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African Journal of Agricultural Research

Table of Contents: Volume 9 Number 40 2 October, 2014

ARTICLES

African Journal of Agricultural Research

Table of Contents: Volume 9 Number 40 2 October, 2014

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Full Length Research Paper

Evaluation of post emergence herbicides on summer groundnut (*Arachis hypogaea* L.) in new alluvial zone of West Bengal

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A field experiment was conducted during summer season of 2010 and 2011 at district seed farm, Kalyani under Bidhan Chandra Krishi Viswavidyalaya, Mohanpur, Nadia, West Bengal, to find out the effect of different post emergence herbicides on summer growing groundnut in new alluvial zone of West Bengal. All the herbicidal treatments as well as weed free check significantly increased the yield and yield attributing characters of groundnut and also reduce the weed population as well as weed biomass production as compared to un-weeded control. Post emergence application of quizalofop Ethyl at 100g a.i./ha was found better to control the weeds as compared to imizathapyr. Among the post-emergence herbicidal treatments pod yield, haulm yield and yield attributes of groundnut was found maximum with the treatment received post emergence application of quizalofop Ethyl at 100 g a.i. /ha. Among all the herbicidal treatments best results was obtained with pre-emergence application of pendimethalin at 1.0 Kg a.i./ ha along with one hand weeding and recorded highest pod yield (2675 Kg/ha), haulm yield (3123 Kg/ha), number of pods/plant (17.3), shelling % (71.3) and SMK % (91.4). This might be due to application of pendimethalin as a pre emergence, suppresses the weed growth at early stage of the crop as a result of better crop stand. Practices of weed control by chemical and /or mechanical gave the significantly better pod yield of groundnut as compared to un-weeded control and pod yield of groundnut was decreased even up to 26 to 47% in un-weeded condition. Among the herbicidal treatments maximum gross return (Rs.80550), net return (Rs.55937) and B:C ratio (3.27) were recorded in the treatment received pre-emergence application of pendimethalin at 1.0 Kg a.i./ ha along with one hand weeding.

Key words: Groundnut, herbicide, pre-emergence, post-emergence, yield, economics.

INTRODUCTION

Groundnut is an important food legume and an oil seed crop in the world which is presently grown in about 90 countries over an area of 25 million hectares under different agro climatic regions. It is grown on large scale in India, China, USA, Senegal, Indonesia, Nigeria, Brazil and Argentina. India accounts for about 34.5% world groundnut area and about 27.3% of world groundnut production while China is the highest producer of groundnut with 37% of total world groundnut production. In West Bengal the total area under groundnut is

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Table 1. Effect of post emergence herbicide on weed flora of groundnut (Pooled over 2010 and 2011).

Treatment	Weed density (Number / m^2)						Weed dry matter (g / m^2)	
	45 DAS			75 DAS				
	NLW	BLW	Total	NLW	BLW	Total	45DAS	75DAS
T_1	179	23	202	243	78	321	207	319
T_2	13	7	20	19	7	26	11	23.1
T_3	17	8	25	39	21	60	26	63.1
T_4	43	12	55	63	22	85	128	236.3
T_5	44	18	62	84	31	115	115.2	226.1
T_6	34	17	51	53	26	79	89.3	146.5
T_7	48	26	74	81	36	117	104.3	169.3
T_8	41	13	54	58	28	86	78.3	138.7
T_9	38	11	49	59	23	82	74.8	102.8
T_{10}	29	16	45	47	29	76	54.4	95.3
T_{11}	44	15	59	54	36	90	42.3	94.1

NLW, Narrow leaved weed ; BLW, Broad leaved weed ; DAS, Days after saowing.

65826 ha and total production is about 113018 tons with average productivity of 1717 kg/ha. The low yield of groundnut in our state as well as in the country can be attributed to many factors, among them many folds losses caused by weeds is of serious nature. Competition of weeds with the crops was observed to be very high during 50 to 60 days period. This period was found to be critical period for crop weed competition (Mahadkar et al., 1993). Weeds when allowed to compete till harvest depleted 162.8 kg N, 21.7 kg P_2O_5 and 141.8 kg K_2O per ha. Herbicides and hand weeding significantly brought down the nutrient removal by weeds and enhanced the uptake of nutrient by groundnut crop (Yadav et al., 1986). Hence, the present experiment was conducted to find out the effect of post emergence herbicide application in groundnut in order to control weed infestation (Table 1).

MATERIALS AND METHODS

Present investigation was carried out during summer seasons of 2010 and 2011 at district seed farm, kalyani under Bidhan Chandra Krishi Viswavidyalaya, Mohanpur, Nadia, West Bengal to study the effect of post emergence herbicides on summer groundnut in new alluvial zone of West Bengal. The soil of the experimental field was alluvium sandy loam in texture with content of organic carbon 0.79%, total N 0.07%, available phosphorus 24.65 kg/ha and Potassium 189.59 kg/ha. The pH of the soil was 6.81. The experiment was laid out in randomized block design with eleven (11) treatments and three (3) replications. The treatment details are as follows: **T_1**= Un-weeded control; **T_2** = Weed free check; **T_3** = Pre-emergence application of pendimethalin at 1.0 Kg a.i. /ha + one hand weeding; **T_4** = Post emergence application of Quizalofop Ethyl at 50 g a.i./ ha 20 DAS ; **T_5**= Post emergence application of Quizalofop Ethyl at 75 g a.i./ ha 20 DAS ; **T_6**= Post emergence application of Quizalofop Ethyl at 100 g a.i./ ha 20 DAS; **T_7** = Post emergence application of Imizethapyr at 50 g a.i./ ha 20 DAS ; **T_8** = Post emergence application of Imizethapyr at 75 g a.i./ ha 20 DAS ; **T_9** = Post emergence application of Imizethapyr at 100 g a.i./ ha 20 DAS ; **T_{10}** = Pre-emergence application of pendimethalin at 1.0 Kg a.i. /ha + T_4 ; **T_{11}**= Pre-emergence application of pendimethalin at 1.0 Kg a.i. /ha +T_8 ; **T_{12}** = farmers practice (one hand weeding + one intercultural operation). The recommended dose of N: P_2O_5 and K_2O was 20:60:40 kg /ha and gypsum at of 400 kg /ha was applied uniformly in all the treatments. The variety TAG 24 was sown in 27th and 30th January during 2010 and 2011 respectively.

RESULTS AND DISCUSSION

Herbicide application to groundnut had the significant influence to increas the groundnut yield. Results indicated that all the herbicidal treatments as well as weed free check resulted in significant increase in yield and yield attributing characters of groundnut along with reduction in the weed population and weed biomass production, compared to un-weeded control.

Effect on weed population and weed dry matter

Best herbicidal response to weed density and weed dry matter production was recorded from T_3 that is, pre-emergence application of pendimethalin at 1.0 Kg a.i. /ha along with one hand weeding. Among the post-emergence herbicidal treatments, best response was observed from application of Imizathapyrat 100 g a.i./ha at 20 DAS (T_9) and application of quizalofop Ethyl at 100g a.i./ha 20DAS (T_6) at 45 DAS and at 75 DAS ,respectively regarding total weed density and weed dry matter production. Best results of weed density and weed dry matter production was found when pendimethalin as a pre-emergence along with quizalofop Ethyl as a post emergence was applied at 20 DAS. Murthy (1982) and Yadav et al. (1983) observed that pendimethalin was most effective in checking most of the monocot as well as dicot weeds and thus by improved the yield components like number of pods per plant, shelling percentage and test weight. Application of pendimethalin at1.5 kg a.i per

Table 2. Effect of post emergence herbicide on yield and yield attributes of groundnut (Pooled over 2010 and 2011).

Treatment	Pod yield (kg/ha)	Haulm yield (Kg/ha)	No.of pods/ plant	Shelling (%)	100 Kernel weight (g)	SMK (%)	Gross return (Rs./ha)	Net return (Rs./ha)	Total cost (Rs./ha)	BCR
T_1	1512	1813	13.2	66.9	37.4	84.0	45360	22742	22618	2.01
T_2	2871	3305	18.6	70.9	38.0	92.6	86130	60505	25625	3.36
T_3	2685	3129	17.3	71.3	37.6	91.4	80550	55937	24613	3.27
T_4	2252	2713	15.4	69.8	37.9	88.9	67560	44440	23120	2. 92
T_5	2383	2911	14.4	70.0	38.0	90.0	71490	48572	22918	3.12
T_6	2409	2941	15.2	69.6	38.2	88.6	72270	49550	22720	3.18
T_7	2082	2546	14.7	69.8	37.3	87.9	62460	39340	23120	2.70
T_8	2109	2598	15.2	70.1	38.1	89.7	63270	40352	22918	2.76
T_9	2211	2731	15.0	69.3	37.3	88.7	66330	43610	22720	2.92
T_{10}	2391	2926	15.8	70.2	38.0	90.6	71730	48414	23316	3.08
T_{11}	2304	2833	16.0	69.9	37.5	89.0	69120	46010	23110	2.99
T_{12}	2598	3181	15.8	70.8	38.6	91.0	77940	52482	25458	3.06
SEm (±)	94.574	71.943	0.709	0.191	0.257	0.546				
CD at 5%	276.858	210.607	2.075	0.559	NS	1.598				

NS, Non significant.

ha showed equal performance in pod yield (16.51 q/ha) with hand weeding at 15 and 35 days after sowing (18.4 q/ha) and was found better in controlling grassy weeds in groundnut (Kondap et al., 1989).

Yield and yield attributes

Effect of different herbicidal treatments on yield and yield attributing characters of groundnut was found to be significant, except in case of 100 kernel weight. Among all the herbicidal treatments, T_3 that is, pre-emergence application of pendimethalin at 1.0 Kg a.i./ ha along with one hand weeding, recorded highest pod yield (2675 Kg/ha), haulm yield (3123 Kg/ha), number of pods/plant (17.3), shelling % (71.3) and SMK % (91.4) as compared to post emergence herbicidal treatments. This might be due to application of pendimethalin as a pre emergence suppresses the weed growth at early stage of the crop as a result of better crop stand. Among the post-emergence herbicidal treatments pod yield, haulm yield and yield attributes of groundnut was found maximum with the treatment received post emergence application of quizalofop Ethyl at 100 g a.i. /ha i,e $T_{6,}$ which was significantly superior to T_7 and T_8 (treatments received Imizathapyr at 50 and 75g a.i./ ha at 20 DAS respectively) but statistically at par with T_9 (Imizathapyr at 100g a.i./ ha at 20 DAS). Among the combinations of pre and post emergence herbicide application, the treatment T_{10} (Pendimethalin at 1.0 kg a.i. / ha and Quizalofop Ethyl at 50 g a.i./ha) and T_{11} (Pendimethalin at 1.0 kg a.i./ha and Imizathapyr 75 g a.i./ha) was statistically at par among each other. From the Table 2 it was found that weed free check (T_2) gave the highest pod yield (2871 kg/ha) as compared to rest of the treatments except T_3 (treatment received pre-emergence application of Pendimethalin at1.0kg a.i./ha and one hand weeding) and T12(one hand weeding and one intercultural operation), those were statistically at par with T_2 .Practices of weed control by chemical and/or mechanical(T_2 to T_{12}) gave the significantly better pod yield of groundnut as compared to un-weeded control (T_1) and pod yield of groundnut was decreased even up to 26 to 47% in un-weeded condition . Groundnut being a deep rooted legume crop proliferation of the root at early stage is essentially required to establish the sufficient numbers of nodule and better crop growth for pegging. Weed growth is faster than crop growth at early stage so controlling of weeds at early stage reduced the crop weed competition and thus providing nutritional security to the crop as result of better pod yield. Maximum gross return (Rs.86130) , net return (Rs.60505) and B:C ratio (3.36) were recorded in the treatment T_2 that

is, weed free check and among herbicidal treatments maximum gross return (Rs.80550), net return (Rs.55937) and B:C ratio(3.27) were recorded in T_3 that is, pre-emergence application of pendimethalin at 1.0 Kg a.i./ ha along with one hand weeding. So it can be revealed that application of quizalofop ethyl at 50 g a.i. / ha can be more effective than imazethapyr at 100 g a.i. / ha in groundnut as post emergence herbicide to control different narrow leaved and broad leaved weeds.

Conflict of Interest

The author(s) have not declared any conflict of interest.

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Full Length Research Paper

Testing market integration and convergence to the law of one price in Indian onions

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Spiraling prices of onions in India undermine the sustainability of current economic growth process and raised the question of price integration among the spatially separated markets. Co-integration test has been used to identify whether onion markets in India share a common linear deterministic trend and the law of one price holds true in view of rising prices in the recent past. The study sourced the wholesale daily prices of major onion markets across the country from January 2010 to March 2011. Augmented Dickey Fuller test statistic has been used to check the presence of a unit root in the time series data. Empirical results indicated the presence of unit root and a strong spatial integration between major markets. The study also confirms the law of one price in Indian onions.

Key words: Co-integration, onion, law of one price.

INTRODUCTION

Onion (*Allium cepa*) is being used as a vegetable and spice for thousands of years by many cultures around the world, and it has immense medicinal and therapeutic value (Sendhil, 2012). It is cultivated across different states in India for domestic consumption and international trade. Among them, Maharashtra accounts for about 40% of India's onion output. Rajasthan is the next major producer followed by Karnataka, Andhra Pradesh and Tamil Nadu. Onion is a commercial crop in India, but the crises of 1998 and 2010 played spoilsport in the economy and the welfare of the producers (Sendhil, 2012). Price surge during the crises led to many economic as well as political implications. The prices have hit the roof across the entire nation and the absence of timely intervention to control them resulted in an explosive situation of prices (Nayyar, 2011). Prices rose by 600% from INR 12 to 18 per kg to INR 60 to 100 per kg during December, 2010 across different markets in the country. Even in the recent past, the wholesale prices hit a two-and-a-half-year high bringing tears in the eyes of millions (Reuters, 2013). Following a 50% increase in the monthly wholesale prices, retail prices increase by cent percent particularly in Indian metros with a wide range of prices across major cities. In comparison to the last year, the

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reported price rise was about 500% in Lasalgoan in Maharashtra, Asia's biggest market. Though supply shock arising from excessive rainfall or drought that spoils the bulbs was a major reason for the spiraling prices, it undermined the sustainability and efficiency of public management in controlling the price of the commodity. The difference in prices that prevailed across onion markets in India raised the question of price integration among the spatially separated markets.

Spatial market integration is a situation in which prices of a commodity in spatially separated markets move together due to arbitrage and the price signals and information are transmitted smoothly across the markets. With the free flow of information in a competitive market, the difference in prices of a product in the two markets would be equal to or less than the transportation cost between them. Empirical studies on market integration of agricultural products typically use bilateral price relationships as an indicator of market integration. This methodology falls under the law of one price (LOOP), which states that if the commodity prices are converted to a common currency, then the particular commodity should be sold at the same price in each country or region.

However, in practice is a very rare case. In case of onions too, there is a great likelihood of market integration and price transmission between markets in India, if the markets are efficient and competitive. In this case, a causal relationship between prices in different spatial markets can be measured (Moodley et al., 2000). Market integration also means that a measurable long-run relationship exists between spatially separated prices for the same product. Thus, even when prices temporarily deviate from each other in the short-run, the differentials should eventually converge in the long-run and the speed of price convergence indicates the degree of market integration. In this context, an attempt has been made with the objective finding the extent of market integration within India and whether the LOOP holds true in the case of onions across different markets in India.

MATERIALS AND METHODS

The study is based on time series data on wholesale daily prices of onion collected from the AGMARKNET portal from January 2010 to March 2011. The data set can be used to analyze the price instability which occurred during 2010 to 2011.

Instability in prices

Instability index was used to examine the extent of variation and risk involved in prices. It was measured by Cuddy-Della Valle Index (Cuddy and Della Valle, 1978; Anuja et al., 2013) which is given as,

Cuddy-Della Valle Instability (%) $= CV \times \sqrt{(1-\overline{R}^2)}$. Where, CV is the coefficient of variation in per cent, and $\overline{R}^2$ is the coefficient of determination from a time trend regression adjusted to its degrees of freedom.

Market integration and price transmission

Estimation of bivariate correlation coefficients between price changes in different markets has been employed as the most common methodology for testing market integration (Cummings, 1967; Lele, 1967, 1971). But it indicates the integration of markets only in the short-run. Economic theory often suggests that certain pairs of economic variables should be linked by a long-run equilibrium relationship. Despite price series move away from the equilibrium point for a while, economic forces may be expected to act so as to restore equilibrium. Granger (1981) proposed the concept of co-integration which states that even though several price time series have unit roots, a linear combination of them would not have a unit root. Since the introduction of co-integration techniques by Engle and Granger (1987), Johansen (1988, 1991, 1994, 1995), Johansen and Juselius (1990), and Goodwin and Schroeder (1991) researchers have applied it on non-stationary data. The Engle and Granger method is basically a bi-variate approach that accommodates relationships only between two price series.

As a result, this shortcoming does not lend itself well to analyzing multivariate systems that characterize, for example, markets with many sellers and buyers. In addition, the results are sensitive to which price series is used to normalize the other. Hypothesis testing on the estimated co-integration vector is also not possible under this approach. The Johansen method is preferred over the Engle and Granger approach and has proven to be popular in the recent literature on market integration (Kumar and Sharma, 2003). Before testing for co-integration, the time series has to be checked for its stationary property. The stationarity properties and the exhibition of unit roots in the time series are substantiated by performing the Augmented Dickey-Fuller (ADF) test. This test is conducted on the variables in level (original price series) and first differences. Price series co-integration arises when the variables are integrated of the same order against the unit root test which identifies the variables that are integrated of order one, or I (1). In a co-integrated equation system:

$$\Delta Y_t = \sum_{i=1}^{k-1} \Pi_i \Delta Y_{t-i} + \alpha\beta' Y_{t-k} + \varepsilon_t$$

where Y_t is the price time series, Δ is the first difference operator ($Y_t - Y_{t-1}$) and matrix $\Pi = \alpha\beta'$ is (n x n) with rank r ($0 \le r \le n$), which is the number of linear independent co-integration relations in the vector space of a matrix. The Johansen's method of co-integrated system is a restricted maximum likelihood method with rank restriction on matrix $\Pi = \alpha\beta'$ (Anuja et al., 2013). The rank of Π can be determined by using λ_{trace} or λ_{max} test statistics. Integration between two markets can be checked in a similar fashion through bi-variate Johansen's test. Since the test is very sensitive to price lag, the choice of lag length should be determined well. After testing for co-integration, the residuals show the deviation from equilibrium and this equilibrium error in the long-run tends to zero. Vector error-correction model (VECM) can be used to capture the deviations from the long-run equilibrium (Brosig et al., 2011). The model is represented as:

$$\Delta A_t = \alpha_0 + \alpha_1 \Delta B_t + \alpha_2 u_{t-1} + \varepsilon_t$$

Where, A_t is the price of market 'A', B_t is the price of market 'B' and u_t is the co-integration vector. The coefficient (α_2) of the error-correction term (u_{t-1}) indicates the speed at which the series returns to equilibrium. If it is less than zero, the series converge to long-run equilibrium and if it is positive and zero, the series diverges from equilibrium.

Table 1. Selected onion markets from different states/union territory.

S/No.	State / Union Territory	Selected market	Basis for selection
1.	Karnataka	Bangalore	Production
2.	Tamil Nadu	Chennai	Consumption
3.	Delhi	Delhi	Wholesale market
4.	Rajasthan	Jaipur	Consumption
5.	West Bengal	Kolkata	Consumption
6.	Maharashtra	Lasalgaon	Production (Asia's biggest market)
7.	Maharashtra	Mumbai	Production
8.	Maharashtra	Nasik	Production

Law of one price (LOOP) analysis

LOOP analysis tests the hypothesis on the coefficients of both α and β using likelihood ratio tests as outlined in Johansen and Juselius (1990). To test for the LOOP, restrictions can be placed and tested on the parameters in the β matrix. In the case of a bivariate system where two price series are examined, the rank of $\pi = \alpha\beta'$ is equal to one, and the dimensions of α and β matrices are 2 x 1. LOOP is tested by imposing the restriction $\beta' = (1, -1)'$. Since the matrix β contains long-run parameters in the system of equations, the test can be considered as a valid one for LOOP in the long-run.

RESULTS AND DISCUSSION

Price behavior of onions in major Indian markets

The price behaviour in major onion markets in India, selected on the basis of production and consumption criteria, (Table 1) was studied with respect to the direction of movement in prices. The price behavior in different markets across the country is depicted in Figure 1. The figure shows the symmetric pattern in the movement of prices in all the markets of the country with peak prices during the months of December (2010) and January (2011) confirming a similar pattern identified by Reddy et al. (2012) in metro cities of India. The plausible reason was supply shock due to unexpected rainfall during the months of September – October which affected the production exhaustively in the major onion growing belts of the country.

Chennai being a region of high consumption of onions with little production, the commodity has to be transported from different parts of the country. Hence, the maximum wholesale price of INR7000/quintal (Table 2) prevailed there which is due to the transportation cost. As expected, the minimum price prevailed in Lasalgaon (INR361/quintal), the Asia's biggest onion market. The average price during the study period was high in Chennai and low in the case of Nasik. As expected, standard deviation and variance was higher in the case of Chennai market. All the markets exhibited a positively skewed distribution. Excluding Chennai and Jaipur, the rest of the markets showed a leptokurtic (slim or long tailed) pattern of probability distribution.

Onion markets integration and price transmission

Market integration is the co-movements or long-run relationship between the spatial prices. The selected markets are tested for co-integration analysis using Johansen's approach. Before co-integration, correlation between different onion markets was carried out to know the short-run integration (Table 3). Correlation analysis revealed a positive co-movement between the onion price series, *apriori*. The results indicated a high degree of significant positive correlation between all the major onion markets that are spatially separated. Before testing for co-integration relationship between different onion market prices, it is mandatory to check the order of integration of the level variables. Hence, for each non-stationary variables, unit root test at their levels as well as first differences were conducted for each market after converting the original series to natural logarithms (Table 4). The results indicated the presence of a unit root at their levels that is, non-stationarity of each market price time series. However, all the non-stationary variables are found to be stationary at their first differences, and therefore, are integrated of order one, I(1) corroborating with the findings of Sidhu et al. (2010) that employed the Augmented Dickey-Fuller (ADF) statistic while studying the integration of wholesale market prices for onions within Punjab state of India. This conformation that each level series is I(1) allows to proceed with the Johansen's co-integration test (Table 5).

The co-integration test revealed the Eigen value and the trace statistic for each market. The test rejected the null hypothesis of no co-integration relationship between the onion market ($r = 0$ to $r \leq 4$) at 5% level of probability indicating the presence of five co-integration vectors between those markets in the long-run (Table 5). The purpose of this analysis was to know whether the onion markets in India are integrated, and thereby price transmission takes place. These findings are also supported by Sidhu et al. (2010) despite their use of Augmented Engle and Granger co-integration test.

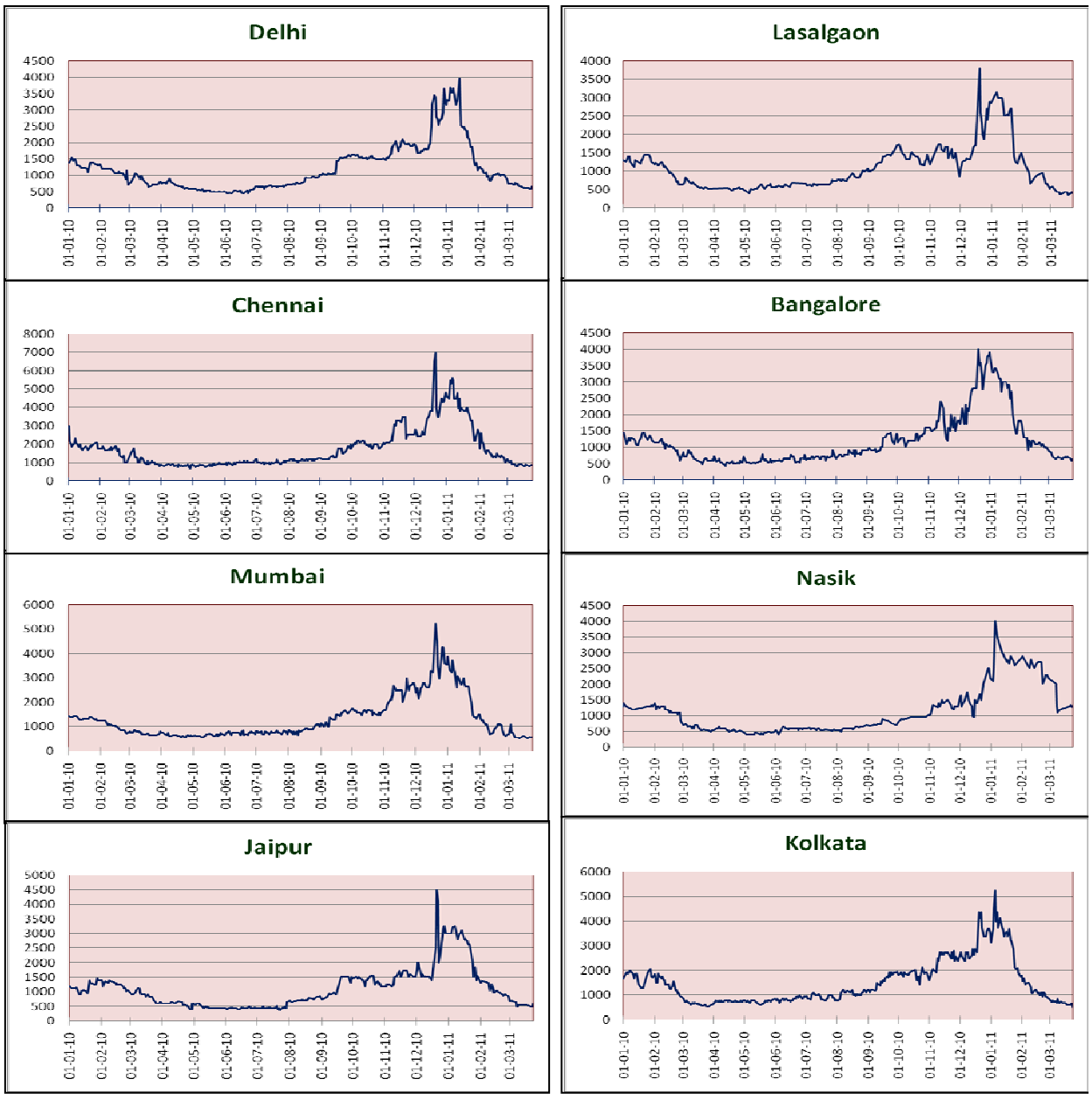

Figure 1. Price behaviour (INR/quintal) of onion in different markets of India.

Yogisha et al. (2006) employed a distributed lag model and found that onion markets in Kolar district of Karnataka were highly integrated attributing to the timely available information on prices. Reddy et al. (2012) also confirmed a high degree of price integration for a majority of the onion markets and they indicated that *inter alia,* prices are governed not only based on market arrivals but also several factors prevailing in other markets like varieties, appearance, moisture content, colour, size and shape of the produce. However, the flow of market information across markets will help to realize the law of one price in onions and the speed of flow can be estimated through the VECM. For this, bi-variate (2 markets) co-integration analysis has been done and the integrated markets were tested with error-correction mechanism. The results of the VECM indicated that most of the estimated coefficients were negative for market 1 and positive for market 2 (Table 6). This indicated that those series with positive coefficients diverge from equilibrium and negative coefficients converge to equilibrium in the short-run. However, further changes in the subsequent periods help the price series to achieve equilibrium in the long-run. The vector error-correction coefficient was estimated at -0.1467 for market 1

Table 2. Summary statistics of onion market prices.

Particulars	Bangalore	Chennai	Delhi	Jaipur	Kolkata	Lasalgaon	Mumbai	Nasik
Observations	448	448	448	448	448	448	448	448
Maximum (INR/quintal)	4000	7000	3975	4500	5250	3800	5250	4000
Minimum (INR/quintal)	425	700	450	370	500	361	525	375
Range (INR/quintal)	3575	6300	3525	4130	4750	3439	4725	3625
Mean (INR/quintal)	1185.30	1724.93	1203.97	1092.57	1501.42	1064.95	1318.87	1044.60
Standard deviation	758.16	1059.75	709.58	681.41	893.19	610.24	852.99	613.20
Variance	574809	1123063	503504	464316	797790	372389	727595	376018
Skewness	1.74	1.78	1.60	1.71	1.33	1.58	1.60	1.59
Kurtosis	2.54	3.36	2.58	3.53	1.39	2.69	2.30	2.70
Instability (%)	56.24	55.97	53.47	57.47	53.90	53.57	58.24	54.50

Table 3. Price correlation between major onion markets in India.

Market (n = 448)	Bangalore	Chennai	Delhi	Jaipur	Kolkata	Lasalgaon	Mumbai	Nasik
Bangalore	1	0.95*	0.95*	0.92*	0.94*	0.92*	0.95*	0.94*
Chennai	0.95*	1	0.95*	0.92*	0.95*	0.93*	0.94*	0.93*
Delhi	0.95*	0.95*	1	0.93*	0.95*	0.94*	0.94*	0.93*
Jaipur	0.92*	0.92*	0.93*	1	0.92*	0.92*	0.89*	0.92*
Kolkata	0.94*	0.95*	0.95*	0.92*	1	0.94*	0.95*	0.94*
Lasalgaon	0.92*	0.93*	0.94*	0.92*	0.94*	1	0.89*	0.93*
Mumbai	0.95*	0.94*	0.94*	0.89*	0.95*	0.89*	1	0.93*
Nasik	0.94*	0.93*	0.93*	0.92*	0.94*	0.93*	0.93*	1

* indicates the significance of Spearman's correlation coefficient at one per cent level of probability.

Table 4. Estimates of ADF test for unit root and lag length based on Akaike Information Criterion (AIC).

Market	Level series		1st differenced series	
	ADF statistic	AIC lag length	ADF statistic	AIC lag length
Bangalore	-1.64	13	-3.15*	17
Chennai	-1.39	5	-12.95*	4
Delhi	-2.14	15	-4.20*	14
Jaipur	-1.44	4	-16.47*	3
Kolkata	-2.08	16	-3.78*	16
Lasalgaon	-2.67	17	-9.05*	7
Mumbai	-1.83	15	-4.05*	14
Nasik	-2.62	17	-3.84*	17

* indicate significance at one per cent level of MacKinnon (1996) one-sided p-values.

(Bangalore) and 0.1761 for market 2 (Chennai). This indicated the speed at which Chennai and Bangalore prices adjust towards the equilibrium in case of any price shocks in other markets. In other words, the coefficient measures the ability of the prices to incorporate shocks or price news available in the market. In this case, Chennai and Bangalore markets absorb 17.61 and 14.67% respectively to bring about the equilibrium in prices. The information flow is more in Chennai market as evident by the magnitude of the coefficient (0.1761). Hence Chennai is more efficient than the Bangalore market in terms of reaction to news on price. Similar kind of interpretation can be done for rest of the markets.

Law of one price (LOOP)

Trace tests showed one co-integrating vector for all the selected markets, the number of common stochastic trends turned out to be one for these markets (Table 7).

Table 5. Estimates of Johansen's multivariate co-integration test.

Data period	: 6/01/2010 to 24/03/2011
Included observations	: 443 after adjustments
Trend assumption	: Linear deterministic trend
Lag length	: 1 to 4
Markets: Lasalgaon, Mumbai, Delhi, Chennai, Jaipur, Kolkata, Nasik and Bangalore	

Null Hypothesis	Eigen Value	Trace Statistic	Critical Value at 5 %	Significance**
r = 0*	0.1418	250.5196	159.5297	0.0000
r ≤ 1*	0.1248	182.7667	125.6154	0.0000
r ≤ 2*	0.0918	123.7211	95.7537	0.0002
r ≤ 3*	0.0708	81.0821	69.8189	0.0048
r ≤ 4*	0.0611	48.5636	47.8561	0.0428
r ≤ 5	0.0230	20.6124	29.7971	0.3822
r ≤ 6	0.0178	10.3184	15.4947	0.2570
r ≤ 7	0.0053	2.3651	3.8415	0.1241

Trace test indicates five co-integrating equation(s) at the 0.05 level, * denotes rejection of the hypothesis at the 0.05 level and ** shows the MacKinnon-Haug-Michelis (1999) probability values.

Table 6. Estimates of Johansen's bivariate co-integration analysis and error correction model.

Markets	Null hypothesis H_0: rank= r	Eigen Value	Trace Statistic	Error-correction estimates		Log Likelihood
				Market 1	Market 2	
Bangalore and Chennai	r = 0*	0.1284	62.81	-0.1467	0.1761	846.75
	r ≤ 1	0.0034	1.54	(0.0322)	(0.0308)	
Bangalore and Delhi	r = 0*	0.0671	31.86	-0.1058	0.0547	1023.70
	r ≤ 1	0.0019	0.87	(0.0260)	(0.0162)	
Bangalore and Jaipur	r = 0*	0.0645	31.72	-0.0237	0.0935	845.51
	r ≤ 1	0.0045	1.99	(0.0204)	(0.0186)	
Bangalore and Kolkata	r = 0*	0.0744	35.00	-0.1286	0.0589	903.85
	r ≤ 1	0.0012	0.53	(0.0262)	(0.0217)	
Bangalore and Lasalgaon	r = 0*	0.0554	26.84	-0.0748	0.0601	869.87
	r ≤ 1	0.0032	1.44	(0.0222)	(0.0198)	
Bangalore and Mumbai	r = 0*	0.0884	42.58	-0.1623	0.0403	889.69
	r ≤ 1	0.0030	1.32	(0.0271)	(0.0241)	
Bangalore and Nasik	r = 0*	0.0816	39.52	-0.1059	0.0878	905.98
	r ≤ 1	0.0035	1.56	(0.0260)	(0.0211)	
Chennai and Delhi	r = 0*	0.0628	29.87	-0.0372	0.1131	1047.68
	r ≤ 1	0.0021	0.95	(0.0170)	(0.0260)	
Chennai and Jaipur	r = 0*	0.0643	31.67	-0.0264	0.1042	869.46
	r ≤ 1	0.0045	2.02	(0.0223)	(0.0207)	
Chennai and Kolkata	r = 0*	0.0873	41.29	-0.1577	0.0589	926.13
	r ≤ 1	0.0013	0.57	(0.0282)	(0.0245)	

Table 6. Contd.

Chennai and Lasalgaon	r = 0* r ≤ 1	0.0668 0.0034	32.37 1.51	-0.1050 (0.0245)	0.0568 (0.0231)	895.30
Chennai and Mumbai	r = 0* r ≤ 1	0.1163 0.0032	56.58 1.44	-0.2010 (0.0279)	0.0263 (0.0262)	913.85
Chennai and Nasik	r = 0* r ≤ 1	0.1040 0.0033	50.48 1.49	-0.1502 (0.0266)	0.0810 (0.0229)	929.34
Delhi and Jaipur	r = 0* r ≤ 1	0.1295 0.0029	63.16 1.31	0.019416 (0.0185)	0.2086 (0.0258)	1071.86
Delhi and Kolkata	r = 0* r ≤ 1	0.0376 0.0010	17.53 0.44	-0.0417 (0.0152)	0.0573 (0.0200)	1110.60
Delhi and Lasalgaon	r = 0* r ≤ 1	0.0564 0.0019	26.73 0.84	-0.0517 (0.0142)	0.0589 (0.0204)	1079.69
Delhi and Mumbai	r = 0* r ≤ 1	0.0730 0.0022	34.78 0.99	-0.0856 (0.0163)	0.0422 (0.0232)	1096.76
Delhi and Nasik	r = 0* r ≤ 1	0.0692 0.0024	33.07 1.08	-0.0676 (0.0156)	0.0653 (0.0208)	1113.10
Jaipur and Kolkata	r = 0* r ≤ 1	0.0459 0.0021	21.91 0.95	-0.0764 (0.0167)	-0.0051 (0.0149)	933.67
Jaipur and Lasalgaon	r = 0* r ≤ 1	0.0466 0.0044	23.24 1.98	-0.0745 (0.0163)	0.0007 (0.0166)	917.81
Jaipur and Mumbai	r = 0* r ≤ 1	0.0573 0.0053	28.72 2.39	-0.0838 (0.0162)	-0.0130 (0.0153)	915.30
Jaipur and Nasik	r = 0* r ≤ 1	0.0531 0.0059	26.98 2.65	-0.0808 (0.0174)	0.0126 (0.0155)	935.57
Kolkata and Lasalgaon	r = 0* r ≤ 1	0.0650 0.0014	30.62 0.64	-0.0680 (0.0207)	0.0752 (0.0228)	977.34
Kolkata and Mumbai	r = 0* r ≤ 1	0.0907 0.0019	43.26 0.83	-0.1354 (0.0233)	0.0699 (0.0250)	985.05
Kolkata and Nasik	r = 0* r ≤ 1	0.0775 0.0018	36.79 0.81	-0.0867 (0.0227)	0.0934 (0.0229)	998.14
Lasalgaon and Mumbai	r = 0* r ≤ 1	0.0483 0.0035	23.61 1.55	-0.0882 (0.0232)	0.0291 (0.0218)	971.87
Lasalgaon and Nasik	r = 0* r ≤ 1	0.1057 0.0039	51.56 1.74	-0.1459 (0.0310)	0.1225 (0.0273)	989.30

Table 6. Contd.

Mumbai and Nasik	r = 0*	0.0683	32.92	-0.0510	0.0976	979.78
	r ≤ 1	0.0030	1.36	(0.0227)	(0.0212)	

* denotes rejection of the null hypothesis at 5 % level of MacKinnon-Haug-Michelis (1999) probability values. The critical value for rejecting the null hypothesis at 5% , H_0: r=0 is 15.49 and r ≤ 1is 3.81. Figures in parenthesis indicate the standard errors.

Table 7. LOOP analysis for onion markets.

Markets	Null hypothesis H_0: rank= r	Number of co-integrated vectors	Number of stochastic trends	LOOP
Bangalore and Chennai	r = 0*	1	1	Yes
	r ≤ 1	1	1	
Bangalore and Delhi	r = 0*	1	1	Yes
	r ≤ 1	1	1	
Bangalore and Jaipur	r = 0*	1	1	Yes
	r ≤ 1	1	1	
Bangalore and Kolkata	r = 0*	1	1	Yes
	r ≤ 1	1	1	
Bangalore and Lasalgaon	r = 0*	1	1	Yes
	r ≤ 1	1	1	
Bangalore and Mumbai	r = 0*	1	1	Yes
	r ≤ 1	1	1	
Bangalore and Nasik	r = 0*	1	1	Yes
	r ≤ 1	1	1	
Chennai and Delhi	r = 0*	1	1	Yes
	r ≤ 1	1	1	
Chennai and Jaipur	r = 0*	1	1	Yes
	r ≤ 1	1	1	
Chennai and Kolkata	r = 0*	1	1	Yes
	r ≤ 1	1	1	
Chennai and Lasalgaon	r = 0*	1	1	Yes
	r ≤ 1	1	1	
Chennai and Mumbai	r = 0*	1	1	Yes
	r ≤ 1	1	1	
Chennai and Nasik	r = 0*	1	1	Yes
	r ≤ 1	1	1	
Delhi and Jaipur	r = 0*	1	1	Yes
	r ≤ 1	1	1	

Table 7. Contd.

Delhi and Kolkata	r = 0* r ≤ 1	1 1	1 1	Yes
Delhi and Lasalgaon	r = 0* r ≤ 1	1 1	1 1	Yes
Delhi and Mumbai	r = 0* r ≤ 1	1 1	1 1	Yes
Delhi and Nasik	r = 0* r ≤ 1	1 1	1 1	Yes

The number of common stochastic trends was determined by subtracting the number of co-integrating vectors from the dimension of the impact matrix given by the number of variables (*n*) included in the co-integration test. The findings of *n* - 1 co-integrating vectors implied that all the prices contain the same stochastic trend and so they are co-integrated in pairs. This suggested that the LOOP holds true for the onion markets across India.

Conclusions

The study of past price behavior and price co-movement in major onion markets in India conclusively support the existence of an integrated market in the commodity. Several conclusions follow from this result. Price transmission occurs between geographically separated markets in onion due to market information flow through diverse channels.

However, the speed of convergence of onion prices to equilibrium depends on the speed of information dissemination, the government's control over the commodity, time-to-time regulations and policy harmonisation within the regions of the country. One of the reasons for market integration is the efficient functioning of markets itself which is clearly evident from the realisation of law of one price. Therefore, commodity based analysis on marketing integration and market research are better suited to understand the price behaviour of different agricultural commodities. This kind of studies is equally important as they provide policy makers with better information on expected market behaviour which will enable the decision making process on resource allocation. More resources should be allocated to those markets having a higher degree of integration and market efficiency. This will help in enhancing the overall efficiency of the marketing function in agricultural commodities and reduce market distortions.

Conflict of Interest

The authors have not declared any conflict of interest.

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Full Length Research Paper

Effect of integrated nutrient management on yield and quality of acid lime (*Citrus aurantifolia* Swingle)

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Integrated nutrient management refers to maintenance of the soil fertility and plant nutrient supply to an optimum level for sustaining the desired crop productivity through optimization of the benefits from all possible sources of plant nutrient in an integrated manner. Therefore, it is a holistic approach, where we first know what exactly is required by the plant for an optimum level of production, in what different forms these nutrients should be applied in soil and at what different timings in the best possible method and how best these form should be integrated to obtain highest productive efficiency on the economically acceptable limits in an environment friendly manner. To identify suitable integration of different sources of nutrients in different ratios with respect to vegetative growth, fruit yield and quality, a field investigation was carried out on integrated nutrient management in acid lime (*Citrus aurantifolia* Swingle) in randomized block design with three replications. There were nine treatments, T_1- Control (500 : 300 : 300 g NPK per tree through fertilizers), T_2- 2/3 RDF + 1/3 through Goat manure, T_3- 2/3 RDF + 1/3 through *Neem* Cake, T_4- 1/3 RDF + 2/3 through Goat manure, T_5- 1/3 RDF + 2/3 through *Neem* Cake, T_6- 50% RDF + 50% through Goat manure, T_7- 50% RDF + 50% through *Neem* Cake, T_8- 100% RDF as Goat manure, T_9- 100% RDF as Neem Cake. The observation on growth parameters of acid lime tree, fruit yield and yield attributes and fruit quality were recorded to study the effect of treatments and their interpretation. Results of the investigation revealed that treatment T_6 (50% RDF + 50% through goat manure) performed best among all treatments. The maximum vegetative growth and yield (7.58 kg tree^{-1}) of fruits having highest fruit length (4.43 cm), fruit diameter (3.99 cm) and fruit weight (35.71 g) was recorded under T_6 treatment. Similarly best quality fruits were also produced with maximum juice (43.37%), TSS (10.42%) and Ascorbic acid (86.33 mg/100 g juice) content and minimum seed (1.15%) and acidity (6.06%) content under the same treatment.

Key words: Integrated nutrient management, acid lime, vegetative growth, fruit yield, fruit quality.

INTRODUCTION

Citrus fruits are popular in subtropical regions of north India mainly due to their hardy nature and good nutritional values. India produced 10,09,000 (Ten lakh nine thousand) tonnes of citrus fruit annually from 1,04,200 (One lakh four thousand two hundred) ha area with the productivity of 9.70 tones ha^{-1} (Tiwari et al., 2013). Rajasthan produces about 1.80 million tones of horticultural produce from an area of 0.95 million hectare

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and accounts for about 1% of total horticulture production of the country. The major of horticulture produce comes from vegetable (40.82%), spices (28.28%) and fruits (26.85%). It is the eighth largest producer state of citrus in the country and forms 3.6% of total production of citrus in the country. The State produces 0.31 million tones of citrus from an area of 0.02 million hectare with productivity of 18.1 tones ha^{-1} (Anonymous, 2011).

Acid lime (*Citrus aurantiifolia* Swingle*)* is a shrubby tree, to 5 m (16 ft), with many thorns. Its trunk rarely grows straight, with many branches, often originating quite far down on the trunk. The leaves are ovate, 2.5 to 9 cm long, resembling orange leaves (the scientific name *aurantiifolia* refers to this resemblance to the leaves of the orange, *C. aurantium*). The flowers are 2.5 cm in diameter, are yellowish white with a light purple tinge on the margins. Flowers and fruit appear throughout the year, but are most abundant from May to September in the Northern Hemisphere. The method of cultivation greatly affects the size and quality of the harvest. Trees cultivated from seedlings take 4 to 8 years before producing a harvest. They attain their maximal yield at about 10 years of age.

Citrus occupies an important place in the fruit industry, but yield levels of citrus orchards are still very low. Out of many factors, poor nutrient status of the soil as well as malnutrition is considered to be the major factors responsible for citrus decline and low yield. Chemical fertilizers are mostly in use for their cultivation, which have some deleterious effects on fruit quality besides adverse effect on soil, water and environmental pollution. An integrated use of organic manures, biofertilizers and chemical fertilizers could help in achieving the goal of obtaining safer food and environment for the people.

Nutrient refers to all those compounds, which are required by the plant as a source of body building material and for the energy, without which, it will not be able to complete its life cycle. The fruit tree nutrition is concerned with the provision of plant with nutrients as well as nutrient uptake and their distribution in the plant nutrition is fertilizer and/ or nutrient application. Fertilizer is one of the major inputs accounting for nearly one-third of the cost of cultivation and its production consumes a lot of energy used in agriculture. Consequent to the global energy crisis, efficient and judicious use of the fertilizers is imperative not only for obtaining more yields per unit area on a sustainable basis, but also to conserve the energy and to avoid the problem of environment quality. These have become key components for the fruit industry growth. The protection of natural resources base for which the intensification of integrated nutrient management in fruit production has become important, because it is directly related to soil and water resources. The land and water are the bases for any sustainable system of agriculture and the improvement should lay the foundation of a production, economically viable, environment friendly and socially acceptable crop production system.

Therefore, we must adopt the most appropriate land investment practices. The new thinking about the soil management technologies needed for the continuous enhancement of the productivity, sustainability of land, arresting the process of land degradation, accelerating the process of land degradation, accelerating the rate of reclamation and restoration of the productivity of lands which have degraded in the past. Therefore, integrated nutrient management is the most appropriate approach for managing the nutrient input. Integrated nutrient management refers to maintenance of the soil fertility and plant nutrient supply to an optimum level for sustaining the desired crop productivity through optimization of the benefits from all possible sources of plant nutrient in an integrated manner. Therefore, it is a holistic approach, where we first know what exactly is required by the plant for an optimum level of production in what different forms these nutrients should be applied in soil and at what different timings in the best possible method; and how best these forms should be integrated to obtain highest productive efficiency on the economically acceptable limits in an environment friendly manner.

Increased chemical fertilizer cost and awareness of environmental pollution have necessitated the use of organic fertilizers for the development of more efficient fertility management program (Bhattarai and Tomar, 2009). Organic fertilizers are apparently environment and farmer friendly renewable source of non-bulky, low cost organic agricultural inputs for improving soil fertility status. Organic manures are fairly good source of nutrients which has direct influence on plant growth like other commercial fertilizers. Mukherjee et al. (1991) and Prasad and Singhania (1989) also reported that application of organic manures with NKP increased the leaf nutrient status of *Khasi* mandarin, which consequently increased the fruit size, weight and yield. Owing to increasing cost of fertilizers, their short supply and sustainability issues gaining importance, it felt essential to reduce the dependence on chemical fertilizers. Therefore, different sources of plant nutrient, viz., chemical fertilizers, organic manures, Vesicular-arbuscular mycorrhiza (VAM) and biofertilizers have to be tried for working out their suitable integration (Singh et al., 2004).

Keeping the above facts into consideration, an investigation on integrated nutrient management in acid lime (*Citrus aurantifolia* Swingle) was carried out to identify the suitable integration of different sources of nutrients with respect to plant growth, yield and quality of acid lime fruits.

MATERIALS AND METHODS

Investigation locale

The investigation was conducted during 2007 to 2008 and 2008 to

Table 1. Properties of experimental soil before experimentation.

Particulars	Value obtained	Reference to the method employed
Mechanical analysis		
Coarse sand	16.10%	International pipette method (Piper, 1950)
Fine sand	25.43%	
Silt	35.78%	
Clay	22.69%	
Chemical analysis		
Available nitrogen (N)	133.10 kgha^{-1}	Subbiah and Asija (1956)
Available phosphorus (P)	18.37 kgha^{-1}	Olsen et al. (1954)
Available potassium (K)	290.63 kgha^{-1}	Metson (1956)
Soil pH	8.2	Richard (1954)
Bulk density	1.39 gm^{-3}	Singh (1980)

2009 at Regional Research Station (Central Arid Zone Research Institute), Pali, Rajasthan, India. The study area represents the transitional climatic conditions between the sub-tropical arid and semi-arid regions and falls in upper Luni basin agro-climatic zone of the arid Rajasthan. It is situated at 25° 47' 13" north altitudes and 73° 18' 42" east longitudes, at an elevation of 220.46 m above the mean sea level. This location received annual average rainfall of 465 mm. Of the total precipitation, 90% was received during July to September.

Experimental details

Six years old acid lime trees having uniform size and vigour were selected. The trees were planted at a spacing of 6 m × 6 m. The water was applied to the trees through ring system of surface irrigation method. About 150 L water tree^{-1} was given per irrigation at an interval of 20 days. An experiment was laid out in a randomized block design with three replications. There were nine treatments, T_1- Control (500 : 300 : 300 g NPK per tree through fertilizers), T_2- 2/3 RDF (Recommended Dose of Fertilizers) + 1/3 through Goat manure, T_3- 2/3 RDF + 1/3 through *Neem* Cake, T_4- 1/3 RDF + 2/3 through Goat manure, T_5- 1/3 RDF + 2/3 through *Neem* Cake, T_6- 50% RDF + 50% through Goat manure, T_7- 50% RDF + 50% through *Neem* Cake, T_8- 100% RDF as Goat manure, T_9- 100% RDF as *Neem* Cake.

Method of application of nutrients

The required quantity of manures (goat manure and *Neem* cake) were weighed by weighing balance separately and applied by broadcasting in the tree basin area, then mixed in the soil properly. Among fertilizers, the total quantity of nitrogen was supplied through urea (46%). Phosphorus and potassium were applied through single super phosphate (16%) and potassium chloride (60%), respectively. Soil application of half dose of nitrogen and full dose of phosphorus and potassium were supplied in July and the remaining half dose of nitrogen was given as top dressing at the time of fruiting in the month of November.

Experimental soil

Experimental soil was loamy sand in nature having low nitrogen, medium phosphorus and high potassium content with 8.2 pH (Table 1).

Vegetative growth parameters

Vegetative growth parameter viz., tree height, spread and stem girth was measured with the help of measuring tape each year just after harvesting of fruits and expressed the pooled values of both the years in cm (tree height and stem girth) and M^2 (tree spread).

Fruit sampling procedure and recording of data

Fruit samples were taken from the plants under different treatments at the time of maturity and analyzed for various physical characteristics. At the time of harvest, ten fully developed fruits were selected randomly from each tree. Length of these fruits was measured longitudinally, fruit diameter transversely with the help of vernier callipers, mean value per fruit calculated and expressed in cm. Weight of the selected fruits was also recorded with the help of physical balance, mean value per fruit calculated and expressed in g fruit^{-1}. For computing the yield of fruits per plant, the matured fruits were harvested and weighed periodically and yield was expressed in kg tree^{-1}.

Juice and seed content in fruits

Juice of the fruits was extracted with the help of juice extractor, from ten randomly selected fruits of each tree, which were washed, dried and weighed. Then filtered through clean muslin cloth, which separated the juice from fruit sacs and seeds. Filtered juice was measured with the help of measuring cylinder in milliliters and expressed as percentage juice content in the fruits on the basis of fruit weight. Similarly, the weight of seeds was recorded with the help of physical balance in grams and expressed as percentage seed content in fruits on the basis of fruit weight. Juice seed ratio was calculated by dividing the values seed (g) by the values of juice (ml).

Chemical composition of fruits

The mature fruits were selected for the study of their chemical composition in the second picking.

(a) T.S.S.: Ten fruits were randomly selected for juice extraction and total soluble solids of the juice were determined by using a 'Zeiss' hand refractometer of 0 to 30% range. The values were corrected at 20°C and expressed as per cent total soluble solids of

Table 2. Effect of integrated nutrient management on vegetative growth of acid lime trees.

Treatment	Tree height (cm)	Tree spread (m^2)	Stem girth (cm)
T_1- Control (500 : 300 : 300 g NPK per tree through fertilizers)	245.28	7.17	36.92
T_2- 2/3 RDF + 1/3 through Goat manure	275.81	9.07	39.25
T_3- 2/3 RDF + 1/3 through *Neem* Cake	291.56	8.51	38.48
T_4- 1/3 RDF + 2/3 through Goat manure	286.83	9.10	41.01
nrrT_5- 1/3 RDF + 2/3 through Neem Cake	289.22	9.54	40.77
T_6- 50% RDF + 50% through Goat manure	319.28	11.37	45.24
T_7- 50% RDF + 50% through *Neem* Cake	287.06	8.54	39.58
T_8- 100% RDF as Goat manure	287.66	8.29	39.23
T_9- 100% RDF as *Neem* Cake	289.22	8.44	39.77
SEm±	**4.28**	**0.26**	**9.59**
CD at 5%	**12.33**	**0.76**	**3.20**

the fruit juice (A.O.A.C., 1990).

(b) Acidity: The acidity was determined by diluting the known volume of clean juice, filtered through muslin cloth with distilled water and titrated against standard N/10 sodium hydroxide (NaOH) solution, using phenolphthalein as an indicator. The appearance of light pink colour was marked as the end point. The result was expressed in terms of per cent acidity of the fruit juice (A.O.A.C., 1990).

$$\text{Total acid} = \frac{Titre \times Normality\ of\ alkali \times Volume\ made\ up \times Eq\ wt\ of\ acid \times 100}{Volume\ of\ sample\ for\ estimate \times Wt.\ or\ volume\ of\ sample\ taken \times 1000}$$

(c) Ascorbic acid: The ascorbic acid of fruit juice was determined by diluting the known volume of juice with 3% metaphosphoric acid and titrating with 2, 6 dichlorophenol indophenol dye solution. The result was expressed as mg of ascorbic acid per 100 g of fruit juice (A.O.A.C., 1990).

(d) Standardization of dye: Standardization of 2, 6 dichlorophenol indophenols dye solution was done. For this purpose, 100 mg pure ascorbic acid was dissolved in 3% metaphosphoric acid and volume was made up to 100 ml. From this, 1 ml ascorbic acid solution was used for titration.

$$\text{Ascorbic acid (mg/100 g p)} = \frac{Titre \times Dye\ factor \times Volume\ made\ up}{Aliquot \times Wt.\ or\ volume\ of\ sample\ taken} \times 100$$

Statistical analysis

The data for two successive years (2007 to 2008 and 2008 to 2009) were pooled together and subjected to statistical analysis. To test the significance of data, the analysis of variance technique was adopted as suggested by Gomez and Gomez (1984). Significance of the difference in the treatment effect was tested by 'F' test. Critical difference value (*p=0.05*) was calculated whereever the 'F' test was significant.

RESULTS AND DISCUSSION

Application of nutrients to acid lime trees through different sources as integrated nutrient management improved vegetative growth, fruit yield and its attributes, juice content and its chemical composition.

Vegetative growth

Results of the investigation (Table 2) revealed that vegetative growth parameters like height, spread, and stem girth of acid lime trees influenced significantly with the application of recommended doses of nutrients through different combinations of organic and inorganic sources. Maximum tree height (319.28 cm), tree spread (11.37 m^2) and stem girth (45.24 cm) was recorded with the application of T_6 (50% RDF + 50% through goat manure). These values were minimum (245.28 cm, 7.17 m^2 and 36.92 cm, respectively) under control (100% NPK nutrients through fertilizers). The tree height, spread and stem girth were taken as indicators for the growth of acid lime tree. The maximum values of these parameters were recorded under treatment T_6, which was superior over rest of all the treatments. It might be due to high nutrient and mineral content present in the combination of inorganic fertilizers (50%) with organic fertilizer, that is, goat manure (50%) in comparison to other sources and treatment combinations. This might also be attributed to the improved nutrient use efficiency with the balanced use of organic and inorganic sources of nutrients. Application of goat manure with NPK fertilizers improved the soil texture and porosity due to bulkiness in nature, which might have helped the plant root development and enhanced the uptake of available nutrients resulting into faster cell division and cell elongation; and consequently increased the tree height, spread and stem girth. These observations were corroborated with the findings of Villasurda (1990) and Yadav et al. (2012) in guava.

Yield and yield attributes

Data presented in Table 3 revealed that integrated nutrient management practices influenced significantly

Table 3. Effect of integrated nutrient management on yield and yield attributes.

Treatment	Fruit length (cm)	Fruit diameter (cm)	Fruit weight (g)	Yield (kg/tree)
T_1- Control (500 : 300 : 300 g NPK per tree through fertilizers)	3.66	3.16	23.40	5.54
T_2- 2/3 RDF + 1/3 through Goat manure	3.75	3.24	24.74	5.69
T_3- 2/3 RDF + 1/3 through *Neem* Cake	3.77	3.53	28.42	5.62
T_4- 1/3 RDF + 2/3 through Goat manure	4.13	3.76	33.55	5.73
T_5- 1/3 RDF + 2/3 through *Neem* Cake	4.16	3.69	30.66	5.93
T_6- 50% RDF + 50% through Goat manure	4.43	3.99	41.26	7.58
T_7- 50% RDF + 50% through *Neem* Cake	4.17	3.90	35.71	6.89
T_8- 100% RDF as Goat manure	3.86	3.72	30.81	6.85
T_9- 100% RDF as *Neem* Cake	3.77	3.75	29.24	6.64
SEm±	0.04	0.04	0.94	0.18
CD at 5%	0.12	0.11	2.70	0.52

Table 4. Effect of integrated nutrient management on chemical constituents of fruits.

Treatment	**Juice content (%)**	**Seed content (%)**	**Juice: Seed ratio**
T_1- Control (500 : 300 : 300 g NPK per tree through fertilizers)	39.17	1.72	0.044
T_2- 2/3 RDF + 1/3 through Goat manure	38.33	1.19	0.031
T_3- 2/3 RDF + 1/3 through Neem Cake	40.17	1.45	0.028
T_4- 1/3 RDF + 2/3 through Goat manure	41.07	1.28	0.031
T_5- 1/3 RDF + 2/3 through Neem Cake	43.12	1.15	0.027
T_6- 50% RDF + 50% through Goat manure	43.73	1.15	0.026
T_7- 50% RDF + 50% through Neem Cake	40.44	1.20	0.030
T_8- 100% RDF as Goat manure	34.90	1.21	0.035
T_9- 100% RDF as Neem Cake	34.71	1.24	0.036
SEm±	0.37	0.04	0.0012
CD at 5%	1.07	0.11	0.0036

the physical characters of fruits (fruit length, fruit diameter and fruit weight) and fruit yield. Highest fruit yield (7.58 $kgha^{-1}$) with maximum fruit length (4.43 cm), fruit diameter (3.99 cm) and fruit weight (41.26 g) was recorded in the acid lime trees fed by treatment T_6 (50% RDF + 50% through goat manure). However, the lowest yield (5.54 $kgha^{-1}$) with minimum fruit length (3.66 cm), fruit diameter (3.16 cm) and fruit weight 23.40 g) were recorded under T_1 treatment (control) where 100% NPK nutrients was applied through fertilizers.

Increase in yield and yield attributing characters with 50% NPK fertilizer doses in association with 50% nutrients through goat manure was due to the optimum supply of plant nutrients and growth hormones at desired amount during entire period of fruit growth, ultimately resulting in accumulation of more photosynthates leading to more length, diameter, fruit weight and yield of fruits. The increase in both number and weight bases might be attributed to the fact that, there was increasing levels of nutrients in assimilating area of crop due to which the rate of dry matter production was enhanced. Similarly, due to rational partitioning of dry matter to economic sink, the yield attributes were improved. The above results are in conformity with the findings of Dalal et al. (2004). Fruit weight and fruit size are highly correlated with dry matter content and balanced level of hormones. Superior physical fruit quality may be due to the fact that, goat manure combined with fertilizers enhances the nutrient availability by enhancing the capability of plants to better solute uptake from rhizosphere; also these are known for accumulation of dry matter and their translocation as well as favour synthesis of different growth regulators. The findings are in accordance with Gawande et al. (1998) and Patel and Naik (2010) in sapota.

Juice and seed content in fruits

Findings of the investigation (Table 4) exhibited that integrated nutrient management influenced significantly the juice and seed content in acid lime fruits and their juice: seed ratio. Maximum juice content (43.73%) with minimum seed content (1.15%) and minimum juice: seed ratio (0.026) was recorded in the fruits produced with the

Table 5. Effect of integrated nutrient management on chemical constituents of fruits.

Treatment	TSS (%)	Acidity (%)	Ascorbic acid (mg/ 100 g juice)
T_1- Control (500 : 300 : 300 g NPK per tree through fertilizers)	10.12	7.02	75.67
T_2- 2/3 RDF + 1/3 through Goat manure	10.20	6.53	78.00
T_3- 2/3 RDF + 1/3 through Neem Cake	10.23	6.77	79.17
T_4- 1/3 RDF + 2/3 through Goat manure	10.22	6.98	80.33
T_5- 1/3 RDF + 2/3 through Neem Cake	10.17	7.07	79.83
T_6- 50% RDF + 50% through Goat manure	10.42	6.06	86.33
T_7- 50% RDF + 50% through Neem Cake	10.13	7.12	82.50
T_8- 100% RDF as Goat manure	10.13	7.10	78.50
T_9- 100% RDF as Neem Cake	10.20	6.76	77.67
SEm±	0.04	0.06	0.81
CD at 5%	0.10	0.18	2.33
CV%	1.47	3.98	4.30

application of treatment T_6 (50% RDF + 50% through goat manure). However, the values of juice content (43.12%), seed content (1.15%) and juice: seed ratio (0.027) recorded in the fruits produced with treatment T_5 (1/3 RDF + 2/3 through *Neem* Cake) were at par with the values obtained under T_6. The minimum juice content (34.71%) was recorded in the fruits produced with application of treatment T_9 (100% RDF as *Neem* cake), while maximum seed content (1.72%) and juice: seed ratio was recorded in the fruits produced under treatment T_1 (control), that is, chemical fertilization. Improvement in physical characters of fruits with respect to fruit size (fruit length and diameter), fruit weight, juice content, seed weight and juice seed ratio in response to integrated nutrient management including organic and inorganic sources of nutrients can be related to assimilate accumulation of the plant. Similar results have been reported by Verma (2010) in phalsa, Yadav et al. (2007) in aonla and Madhvi et al. (2008) in mango.

Chemical composition of fruits

It is inferred from the data presented in Table 5 that chemical composition of acid lime fruits with respect to TSS, acidity and ascorbic acid content was significantly affected with integrated nutrient management. The highest TSS (10.42%) and ascorbic acid (86.33 mg/100 ml juice) with minimum acidity (6.06%) were recorded in the fruits produced with the application of treatment T_6 (50% RDF + 50% through goat manure). However, the minimum TSS (10.12%) and ascorbic acid (75.67 mg/100 g juice) were recorded in the fruits produced under control (T_1 treatment).

These findings are in accordance with the results of Mahendra et al. (2009) in ber. The quality improvement in fruits may be due to proper supply of nutrients and induction of growth hormones, which stimulated cell division, cell elongation, increase in number and weight of fruits, better root development and better translocation of water uptake and deposition of nutrients. This might be attributed due to the improved fertilizer use efficiency with the application of organic source of nutrients (Ranjan and Gosh, 2006) in sweet orange and Ram et al. (2007) in guava.

Conclusion

On the basis of obtained experimental findings, it can be concluded that among different treatments of integrated nutrient management, application of T_6 (50% RDF + 50% through goat manure) gave best results with respect to vegetative growth, yield, fruiting and quality of fruits of acid lime. Hence, application of treatment T_6 (50% RDF + 50% through goat manure) is highly recommended to enhance growth of trees and consequently produce high yield of good quality fruits.

Conflict of Interest

The authors have not declared any conflict of interest.

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Full Length Research Paper

Geochemical evaluation of high-fluoride and nitrate groundwater: A Case Study from Qinkenpao area, Daqing, China

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Groundwater quality problems have emerged in many geographical areas due to natural environmental processes and human intervention in the geosystems. Hydrogeochemical appraisal of fluoride contaminated groundwater in Qinkenpao area, Daqing State, China is carried out by means of groundwater quality investigations. Results obtained from aqueous speciation modeling using AQUACHEM 5.1 reveal that the groundwater is undersaturated with fluorite and with calcite. The samples also contained high concentrations of nitrate, which is a serious water quality issue due to the impact of human activity. Factor analysis modeling demonstrated that NO_3^- in the system are produced by anthropogenic sources. Strong correlation observed between NO_3^- and Ca^{2+}, Mg^{2+}, Cl^- and F^-, suggesting that they have the same origin. The factor analysis indicates that sodium plus nitrate bicarbonate groundwater have a high loading factor for fluoride, whereas that for calcium chloride and magnesium chloride groundwater is low. The plausible geochemical reactions in the area of study are dissolution of calcite and dolomite, carbon dioxide and sulphate minerals with ion exchange.

Key words: Aqueous speciation modeling, fluoride, nitrate, groundwater, hydrogeochemical, factor analysis.

INTRODUCTION

Water is an essential natural resource for sustaining life and environment that we have always thought to be available in abundance and a free gift of nature. However, chemical composition of surface or subsurface is one of the prime factors in which the suitability of water for domestic, industrial or agricultural purpose depends. Freshwater occurs as surface water and groundwater. Though groundwater contributes only 0.6% of the total water resources on earth, it is the major and preferred source of drinking water in rural as well as urban areas, particularly in developing countries like China because treatment of the drinking water, including disinfection is often not required. But in the area of economical growth, groundwater is getting polluted due to urbanization and

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industrialization.

Presence of various hazardous contaminants like fluoride, nitrate, arsenic, pesticides, other heavy metals etc. in groundwater has been reported from different parts of China. The occurrence of high fluoride and nitrate concentration in groundwater has now become one of the most important health-related geoenvironmental issues in many countries of the world. Fluoride and arsenic are the two main contaminants in groundwater present naturally and artificially in China, India, Egypt, Morocco, Saudi Arabia, Argentina, Canada etc., causing a set of health symptoms known commonly as fluorosis and arsenicosis (Mameri, 1998).

In groundwater, the natural concentration of fluoride depends on the geological, chemical and physical characteristics of the aquifer, the porosity and acidity of the soil and rocks; the temperature, the action of other chemical elements like calcium and bicarbonates, and the depth of the aquifer (Chandra et al., 1981; Largent, 1961).

Fluoride has a significant mitigating effect against dental caries if the concentration is approximately 1 mg/L. However, continuing consumption of higher concentrations can cause dental fluorosis and in extreme cases even skeletal fluorosis. The maximum tolerance limit of fluoride in drinking water specified by the World Health Organization (WHO, 1984) is 1.5 mg/L. High fluoride concentrations are especially critical in developing countries, largely because of lack of suitable infrastructure for treatment.

If fluoride concentration in drinking water is more than 1.0 mg/L, it would result in fluorosis (dental fluorosis and skeletal fluorosis) for human beings, especially for children and pregnant woman. This is a result of the destruction of metabolic calcium and phosphorus, as well as inhibition of active enzymatic processes in the human body. Thus, the function of the endocrine system gets interrupted, leading to fluorosis (Khandare et al., 2005).

Once nitrates get into the groundwater, the greatest danger is for babies less than 1 year old. Small babies have bacteria in their digestive tract that converts nitrate into nitrite, which is toxic. Nitrite reacts with a substance in the blood called hemoglobin. When nitrites are present, hemoglobin will preferentially combine with nitrite instead of oxygen. The new substance formed is called methemoglobin and does not carry oxygen. As the amount of methemoglobin increases, the amount of oxygen in the blood decreases, eventually causing internal suffocation (Walton, 1951; WHO, 1996; USEPA, 1997).

The most common symptom of nitrate poisoning in babies is a bluish color to the skin, particularly around the baby's eyes and mouth. The blood will also turn a chocolate-brown color, which reflects the lack of oxygen.

These symptoms of nitrate toxicity are commonly referred to as the "blue-baby" syndrome.

Except the municipality of Shanghai, every province in China has areas that are afflicted with endemic fluorosis (Zan-dao and Yan, 2002). Unfortunately, the problem is often serious and is usually difficult to prevent or correct. The major sources of fluoride ingestion are from drinking water, coal burning, and tea bricks. For the prevention of dental caries, a drinking water standard for fluoride of 1.0 to 1.5 mg/L had been widely adopted. In China, the standard in the 1950s was 1.5 mg/L. In 1970s, it was lowered to 0.5 to 1.0 mg/L, where it has remained till date (Zan-dao and Yan, 2002).

Recently, factor analysis has been used with remarkable success by many investigators as a tool in the study of groundwater chemistry (Lawrence and Upchurch, 1983). In this study, hydrogeochemical evaluation of high-fluoride and nitrate groundwater in Daqing City, Qinkenpao area, China, is carried out (i) to characterize fluoride and nitrate contamination in the groundwater; and (ii) to identify plausible geochemical reactions under the prevailing hydrogeological conditions, as well as their role in mobilizing fluoride and nitrate concentration.

METHODOLOGY

Area of study

Located in the North Temperate Zone, Daqing belongs to the continental monsoon climate and is affected by the cold air mass from inland Mongolia and the monsoon comes from the warm air mass of the ocean. The area of study located between latitudes 46°32'28" N - 46°42'18" N and longitudes 125°04'19" E - 125°25'14" E covers an area of 549 km^2 (Figure 1). It is semi-arid area characterized by hot dry summers 24°C (April to June) and cold dry winters -26°C (November to February). The annual rainfall is 418 mm, mainly during June to September. Most of the area is flat and plain, where the surface elevation ranges between 174 to 149 m a.s.l.

Geology and hydrogeology

Quaternary sediments are widely distributed in this area, and the thickness ranges from 20 to 40 m. The shallow groundwater belongs to unconfined and confined groundwater, which is located in the quaternary sediment. Unconfined aquifer lies below loess-like loam, and the permeability coefficient is 0.5 m/d. This aquifer lithology is sand and gravel, with the thickness changes from 1 to 10 m and groundwater depth below ground varies from 2 to 8 m. Confined aquifer mainly occurs in sand and gravel, which has a close hydraulic connection with unconfined aquifer. It is important to note that there is a lack of tertiary sediment in this area, and the mudstone aquiclude (about 30 m), which belongs to cretaceous sediment, exists in the underlying cretaceous gravel aquifer and the upper quaternary aquifer. The hydraulic gradient in this area is not large, and the runoff cycle is poor. Moreover, the main recharge of groundwater in this area is from atmospheric precipitation and groundwater runoff, and discharge is via evaporation and lateral runoff (Figure 2).

Sampling and analytical procedure

Groundwater quality monitoring was carried out for 19 well locations in the area of study during post-monsoon 2010. The details of

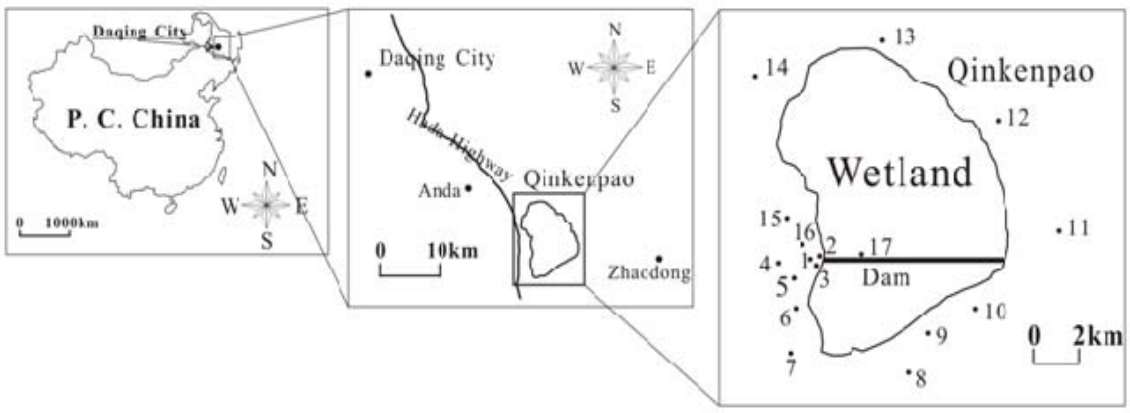

Figure 1. Map of Sartu District, Daqing City, China.

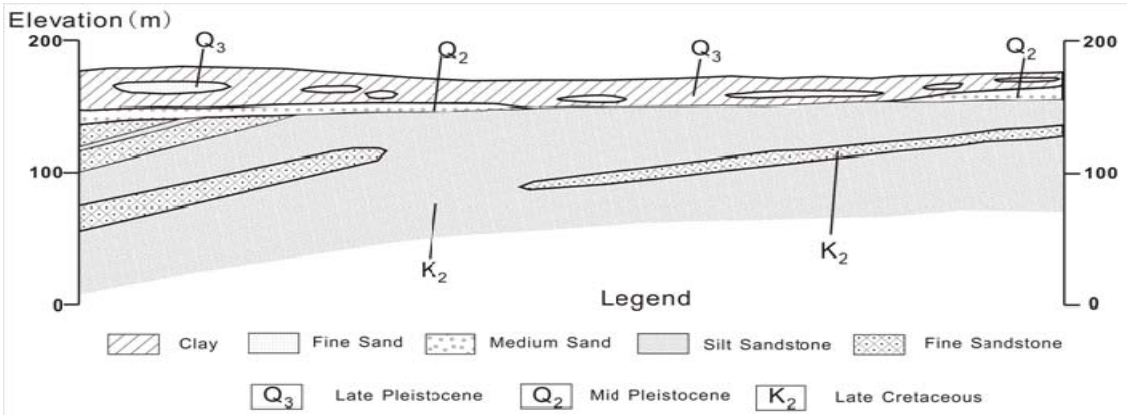

Figure 2. Geological sketch of aquifers in the study area.

location, type and depth of wells are given in Table 1. The polyethylene bottles were used to collect the samples of surface water and groundwater. Water samples were collected from September, 2010. Physicochemical parameters, including temperature, pH, electrical conductivity (EC) and total dissolved solid (TDS), were conducted in the field. Other samples were transported to the laboratory for testing. Major components (HCO_3^-, Cl^-, SO_4^{2-}, NO_3^-, Ca^{2+}, Mg^{2+}, NH_4^+, Na^+ and K^+ and F^-) were measured in laboratory within 10 days of sampling. Samples were stored in the dark in a cold room at 4°C. All of these negative and positive ions are tested using DX-120 ion chromatography, and the detection limits are shown in Table 2 (EPA, 2009).

The cation-anion balance error of 17 samples collected in post-monsoon 2010 is within the permissible limit of 5% and the remaining 2 samples with higher ion balance error were not considered in the study.

RESULTS AND DISCUSSION

Fluoride and Nitrate hydrogeochemistry and aqueous speciation modeling

Concentrations of various groundwater quality parameters obtained for samples collected in post-monsoon 2010 are shown in Table 2. The water type for each sample is also indicated. It is evident that groundwater consists mainly of types Na-HCO_3, Ca-HCO_3 and Ca-NO_3. The calcium concentration ranges between 6.25 and 193.5 mg/L, fluoride between 0.58 and 3.4 mg/L and nitrate between 0 and 624.1 mg/L. The correlation coefficients among various groundwater quality parameters were obtained to investigate their interdependence. The correlation matrix for the hydrochemical data of September 2010 is shown in Table 3. This shows that fluoride and nitrate have a positive relationship with all species, but sulfate and pH for nitrate, and further pH for fluoride have a negative relationship.

The NO_3^- concentrations at some locations (60% of the wells) exceeded the 50 mg/L limit specified by the WHO drinking water standard.

Groundwater also contained high concentrations of NO_3, which indicates a serious water quality issue. This NO_3 originates from the soil surface and enters the groundwater via infiltration. However, due to the influence of land use, there was still a high level of horizontal

Table 1. Well location, and depth of well.

Location	Latitude, N	Longitude, E	Altitude (m a.m.s.l.)	Depth of well (m b.g.l.)
1	46°19′07.1″	125°29′07.0″	163.2	100.0
2	46°19′08.4″	125°29′11.1″	156.9	40.0
3	46°19′05.4″	125°29′08.7″	158.3	34.0
4	46°19′04.0″	125°28′09.0″	170.6	20.0
5	46°18′47.3″	125°28′34.0″	167.5	65.0
6	46°18′02.4″	125°28′42.1″	164.7	10.0
7	46°17′01.0″	125°28′30.0″	163.5	32.0
8	46°16′35.0″	125°32′44.0″	174.2	10.0
9	46°17′30.0″	125°33′21.0″	166.4	20.0
10	46°18′02.8″	125°35′04.0″	164.5	20.4
11	46°19′47.5″	125°38′03.9″	153.8	20.0
12	46°22′16.5″	125°35′50.2″	150.3	7.8
13	46°24′07.6″	125°31′42.5″	156.7	20.0
14	46°23′16.8″	125°27′15.6″	165.3	50.0
15	46°20′04.6″	125°28′23.1″	162.4	37.0
16	46°23′16.8″	125°27′15.6″	167.8	50.0
17	46°19′06.6″	125°29′52.4″	152.8	-

Table 2. Hydrochemical parameters for post-monsoon 2010 (concentrations are in mg/L).

Location	TDS	pH	K	Na	Ca	Mg	Cl	F	SO_4	HCO_3	NO_3	Water Type
1	365.44	8.77	0.56	58.50	51.22	21.76	53.23	1.82	13.60	159.26	5.49	Ca-HCO_3
2	671.96	7.88	0.03	112.06	68.52	28.54	36.45	1.44	190.80	214.33	19.78	Na-SO_4
3	419.29	8.83	0.72	61.82	55.05	25.12	11.51	1.75	13.05	215.52	34.74	Ca-HCO_3
4	1003.87	7.26	0.61	69.03	147.50	39.42	166.21	0.86	22.64	223.56	334.05	Ca-NO_3
5	429.70	7.82	0.64	100.23	44.17	13.57	34.62	0.58	30.18	176.23	29.48	Na-HCO_3
6	576.19	8.23	0.57	44.71	103.14	26.87	75.19	1.18	25.17	164.62	134.75	Ca-HCO_3
7	345.95	8.24	0.95	61.69	42.44	12.68	16.45	0.65	19.98	191.11	0.00	Na-HCO_3
8	1228.24	7.36	0.43	39.72	193.50	39.62	198.30	1.30	36.00	157.77	561.60	Ca-NO_3
9	501.95	7.46	0.33	39.15	89.94	26.31	17.25	1.01	20.28	247.07	60.62	Ca-HCO_3
10	1451.51	7.78	3.20	99.04	166.96	49.53	315.25	2.25	88.90	310.48	415.90	Ca-Cl
11	1910.79	7.65	2.24	340.78	90.72	39.06	314.30	3.40	91.05	405.14	624.10	Na-NO_3
12	1731.95	9.75	1.26	451.12	31.23	38.27	186.24	1.36	124.13	717.41	180.93	Na-HCO_3
13	1123.04	7.66	1.47	121.08	130.06	42.29	201.50	0.96	75.85	276.84	272.98	Ca-Cl
14	532.49	7.70	0.95	71.59	75.36	29.10	26.84	1.52	8.81	262.85	55.47	Ca-HCO_3
15	654.17	7.98	1.02	121.18	72.93	30.59	66.52	2.94	31.37	294.70	32.91	Na-HCO_3
16	1120.36	7.89	0.74	59.67	161.13	42.04	190.99	1.46	24.81	178.61	460.91	Ca-NO_3
17	1181.18	9.40	1.30	340.01	6.25	9.58	207.98	0.97	0.00	363.17	251.92	Na-HCO_3
	Detection limit		0.10	0.10	0.10	0.10	0.10	0.10	0.25	3	0.20	

variation in groundwater quality in the upper aquifer. NO_3 and cation concentrations (Ca^{2+} and Mg^{2+}) showed strong correlations indicating that they originated from the same sources (Figure 3).

The upper limits of the point distribution for observed fluoride and calcium concentrations form a hyperbolic curve that suggests solubility of calcium- and fluoride-containing minerals controlling the fluoride concentration (Figure 4). High fluoride with very low calcium and magnesium in water may be due to prior precipitation of $CaCO_3$ from water and only limited incorporation of fluoride in the calcite structure, so that there is always a net balance of fluoride in the solution.

Aqueous speciation modeling was carried out using PHREEQC (Parkhurst and Appelo, 1999) geochemical code via AquaChem 5.1 (Waterloo Hydrogeologic Inc.,

Table 3. Correlation coefficients matrix for post-monsoon 2010 hydrochemical data.

Element	Ca	Mg	Na	K	Cl	F	SO_4	HCO_3	NO_3	TDS	pH
Ca	1	0.784	-0.441	0.188	0.478	0.1	-0.13	-0.314	0.656	0.319	-0.691
Mg		1	0.025	0.424	0.637	0.4	0.226	0.236	0.627	0.637	-0.388
Na			1	0.397	0.489	0.259	0.441	0.896	0.227	0.702	0.597
K				1	0.754	0.486	-0.013	0.442	0.528	0.632	0.053
Cl					1	0.373	0.143	0.429	0.888	0.92	-0.046
F						1	0.126	0.25	0.271	0.395	-0.05
SO_4							1	0.339	-0.06	0.334	0.107
HCO_3								1	0.145	0.676	0.547
NO_3									1	0.806	-0.287
TDS										1	0.079
pH											1

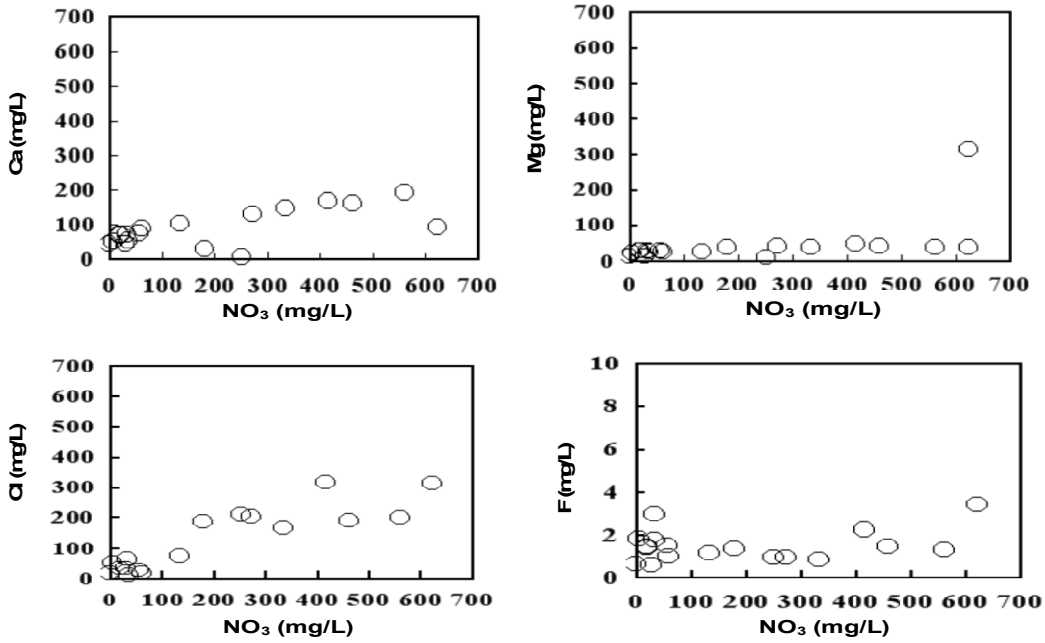

Figure 3. Cation (Ca^{2+} and Mg^{2+}) and anion (F^- and Cl^-) versus NO_3^- concentrations in groundwater.

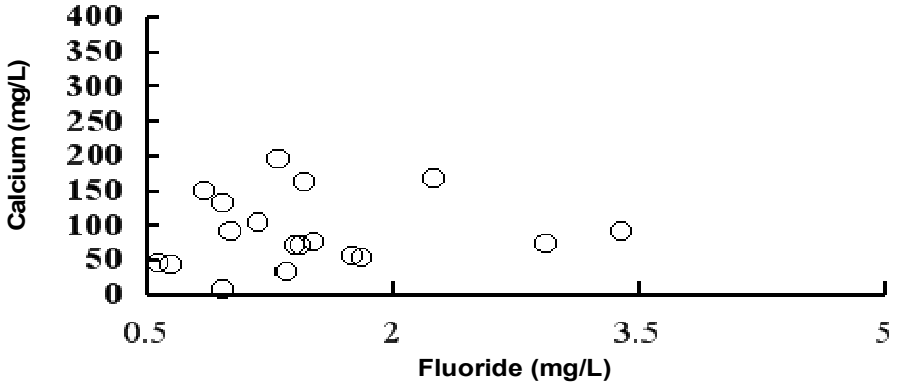

Figure 4. Relationship between calcium and fluoride concentration.

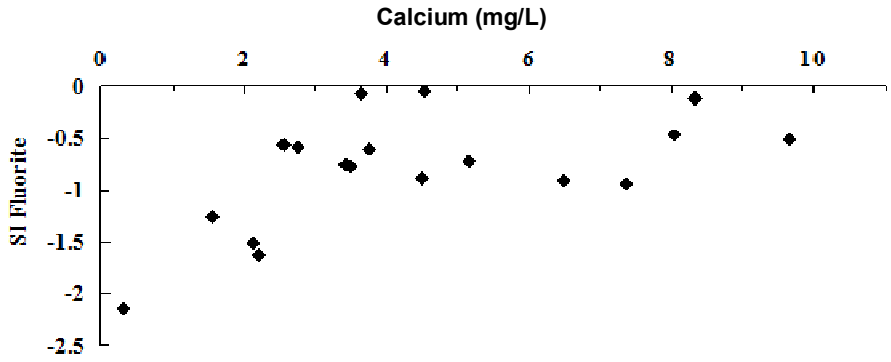

Figure 5. Plot of saturation indices of fluorite versus calcium concentration.

Table 4. Saturation index.

Well No.	SI anhydrite	SI aragonite	SI calcite	SI dolomite	SI fluorite	SI gypsum	SI halite
1	-2.0696	-0.3273	-0.1799	-0.4365	-0.0453	-1.8335	-5.6201
2	-2.3956	-0.4054	-0.258	-0.6096	-0.0716	-2.1586	-6.6778
3	-1.7993	-0.1499	-0.0024	-0.2443	-0.1128	-1.5628	-6.1418
4	-2.8094	-0.7648	-0.6174	-1.3233	-0.5607	-2.5723	-7.0696
5	-2.8246	-0.6192	-0.4717	-1.0003	-0.5872	-2.5875	-7.7168
6	-2.9105	-0.4228	-0.2753	-0.679	-0.6089	-2.6735	-7.2957
7	-2.3311	-0.3763	-0.2288	-0.7522	-0.4667	-2.0946	-6.5728
8	-1.6651	-0.6075	-0.4601	-1.0192	-0.7503	-1.4281	-6.975
9	-1.6293	-0.5938	-0.4463	-0.9955	-0.7734	-1.3923	-6.9723
10	-2.3233	-0.5106	-0.3632	-0.3509	-1.2585	-2.0868	-5.7029
11	-2.1222	-0.3693	-0.2219	-0.8428	-0.5083	-1.8858	-6.7409
12	-2.3643	-0.5152	-0.3677	-1.0342	-0.7208	-2.1273	-7.0639
13	-2.4764	-0.3743	-0.2269	-0.7026	-0.8849	-2.2394	-7.7493
14		-1.4277	-1.2803	-2.0872	-2.1392		-5.7602
15	-1.9107	-0.2697	-0.1223	-0.445	-0.9089	-1.6741	-6.2329
16	-2.3758	-0.297	-0.1496	-0.5838	-0.9375	-2.1391	-6.5602
17	-2.6851	-0.7539	-0.6064	-1.4538	-1.5082	-2.4479	-7.5497

2009). The saturation indices of fluorite were computed for all three sets of temporal data, sampled as well as earlier hydrochemical data. Figure 5 shows the plot of these saturation indices versus calcium concentrations. Results show that the groundwater is undersaturated with calcite, having a saturation index of -0.0024 to -0.6174 with an equilibrium state. The groundwater is undersaturated with fluorite, having a saturation index of -0.0453 to -2.1392 (Table 4).

Multivariate analysis

The factor analysis was carried out using the hydrochemical data of the area of study. These data were considered for multivariate analysis without combining sampled data to make the homogeneous data set with reference to dynamic changes in hydraulic stresses, land-use characteristics and pollutant sources. The chemical constituents considered for factor analysis for the unconfined aquifer were calcium, magnesium, sodium, potassium, bicarbonate, sulphate, chloride, fluoride, nitrate, pH and TDS. The principal component analysis was carried out using Statistical Package for Social Services (SPSS) software. The two-factor model results revealed that the first two eigenvalues extracted from the matrix account for more than 72% of total variance, which shows that the hydrochemical data is well posed. A varimax rotated component matrix with Kaiser (1958) normalization was used for principal component analysis. The interpretation of factors was made in terms of the square of the coefficients of that factor. The rotated component matrix for the geochemical data is given in Table 5. There is almost identical loading for sodium,

Table 5. Rotated component matrix.

Parameter	Factor 1	Factor 2
NO_3	0.910	0.121
Mg	0.870	-0.228
Cl	0.858	0.411
Ca	0.833	-0.510
TDS	0.746	0.630
K	0.600	0.455
F	0.420	0.329
Na	0.324	0.973
HCO_3	0.365	0.919
pH	-0.277	0.823
SO_4	0.185	0.352

magnesium, nitrate and bicarbonate. Therefore, the variance in the chemical composition of the hydrochemical system is controlled by sources of sodium and bicarbonate. For Factor 1, the sum of the squares of calcium and magnesium (1.45) is approximately that of nitrate and chloride bicarbonate, sulphate and chloride (1.56). Thus, the combined relationship suggests that there is more than one component or more than one solid phase that adds or removes calcium, magnesium, nitrate and chloride into the groundwater. For Factor 1, there is no replacement mechanism as there is a lack of mutually exclusive components. Nitrate is naturally present in groundwater at very low concentrations, and its source is human activity such as domestic or industrial waste or agricultural. Factor 1 shows calcium chloride and magnesium chloride waters. Factor 2 shows that the sum of the squares of sodium and potassium (1.15) is approximately that of bicarbonate, sulphate and chloride (1.37). The combined relationship suggests that there is more than one component or more than one solid phase that adds or removes sodium, potassium, bicarbonate, sulphate and chloride. The presence of negative correlations indicates that some components are controlled by equilibrium with the minerals in the aquifers. Thus, there is a reaction path by which one set of chemical products replaces another set. Moreover, it is observed that there is positive correlation between calcium and fluoride for both the factors.

For Factor 1, there is probably dissolution of fluorite, calcite and dolomite, whereas for Factor 2, the decrease in fluoride may be due to adsorption on clay surfaces. Thus, these results obtained from factor analysis help in understanding the possible grouping of chemical constituents in the groundwater.

Geochemical reactions

The actual changes in concentrations of chemical species as a function of sulphate concentration help in obtaining the information on possible geochemical reactions that may be occurring in the area of study. To investigate this, analysis was carried out using hydrochemical data. The increase in calcium and magnesium and pH as sulphate increases are shown in Figure 6a-c. The groundwater in the area of study is undersaturated with calcite and dolomite (Table 4), and dissolution of calcite and dolomite adds calcium and magnesium to the groundwater. Calcite dissolution causes a pH increase due to consumption of H^+ by carbonate during the dissolution process. The decrease in the CO_2 in the solution leads to the dissolution of calcite and dolomite and CO_2 dissolution, thereby increasing the magnesium and calcium concentration in the solution. The geochemical equations for possible reactions in the aquifer are as follows:

$$CaCO_3 \rightarrow Ca + CO_3 \quad (1)$$

$$CaMg(CO_3)_2 \rightarrow Ca + Mg + 2CO_3 \quad (2)$$

$$CO_3 + H^+ \rightarrow HCO_3 \quad (3)$$

$$CO_2(g) \rightarrow CO_2(aq) \quad (4)$$

$$NaCl \rightarrow Na + Cl \quad (5)$$

$$KCl \rightarrow K + Cl \quad (6)$$

Where (g) in Equation 4 refers to the gaseous phase and (aq) to the aqueous phase.

Although the trends in calcium, magnesium and pH with sulphate are evident in the groundwater in the area of study, there is variation in the temperature and other reactions in addition to dedolomitization or dissolution of carbonates. Dedolomitization is a specific geochemical process and has been reported by Plummer et al. (1990)

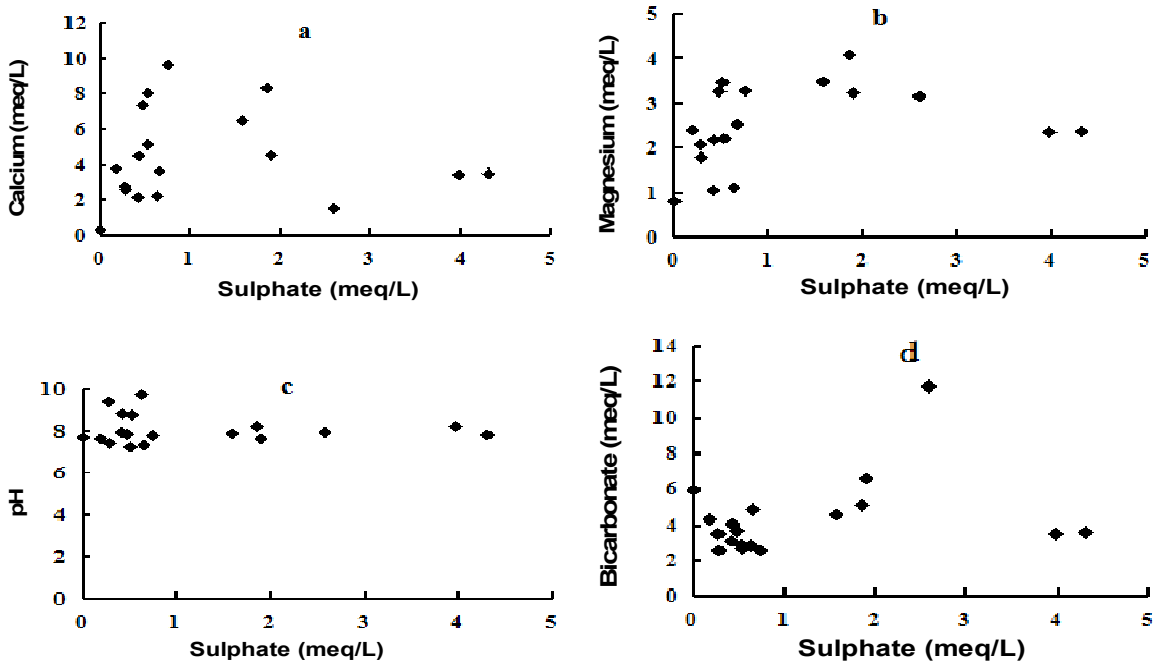

Figure 6. Plot of calcium, magnesium, pH and bicarbonate alkalinity as a function of sulphate.

and Kloss and Goebelbecker (1992). The plot of bicarbonate alkalinity as a function of dissolved sulphate concentration (Figure 6d) shows that bicarbonate alkalinity is decreasing as sulphate increases, and is possibly indicative of dedolomitization or carbonate dissolution. Cation exchange with calcium and magnesium cations could contribute to additional bicarbonate on the flow path with uptake of calcium and magnesium and release of sodium from exchange sites on clay minerals causing dissolution of carbonate minerals. The molar concentrations of sodium plus potassium are slightly more than the concentration of chloride. This is indicative of presence of evaporites. These waters have high bicarbonate concentration. Thus, there is a tendency to form sodium-bicarbonate waters. For the groundwater in the area of study of Qinkenpao area, the sodium bicarbonate water is derived from the dissolution of carbonate minerals.

Conclusion

Fluoride (0.58 to 3.4 mg/L) and nitrate (0 to 624.1 mg/L) concentrations in groundwater are observed in large parts of the Qinkenpao area, Daqing, China, and are a matter of concern for drinking water supply since it exceeds the maximum permissible fluoride concentration of 1.5 mg/L and nitrate concentration of 50 mg/L for public water supply systems. Factor analysis showed that sodium bicarbonate waters have high fluoride, whereas calcium chloride and magnesium chloride waters have low factor loading for fluoride. Results obtained from aqueous speciation modeling reveal that the groundwater is undersaturated with calcite, indicating dissolution of calcite and dolomite, and undersaturated with fluorite, possibly indicative of its dissolution. The geochemical reactions in the area of study indicate dissolution of calcite and dolomite, carbon dioxide and sulphate-bearing minerals with ion exchange.

Conflict of Interest

The author(s) have not declared any conflict of interest.

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Full Length Research Paper

Effect of culture filtrate of *Curvularia inaequalis* on disease control and productivity of grape cv. Isabel

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This study aimed to evaluate the effect of a culture filtrate of the saprophytic fungus *Curvularia inaequalis* on the control of the foliar diseases downy mildew and isariopsis leaf spot of grapevine and on the incidence and severity of grape fruit mildew under field conditions. This study also analyzed the physical and physicochemical characteristics of fruits that were treated preharvest during standard season production and off season production. Concentrations of 0 (absolute control), 1, 5, 10 and 20 ml L^{-1} culture filtrate were tested. The standard treatments that were used as controls were bordeaux mixture, mancozeb, acibenzolar-S-methyl and *Agaricus blazei*. In the case of downy mildew, the concentrations reached a control of approximately 56% compared to that of the absolute control treatment during both seasons. The reduction of the isariopsis leaf spot was approximately 54 and 42% in the standard season and off season, respectively. The severity of fruit mildew was controlled in 62% of the cases, but the incidence was controlled in approximately 11% of the cases. Postharvest, the pH and relationship between solids and acidity (TSS/TTA) of the fruit pulp remained in the ideal range, the fruits showed low juice yield and high total titratable acidity, and the bunch weight, number of bunches and yield per plant were higher in the off season than in standard season and were affected by weather conditions and the pruning season.

Key words: *Vitis labrusca*, biological control, agro-ecology, organic production.

INTRODUCTION

In Southern Brazil, as in other worldwide grape-producing regions, downy mildew caused by *Plasmopara viticola* (Berk and Kurt) Berlese and de Toni is a major disease in grapevines (Dalbó and Schuck, 2003), affecting all of the green plant organs, mainly leaves, inflorescences and young fruits. However, under climatic conditions of high rainfall and temperatures that are favorable to fungal growth, *Isariopsis clavispora* (Berkeley and Curtis) Saccardo, the etiologic agent of isariopsis leaf spot, becomes highly dangerous to vineyards due to the premature defoliation of plants which can cause branch aging and weakening in the following season (Lenz et al., 2009).

Disease management in vineyards still constitutes a major problem for viticulture and when not controlled seriously can be responsible for significant losses in

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production both in the quantity and quality of berries and their products (Özer et al., 2012; Pinto et al, 2013). According to Chavarria et al. (2007), the grape producer sprays chemicals weekly to guard the harvest, applying the fungicides cymoxanil, azoxystrobin and mancozeb, among other synthetic chemicals that are used in controlling mildew. To control isariopsis leaf spot, the recommended treatments for grape downy mildew and anthracnose are usually enough (Amorim and Kuniyuki, 2005).

When considering the economic and environmental costs of such applications as well as the increasing restrictions that are related to the presence of toxic residues on the fruits, the search for new alternatives is necessary. Among these alternatives, biological control may be an important and justifiable technical alternative, mainly for wine-growers in an agroecological production system, which has few options at a commercial level for controlling these diseases. Therefore, saprobe fungi currently receive special attention as controlling agents as well as potential resistance inducers.

Similarly, the fungus *Curvularia inaequalis* (Shear) belongs to the group of Hyphomycetes dematiaceous fungi and is pathogenic to plants and/or is saprophytic. *C. inaequalis* produces chloroperoxidase, a vanadium enzyme that has similar bromoperoxidase properties and forms hypochlorous acid (HOCl), a strong oxidizing agent and bactericide (Van Schijndel et al., 1993; Hemrika et al., 1999). Hansen et al. (2003) found that the haloperoxidase system of *Curvularia* rapidly acts as an antimicrobial agent against a broad spectrum of bacteria (*Pseudomonas* spp., *Escherichia coli*, *Serratia marcescens*, *Aeromonas salmonicida*, *Shewanella putrefaciens*, *Staphylococcus epidermidis* and *Listeria monocytogenes*), yeasts (*Candida* sp. and *Rhodotorula* sp.) and filamentous fungi (*Aspergillus niger*, *Aspergillus tubigensis*, *Aspergillus versicolor*, *Fusarium oxysporum*, *Penicillium chrysogenum* and *Penicillium paxilli*) when cultivated in suspension.

This study aimed to determine the antagonistic potential of the saprophytic fungus *C. inaequalis* which was obtained from the litter of the semi-arid region of the northeastern Brazil in relation to the severity of grape foliar diseases (downy mildew and isariopsis leaf spot) and the incidence and severity of downy mildew in grape berries cv. Isabel under field conditions and to analyze the physical and physicochemical characteristics of fruits that were treated pre-harvest during the standard season and off season using culture filtrates of the saprophytic fungus.

MATERIALS AND METHODS

Obtaining the culture filtrate of the saprophytic fungus *C. inaequalis* from the Brazilian semiarid region

Discs of the saprophytic fungus *C. inaequalis* (CUI) mycelium were transferred to an Erlenmeyer flask containing liquid culture medium potato-dextrose (PD) in the proportion of five mycelial discs for 1000 ml liquid medium. The cultivation was carried out at 25±1°C for seven days in a 12 h photoperiod. After this period, the suspension was filtered through a thin fabric to obtain the culture filtrate.

Field experiment

The experiment was conducted during two consecutive seasons from July to December 2011 (standard season) and from February to June 2012 (late season) in a commercial vineyard with cv. Isabel in Marialva County, Paraná State, Brazil. The county's geographical coordinates are latitude 23°29'8"S, longitude 51°47'34"W and altitude 644 m.a.s.l. (Iapar, 2000). Four-year-old plants that were grafted on Paulsen 1103 rootstock were spaced in 2.5 m x 2.0 m arrays (plant density approximately 5,000 plants ha^{-1}) and trained in a trellis system.

The applied treatments were 0, 1, 5, 10 and 20 ml L^{-1} (0, 1, 5, 10 and 20%, respectively) of the culture filtrate CUI. The zero concentration (0 ml L^{-1} filtrated CUI) consisted of an absolute control without treatment. Furthermore, the standard treatments that were used as controls were bordeaux mixture in a proportion of 1:1:100 (copper sulfate: quicklime: water, v:v:v), mancozeb at 2.5 g p.c. L^{-1} (Manzate® 800, Dow AgroSciences Industrial Ltda.), acibenzolar-S-methyl at 0.05 g p.c. L^{-1} (Bion®, Syngenta Proteção de Cultivos Ltda.) and *Agaricus blazei* (1 g of dry powder per 14 ml water).

Fungicides were sprayed with hand sprayer weekly at approximately 7:00 a.m. under the proper environmental conditions aiming for a high-quality application (Guarany®, adjustable conic nozzle) using three liters of the solution per treatment. The fungicides were applied until the point of runoff from the onset of sprouting on September 1, 2011 for the first season and on February 14, 2012 for the second season for a total of 10 and 8 applications per crop season, respectively. The surrounding plants were treated weekly with mancozeb.

At the appearance of the first symptoms of downy mildew, the disease severity was evaluated at three leaves in the apex of four branches per plant that was previously identified using the diagrammatic scale that is described by Azevedo (1997). The severity assessment of the isariopsis leaf spot began at the end of each crop season by examining the six leaves in the basal area of the plants that were previously identified through the diagrammatic scale that is described by Lenz et al. (2009).

Data on the severity were determined through the area under the disease progress curve (AUDPC) according to Shaner and Finney (1977). Five evaluations for downy mildew and four evaluations for isariopsis leaf spot were respectively conducted at intervals of seven days during both the first and second seasons.

At the end of the experiment during the first season, the symptoms of downy mildew on grape fruits were identified and the incidence and severity of that disease were evaluated. In the following season, downy mildew was not present on the fruits.

The experimental design was a randomized block pattern of nine treatments and five replicates with one plant per plot during the two evaluation periods. The results were submitted to a variance analysis, to a polynomial regression analysis and to a mean comparison by the Tukey test at 5% probability using the statistical program SISVAR (Sisvar 5.1 Build 72, UFLA) (Ferreira, 2011).

Physical and physicochemical determinations

At the end of the field experiment during each season, the number of grape bunches per plant were counted and then harvested for physical and physicochemical analyses of the grape fruits that were produced after each treatment. For these analyses, each treatment

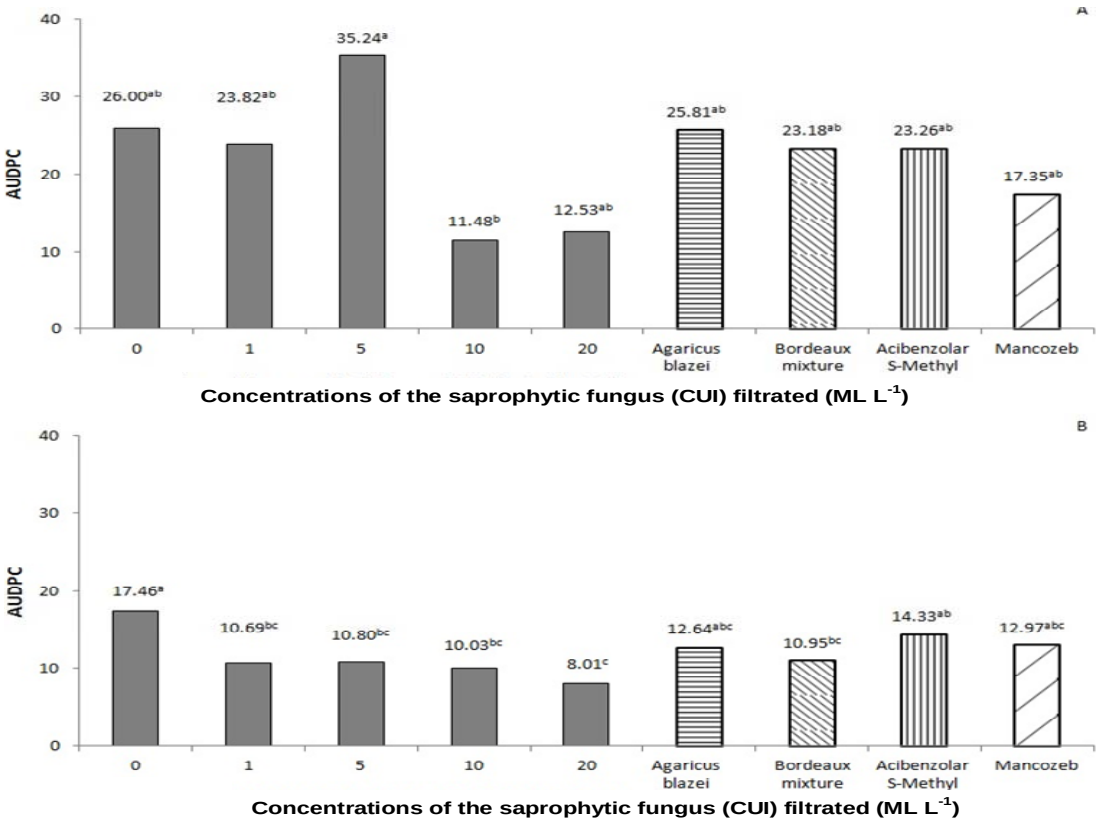

Figure 1. Effect of concentrations of the saprophytic fungus (Curvularia inaequalis) filtrated, in the area under the disease progress curve (AUDPC), on downy mildew in grape cv. 'Isabel', in the first (A) and second year of study (B) under field conditions. [1] Averages followed by different letters differ by the Tukey test (p>0.05). * Significant at 5% probability by test F. The data on severity were transformed through X root for performing the statistical analysis.

consisted of five replicates with three to four bunches of grapes per plot during both of the evaluation periods. The plants were harvested when the grapes presented on average 14 ºBrix.

The first analyses were as follows: a) cluster weight (g) determined using a digital analytical balance; after the extraction of the grape juice, the samples were homogenized and subjected to the following analysis; b) total soluble solids (TSS, °Brix) determined using a portable refractometer (Instituto Adolfo Lutz, 1985); c) total titratable acidity (TTA, %) determined using a titration meter with a 0.1 N NaOH solution; the values are expressed in grams of tartaric acid per 100 ml wort (Instituto Adolfo Lutz, 1985); d) relationship between solids and acidity determined using the ratio TSS/TTA; e) pH determined in using a potentiometer according to the norms that were established by Instituto Adolfo Lutz (1985); f) productivity per plant (g); and g) juice yield (%).

The results were submitted to a variance analysis to a polynomial regression analysis and to a mean comparison by the Tukey test at 5% probability using the statistical program SISVAR (Sisvar 5.1 Build 72, UFLA) (Ferreira, 2011).

RESULTS

Field experiment

In this study, concentrations of 10 and 20 ml L^{-1} of the culture filtrate CUI in the first cycle were statistically similar to the standard treatments and reduced the severity of downy mildew by 55.84 and 51.80%, respectively compared to the absolute control treatment. The standard treatments mancozeb, bordeaux mixture and acibenzolar-S-methyl decreased the disease in 33.26, 10.82 and 10.54% of the cases, respectively when compared to the absolute control treatment.

During this season, there was a downtrend AUDPC treatment 1 ml L^{-1} filtrate CUI with an increase of the disease in relation to the 5 ml L^{-1} filtrate treatment followed by stabilization by the 10 and 20 ml L^{-1} filtrate treatment. This pattern led to a low R^2 for the linear regression and non-significant R^2 for the quadratic, requiring further study (Figure 1A). The regression analysis of the second cycle was similar to that of the 2011 harvest, although the results of the AUDPC were more stable (Figure 1B).

In the second year of the study, there were similar reductions in the severity of downy mildew. The highest concentration (20 ml L^{-1}) of the filtrated CUI showed a control of 54.13% when compared to the absolute control treatment. The concentration of 20 ml L^{-1} filtrate was also not significantly different from the other concentrations (1, 5 and 10 ml L^{-1}) or from the bordeaux mixture, *Agaricus*

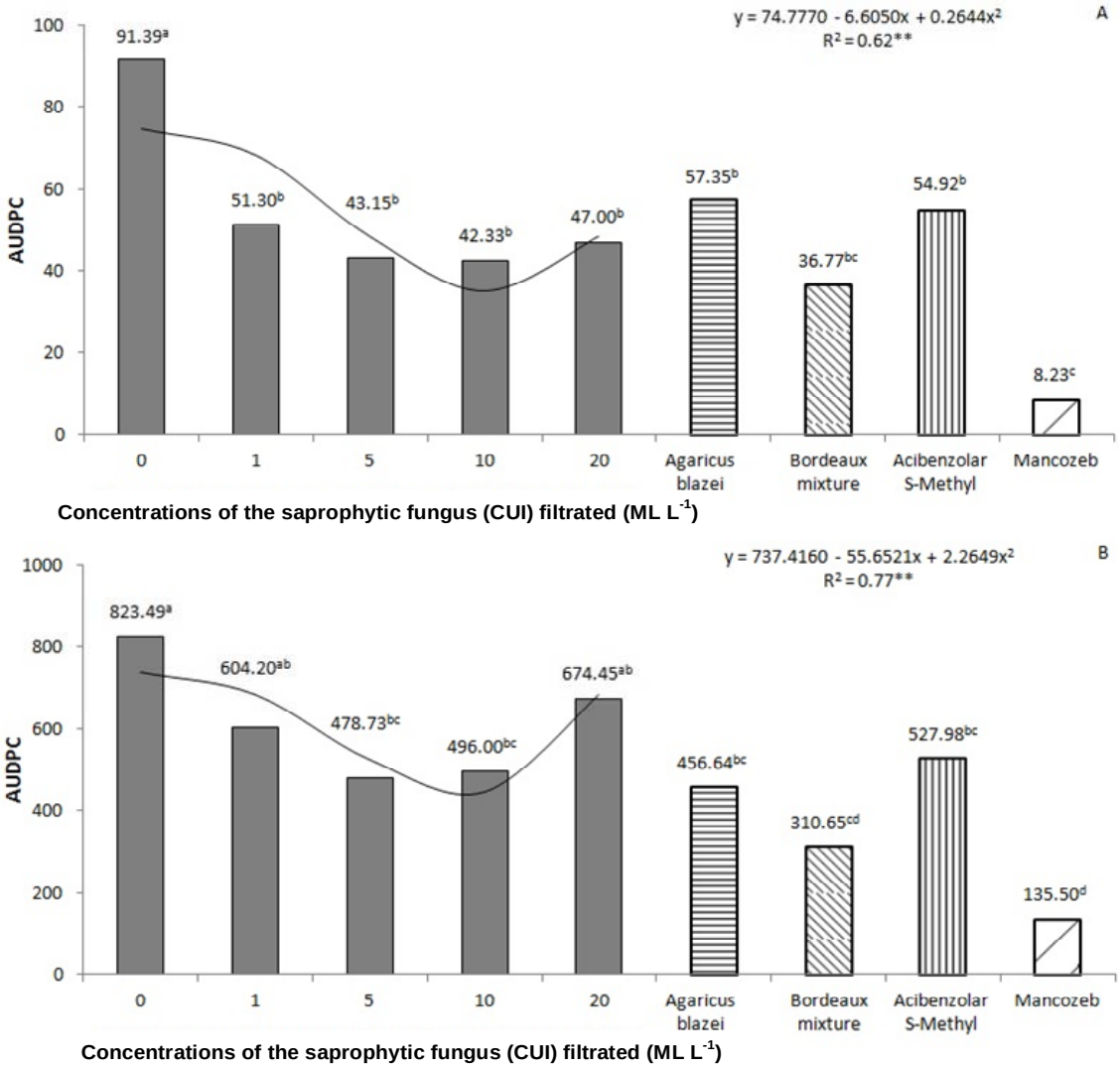

Figure 2. Effect of concentrations of the saprophytic fungus (*Curvularia inaequalis*) strain, in the area under disease progress curve (AUDPC), in isariopsis leaf spot of grapevine cv. 'Isabel', in the first (A) and second years of study (B), under field conditions. [1]Averages followed by different letters differ by the Tukey test ($p>0.05$). ** Significant at 1% probability by test F.

blazei and mancozeb which presented smaller reductions in the disease, decreasing the downy mildew severity by 38.76, 38.17, 42.53, 37.28, 27.61 and 25.72%, respectively, compared to the absolute control treatment (Figure 1B).

Regarding the AUDPC of isariopsis leaf spot in both the first and the second cycles, the concentrations of 1, 5, 10 and 20 ml L^{-1} did not differ significantly from the standard treatments with the bordeaux mixture, *A. blazei* and acibenzolar-S-methyl. In the first cycle, the reductions were 43.87, 52.78, 53.68, 48.57, 59.76, 37.24 and 39.90%, respectively, compared to the absolute control treatment (Figure 2A). Similar to the previous year, the reduction of the severity of isariopsis leaf spot in the second cycle ranged from 18 to 62% (Figure 2B). In both years, studying the AUDPC of this disease, there was a quadratic effect in function to the concentrations of filtrated CUI utilized.

For the incidence of downy mildew on grape bunches, the concentrations of 10 and 20 ml L^{-1} filtrate CUI were statistically similar to the standard treatments of *A. blazei*, mancozeb and bordeaux mixture. The reductions in the incidence rates were 9.89 and 11.14% for 10 and 20 ml L^{-1} filtrate, respectively, and 6.21, 34.75 and 59.07% for the standard treatments, respectively, compared to the absolute control treatment (Table 1).

However, regarding the mildew severity on grape bunches, the reductions ranged from 8 to 86%. The concentrations of 1 and 10 ml L^{-1} filtrate CUI did not differ statistically from the standard treatments. Furthermore, the 5 and 20 ml L^{-1} filtrate concentrations were statistically equivalent to the 10 ml L^{-1} concentration

Table 1. Incidence and severity index of downy mildew on grape bunches treated with saprophytic fungus strain Curvularia inaequalis (CUI). Marialva County, Paraná State, Brazil, 2011 harvest.

Treatments (ml L^{-1})	Incidence of Disease (%)	Severity Index
0	24.01^{bc}	2.52^{a}
1	27.91^{ab}	0.96^{cd}
5	43.36^{a}	2.30^{ab}
10	21.63^{bc}	1.10^{bcd}
20	21.33^{bc}	1.75^{abc}
Agaricus blazei	22.52^{bc}	0.98^{cd}
Bordeaux mixture	9.83^{c}	0.35^{d}
Acibenzolar S-Methyl	24.91^{bc}	1.31^{abcd}
Mancozeb	15.67^{bc}	0.70^{cd}
CV (%)	33.92	46.75

*Averages followed by the same letter in columns do not differ significantly by Tukey test at 5% probability. The severity index data were processed by the root of x to perform statistical analysis.

and to the standard treatment acibenzolar-S-methyl, despite the 5 ml L^{-1} concentration exhibiting the lowest culture filtrated performance. In relation to the absolute control treatment, the concentrations of 1, 5, 10 and 20 ml L^{-1} filtrate decreased the severity of downy mildew on grape bunches by 61.85, 8.63, 56.32 and 30.23%, respectively. The standard treatments acibenzolar-S-methyl, *A. blazei*, bordeaux mixture and mancozeb decreased the severity by 47.93, 61.14, 72.16 and 86%, respectively, compared to the absolute control treatment (Table 1).

Physical and physicochemical determinations

When analyzing the effect of the concentrations of the CUI culture filtrate on the physical and physicochemical characteristics of grape fruits, in general, the juice pH did not vary greatly between the two years ranging from 2.92 to 3.23. In the first cycle, the pH ranged from 3.07 to 3.23 (Table 2) and in the second cycle from 3.02 and 3.18 (Table 3).

In the first cycle, the values for total soluble solids (TSS) reached the minimum value established for a grape juice with identity and quality which is 14.00 °Brix except for the standard treatments acibenzolar-S-methyl (12.50 °Brix), *A. blazei* (12.66 °Brix) and bordeaux mixture (13.55 °Brix) (Table 2). However in the second cycle, the TSS values for all of the treatments were inferior to the established minimum, except for the treatment of mancozeb which showed 15.52 °Brix (Table 3). In relation to the total titratable acidity (TTA) in the first cycle, there was no significant difference between the treatments (Table 2) ranging from 0.90 to 1.37% tartaric acid. In the following year, there was also no significant difference between the treatments (Table 3).

The TSS/TTA ratio is indicative of the grape juice quality, that is, the degree of sweetness, and the limiting values that were established by legislation are between 15 and 45 (Miguel et al., 2009; Pinheiro et al., 2009). However, in the first cycle, the average values for this ratio ranged from 10.06 to 20.25, which were transformed by log (x) to perform statistical analyses. There was no significant difference between the treatments in the first cycle (Table 2). In the next cycle, the TSS/TTA ratio ranged from 16.66 to 27.05, which is consistent with the values that are required for quality standards. The concentration of 5 ml L^{-1} CUI filtrate did not differ statistically from the standard treatments of mancozeb, bordeaux mixture and *A. blazei*, which showed the highest TSS/TTA values (Table 3).

Regarding the juice yield in the first cycle, there was a significant difference between the treatments and the bordeaux mixture had the highest yield (44.39%), followed by acibenzolar-S-methyl, mancozeb, and 0, 1 and 10 ml L^{-1} filtrated (Table 2). In the following year, the juice yield ranged from 8.59 to 19.22, and there was no significant difference between the treatments in this cycle (Table 3).

In the two cycles, there was no significant difference between the treatments for the variable bunch weight. In the first cycle, the mean values varied from 46.75 (*Agaricus blazei*) to 89.74 g (mancozeb) (Table 2), and in the next cycle from 175.92 (acibenzolar-S-methyl) to 268.27 g (20 ml L^{-1} culture filtrate) (Table 3).

For the variable number of bunches per plant, it was noted that concentrations of 0, 5 and 20 ml L^{-1} filtrate CUI in addition to the standard treatment mancozeb produced the highest results ranging from 26.20 to 41.80 bunches per plant. The other treatment ranged from 12 to 18 bunches per plant (Table 2). In the second cycle, there was no significant difference between the treatment

Table 2. Average values of pH, total soluble solids (TSS), total titratable acidity (TTA), ratio (TSS/TTA), juice yield, bunch weight, bunches per plant and plant yield for grapevine cv. 'Isabel' treated with the strain from saprophytic fungus Curvularia inaequalis (CUI), in Marialva County, Paraná State, Brazil, 2011 harvest.

Treatment (ml L^{-1})	pH	TSS (°Brix)	TTA (%)	TSS/TTA	Juice yield (%)	Bunch weight (g)	Bunches per plant	Yield per grapevine (kg)
0	3.09^{ab}	15.74^{abc}	1.29^{a}	1.10^{a}	35.66^{ab}	73.96^{a}	28.80^{abc}	2.14^{ab}
1	3.23^{a}	17.24^{a}	1.02^{a}	1.23^{a}	36.52^{ab}	48.80^{a}	18.40^{bcd}	0.90^{b}
5	3.07^{ab}	14.58^{abc}	1.03^{a}	1.20^{a}	26.09^{b}	79.77^{a}	31.80^{ab}	2.61^{ab}
10	3.10^{ab}	15.53^{abc}	1.08^{a}	1.17^{a}	34.67^{ab}	60.11^{a}	12.40^{d}	0.99^{b}
20	3.09^{ab}	14.90^{abc}	1.27^{a}	1.08^{a}	27.84^{b}	49.80^{a}	26.20^{abcd}	1.40^{b}
Agaricus blazei	2.73^{c}	12.66^{bc}	1.08^{a}	1.13^{a}	29.84^{ab}	46.75^{a}	13.00^{cd}	0.70^{b}
Bordeaux mixture	2.98^{abc}	13.55^{abc}	1.13^{a}	1.07^{a}	44.39^{a}	47.80^{a}	16.60^{bcd}	1.04^{b}
Acibenzolar S-Methyl	2.95^{bc}	12.50^{c}	1.37^{a}	0.97^{a}	39.99^{ab}	49.32^{a}	17.00^{bcd}	1.04^{b}
Mancozeb	3.21^{ab}	16.58^{ab}	0.90^{a}	1.27^{a}	38.11^{ab}	89.74^{a}	41.80^{a}	3.83^{a}
CV (%)	4.42	12.85	31.36	18.29	21.42	36.82	33.60	59.82

Averages followed by the same letter in columns do not differ significantly by the Tukey test at 5% probability. TSS/TA ration data were transformed to log (x) to perform statistical analysis.

Table 3. Average values of pH, total soluble solids (TSS), total titratable acidity (TTA), TSS/TA ratio, juice yield, bunch weight, bunches per plant and plant yield for grapevine cv. 'Isabel' treated strain from saprophytic fungus Curvularia inaequalis (CUI), in Marialva County, Paraná State, Brazil, 2012 harvest.

Treatment (ml L^{-1})	pH	TSS (°Brix)	TTA (%)	TSS/TTA	Juice yield (%)	Bunch weight (g)	Bunches per plant	Yield per grapevine (kg)
0	3.08^{ab}	12.74^{b}	1.30^{a}	17.62^{b}	0.31^{a}	208.84^{a}	25.80^{a}	1.46^{a}
1	3.04^{abc}	10.80^{b}	1.24^{a}	16.66^{b}	0.27^{a}	208.84^{a}	20.60^{a}	1.34^{a}
5	3.02^{bc}	11.96^{b}	1.34^{a}	21.45^{ab}	0.32^{a}	221.85^{a}	21.40^{a}	1.24^{a}
10	3.06^{abc}	12.00^{b}	1.26^{a}	19.25^{b}	0.31^{a}	223.18^{a}	35.20^{a}	2.37^{a}
20	3.18^{a}	12.66^{b}	1.24^{a}	19.55^{b}	0.35^{a}	268.27^{a}	23.40^{a}	1.61^{a}
Agaricus blazei	3.07^{abc}	11.96^{b}	1.34^{a}	21.54^{ab}	0.30^{a}	179.14^{a}	22.20^{a}	1.01^{a}
Bordeaux mixture	3.12^{ab}	13.30^{ab}	1.28^{a}	21.90^{ab}	0.33^{a}	240.29^{a}	26.50^{a}	1.56^{a}
Acibenzolar S-Methyl	3.06^{abc}	10.96^{b}	1.30^{a}	18.36^{b}	0.27^{a}	175.92^{a}	27.80^{a}	1.28^{a}
Mancozeb	2.93^{c}	15.52^{a}	1.32^{a}	27.05^{a}	0.31^{a}	229.83^{a}	21.40^{a}	1.14^{a}
CV (%)	2.38	10.63	7.08	15.64	15.72	38.59	57.29	68.52

Averages followed by the same letter in columns do not differ significantly by the Tukey test at 5% probability. The TTA data were transformed to $(1/\sqrt{x})$ to perform statistical analysis.

ranging from 20.6 to 35.20 bunches per plant (Table 3).

Tables 2 and 3 present the plant production for the concentrations of the filtrate CUI in the first and second crop cycles, respectively. In the first cycle, there were higher productivities for the treatments with mancozeb (3.83 kg), 0 ml L^{-1} culture filtrate (2.14 kg) and 5 ml L^{-1} of culture filtrate (2.61 kg) in which the last two treatments

did not differ from the other treatments. In the following season, there was no difference between the treatments, and the plant yield ranged from 1.01 (*A. blazei*) to 2.37 kg per plant (10 ml L^{-1} filtrate).

DISCUSSION

Since 1979, there have been reports on grapevine disease control using strains of antagonistic fungi that are supported by the results presented in this work. For example, Bogdanova et al. (1979) reported that *Fusarium gibbosum* strains that were isolated from grapevine leaves and fruits restrained the disease development when sprayed onto detached leaves before inoculation with *P. viticola* zoospores.

Falk et al. (1996) analyzed from 1992 to 1995, the downy mildew incidence and severity on bunches and leaves of grapevine cv. Chancellor and cv. Lakemont that were treated with a microconidial suspension of *Fusarium proliferatum* G6. The authors observed that for cv. Chancellor in 1992, the grapevines that were treated with the microconidial suspension of *F. proliferatum* G6 showed a 71% reduction in the downy mildew severity. This treatment was not significantly different from the treatment with mancozeb which is similar to the results presented in this work and the various concentrations of the filtrated CUI in both of the evaluated years did not differ from the standard treatments that exhibit control over this disease. However, in relation to the cv. Lakemont, the authors found that the reduction of the incidence of downy mildew in grapevine leaves that were treated with the antagonist suspension decreased from 68.7 to 37% (control 46%) in 1992 and from 40.8 to 20.8% (control 49%) in 1994. The disease severity was reduced by 79 and 67% in 1992 and 1994, respectively.

Previously, Musetti et al. (2006) isolated 126 endophytic fungi from grapevine leaves and tested their activities on *P. viticola* on leaf discs. Of these isolates, only five fungi that were identified as *Alternaria alternata* induced ultra-structural changes in the mycelium of *P. viticola* and restrained sporulation. The most likely hypothesis is that *A. alternata* produces toxic compounds similar to other antagonistic microorganisms.

The results from this study in the control of both downy mildew and isariopsis leaf spot can most likely be explained by the possible occurrence of chlorination as a result of antagonistic interactions between microorganisms through the chloroperoxidase system presented by antagonist microbial interactions in terrestrial ecosystems (Bengtson et al., 2009). In addition, the haloperoxidase system may be involved, such as that of *Curvularia* which has a broad spectrum of action and exerts its lethal effect against bacteria, yeasts and filamentous fungi. It is likely but has not been experimentally verified that the haloperoxidase of *Curvularia* oxidizes halogenates, such as bromide, chloride and iodide, which in the presence of hydrogen peroxide produce reactive oxygen species with antimicrobial properties (Wolfson and Sumner, 1993).

However, one cannot categorically support these explanations, although the *C. inaequalis* filtrate produced some substance(s) that operate in the partial control of the studied diseases. However, it is unknown whether this production is due to a direct action and/or induction of resistance in grapevines. In addition to most of the studies on this subject which are old, few studies were found in the available literature on these antagonistic microorganisms.

Nevertheless, before biological control by any other biocontrol agents can be practically implemented, it is essential to determine how this biological control may be affected by changing environmental conditions. Overall, it is important to learn as much as possible regarding the ecology of these biocontrol organisms and their interactions with the pathogen, host plant and soil and rhizosphere microbial communities and their surrounding environments. Many attempts at biological control have resulted in inconsistent or unsatisfactory disease control under varying environmental conditions and locations (Larkin and Fravel, 2002), as in the case of this study in which the results obtained in the control of isariopsis leaf spot were similar during both years of study; but the AUDPC values were approximately 10 times higher in the first crop season when compared to the second crop season.

This result is mainly due to the favorable conditions for disease development in the second year, making it an unusual year for controlling this disease with a large inoculum pressure of the etiologic agent and conditions of warm and humid weather (high temperatures with an average minimum temperature of 17°C and average maximum temperature of 28°C and a relative humidity of approximately 80.5% mainly in the last month of the experiment during which the disease was evaluated) (Figure 3). Furthermore, the experiment was conducted in the off season during the second year with rainy periods occurring between December and February. This pruning requires more frequent spraying of the vineyard to ensure good control of diseases during the cycle. Therefore, depending on the climate, farmers usually spray every two or three days, but in both experimental years, weekly sprays were performed which may have influenced the increased severity of the disease during the second year compared to the first.

According to Sharma et al. (2009), since 1984 several reports in the related literature have demonstrated the potential of microbial antagonists in the biological control of postharvest diseases of fruits and vegetables. In this sense, some microbial antagonists that have been used for the successful control of postharvest diseases of grapevine include *Aureobasidium pullulans* on grapevine gray mold (*Botrytis cinerea*) (Schena et al., 2003) and soft rot (*Monilinia laxa*) (Barkai-Golan, 2001), as well as

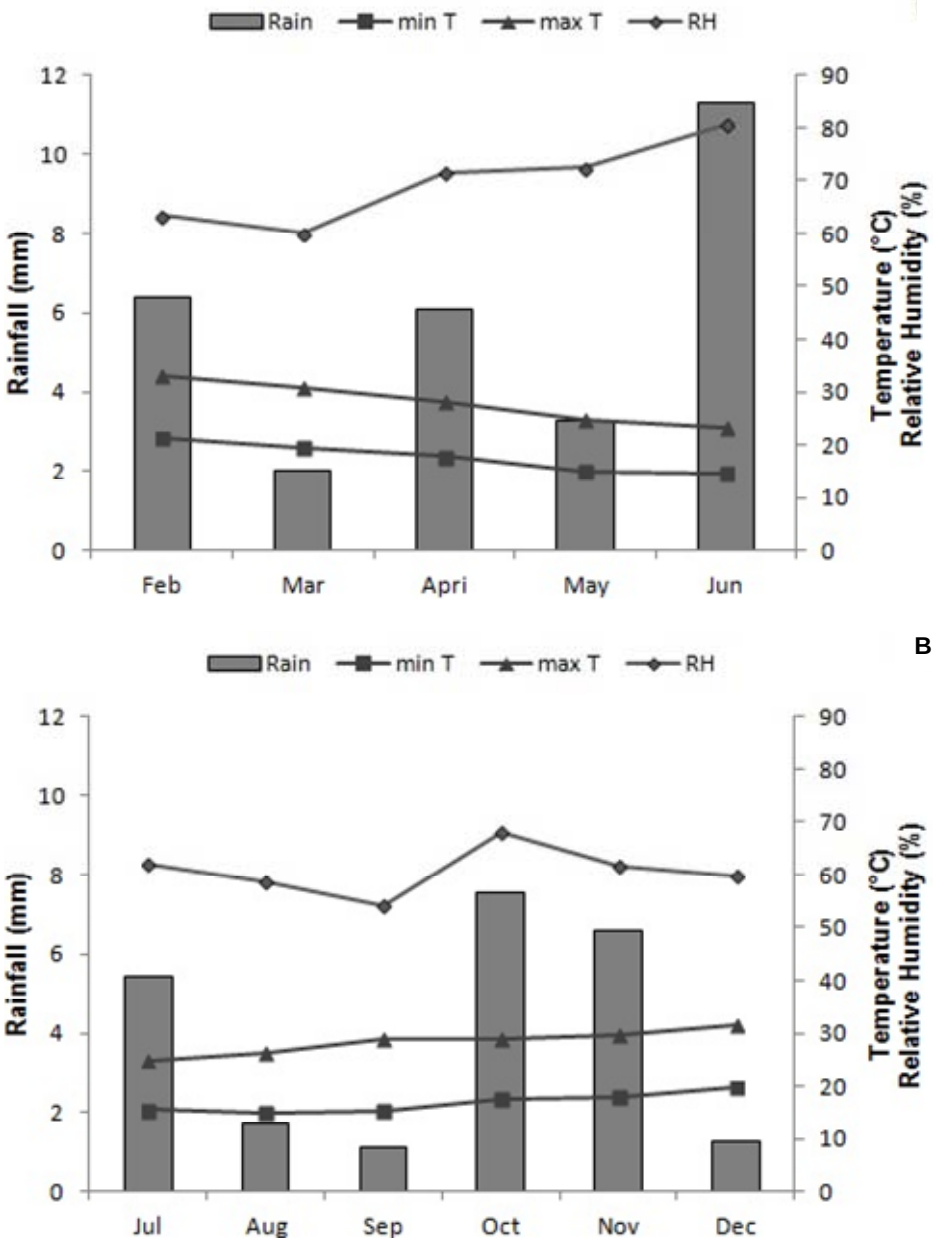

Figure 3. Monthly rainfall (mm), minimum and maximum monthly average temperature (°C) and monthly relative humidity prevailing in the vineyard studied, the periods from July to December 2011 harvest (A) and February to June 2012 harvest (B), in Marialva County, Paraná State, Brazil.

Metschnikowia fructicola (Karabulut and Baykal, 2003) and *Trichoderma harzianum* (Batta, 2007) both on *B. cinerea*.

Cañamás et al. (2011) studied the efficiency of different formulations of *Candida sake* CPA-1 in vineyards of cv. Cabernet Sauvignon on the incidence and severity of *Botrytis cinerea* (gray mold) and observed that in the first year of study, the treatments reduced the incidence of disease between 36 and 40% and that the severity in all of the treatments with *C. sake* was equivalent to the severity with conventional fungicide treatment. The following year, the treatments with *C. sake* cells without heat treatment and that were formulated in liquid solution combined with Fungicover® 5% and the conventional fungicide reduced the incidence of gray mold by up to 90% and the severity from 2.7% to 0.12 and 0.07%, respectively.

Regarding the physicochemical analyses according to Rizzon et al. (2004), the ideal pH range for a good quality grape juice is between 3.1 and 3.3 similar to the values

that were found in this study, although during the second year of study, the pH was more acidic than in the first year. In addition, the use of grapes with a TTA from 0.4 to 0.6%, lower than those values that were obtained in this study is recommended.

Pereira et al. (2008), in analyzing the grape juices of different cultivars, found a total titratable acidity of 0.8% tartaric acid for the cv. Isabel. The acidity in grapes comes from tartaric, malic and citric acids, and this characteristic is related to the edaphic and environmental conditions, especially near the ripening phase in addition to the cultivar and cultivation methods that are used during grape development (Santana et al., 2008) which can explain the high tartaric acid content in the grape juices of this experiment.

Gomes et al. (2011) found that grapevines cv. Isabel that were treated with fungicides showed the lowest proportions of pulp yield differing from the other alternative treatments. The maximum pulp yields that were obtained for each treatment were 72.31% EcoLife® 95 days after pruning (DAP); 69.39% EcoLife® + 71 potassium phosphite 102 DAP; 63.43%, potassium phosphite 102 DAP; 66.67% fungicides 95 DAP; 70.84% Rocksil® 95 DAP; and 81.31%, Agro-Mos® 95 DAP. The results of Gomes et al. (2011) were higher than those that were obtained in this work; however, Di Piero et al. (2005) explain that in some cases, the use of elicitors for the variable weight, bunch weight and berry diameter may prejudice the yield most likely due to the dose and the number of applications which require additional study. In addition, the allocation of plant resources for defense can generate greater energy expenditure.

In the studies of Sato et al. (2008), the average grape bunch weight (cv. Isabel) in northern Paraná State was 125.1 g, and Pereira et al. (2008) averaged 130.55 g, but Kishino et al. (2007) state that the grapevine Isabel produces an average bunch weight of approximately 200 g. According to Grangeiro et al. (2002), the climatic conditions, mainly temperature and luminosity at the time of blooming differentiation may be mainly responsible for the increase in the bunch mass. Furthermore, Neis et al. (2010) concluded that the average productivity of grapevines is higher when pruning is conducted in March and April; these factors could explain the increase in the bunch mass and subsequent yield per plant during the second cycle of this work.

Assis et al. (2011) studied the productive behavior of grapevine cv. Isabel and obtained similar results to those of this study observing a value of 47.4 bunches per plant. Previously, Sato et al. (2008) observed in different rootstocks that the cultivar Isabel had higher number of bunches than those that were obtained by Assis et al. (2011) and in this study in which an average of 76.1 bunches per plant was observed.

The productivity values of this study were lower than those obtained by Sato et al. (2008) which showed a productivity of 9.60 kg per plant (cv. Isabel). Gomes et al. (2011) analyze the effect of resistance inducers on the productivity of grapevine cv. Isabel observed reductions at different levels when the plants were treated with Agro-Mos®, Rocksil®, EcoLife® and EcoLife® + potassium phosphite compared to plants that were treated with fungicides and potassium phosphite, indicating that plants possibly utilize their resources to defend themselves, creating a state of preconditioning that results in an associated adaptive cost, that is, low productivity which may also explain the values that were obtained in this study.

Based on the results of this study, the culture filtrate from the saprobe fungus *C. inaequalis* was efficient in controlling the foliar diseases downy mildew and isariopsis leaf spot, as well as restraining the severity of downy mildew on grape bunches. However, with the weather conditions being extremely favorable for disease development and high inoculum pressure, the efficiency was lower. Postharvest of the grape fruits showed no physicochemical changes regarding the use of this product, but when analyzing the physical features, poor performance was observed which may have been generated by both the experiment development time and climatic conditions as well as by environmental penalties caused by the dose applied and/or the amount of the elicitor applications. Thus, further studies are required in order to clarify issues related to the physical and physicochemical characteristics.

Conflict of Interest

The authors have not declared any conflict of interest.

ACKNOWLEDGMENTS

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Transhumant migration of Baruwal sheep (*Ovis aries*): A case study from the Kanchenjunga conservation Area of Nepal

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The transhumant sheep production is traditional farming system adopted by the herders of the Kanchenjunga Conservation Area (KCA) of Nepal. A rapid field survey by was organized in the KCA of Nepal in order to know the basic of transhumant sheep production in the focus group discussion (FGD), it was learned that the current number of herders and sheep population were sharply declined since the last two decades in KCA. Among the problems identified, conflict of sheep herders due to grazing overlap with the yaks and chauries at high altitude summer pastures, farming systems overlap at low altitude in winter, and the legal conflict between the conservation area management and the sheep herders were prominent. The rapidly growing commercial cultivation of cardamom in suburb of villages at low altitude areas (<2000 m) restricted the sheep migration during winter and, the net benefit gained through transhumant sheep herding was often lower than that of cardamom cultivation. This study would provide the base for future experiments on transhumant sheep production. In general, the scope of transhumant sheep production in view of livelihood of the shepherds were discussed briefly in this case study.

Key words: Transhumance system, grazing route, alpine pasture, Baruwal sheep, conservation area.

INTRODUCTION

The transhumance system is characterized by the seasonal movement of livestock to high altitude alpine pastures for grazing in summer and progressive downward movement and lying at mixed forest areas in winter (Dong et al., 2009a). Among 17 different livestock species reared in Nepal, sheep are one of the important ruminant species for livelihood (Wilson, 1997). Transhumant sheep farming is a traditional practice in the northern high altitudinal regions of Nepal (Joshi et al., 2004). Among the total sheep population available in the country, 60% of them are reared under the transhumance system (LMP, 1993) and Baruwal sheep is one of the important native sheep breeds of Nepal kept in transhumance as a multipurpose small ruminant (Wilson, 1997; Joshi et al., 2004). In KCA, Baruwal sheep are raised in mid hills (2000 to 3000 m) and their crossbreds

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with Kagi, can migrate from 1500 to 4500 m a.s.l, is reared on extensive numbers in transhumance, mostly by the Gurungs and Kirats ethnic groups (KCA, 2004). The sheep herders move their flocks upward in early summer for utilizing the alpine pasture and downward in late autumn/early winter to protect their herds from winter cold weather at high altitude. However, transhumant sheep production is declining in terms of practices, size of flocks reared and overall keeping due to lack of advanced management, and socio-economic factors (Joshi et al., 2004). A study was thus conducted in the KCA in order to get basic information with regard to the transhumant Baruwal sheep production so that the migration system, constraints faced by herders and future scope of transhumance sheep farming in the Kanchenjunga conservation area of Nepal would be possible to learn for research and development.

METHODOLOGY

The following three step procedures were followed with the common principles of rapid rural appraisal (Chambers, 1994) to collect the information on basic of transhumant sheep production in KCA.

Identification of shepherding villages in KCA

The sheep herding communities around the KCA were first identified from the KCA office in Taplejung district of Nepal. The transhumances sheep herding communities of Lelep/Lungthung area which were close to the KCA headquarter at Lelep were selected for this case study. Four flocks were chosen from the herders each two from the Gurung and Kirat ethnicities moving the herds together to high altitude in summer months.

Focus group discussion (FGD)

A checklist was prepared in consultation with the KCA staffs and 3 local key informants with covering history, current practices, challenges and interrelationship of sheep rearing with other farming components from the holistic perspectives. Later, a pre-planned and organized discussion was organized with villagers following the steps of rapid rural appraisal in Lungthung of KCA and five key informants (consultation with the ages and old shepherds) interview was taken focusing transhumance migration, animal management, productivity and current situation of transhumant sheep farming by the local villagers. The participants were the local farmers and former transhumant sheep herders. The FGD was organized intensively with 30 local peoples of Lungthung of KCA and the discussion was set according to the checklist. The discussion with participants was completed in two consecutive days. Information collected during FGD were verified further with key informants' so that reliability of the information would be better established to discuss with the shepherds during field visit (shepherds on pastures).

Field visit

The information collected during FGD and key informants interview were further confirmed with the four shepherds in the field who were migrating the flocks and were available at Mauma pasture at 4000 m a.s.l in KCA. The questions concerning the livelihood and traditional migratory sheep herding were further elaborated together with the shepherds.

Calculation of net benefit from sheep herding

First the revenue resources and the items of cost of shepherding were collected from FGD and further revised in consultation with the shepherds. The net benefit of sheep farming was further calculated from 120 sheep that were jointly herded and from which the total benefit were shared by four herders. The monetary values both for items of revenue and costs were first recorded in local currency altogether for four herders and further expressed to the US dollar. Information was further summarized from four herders having a total of 180 sheep. The net benefit of shepherding was calculated as:

Net benefit = Gross revenue - Gross expenses

RESULTS

The results and explanations in this study were based on the above mentioned criteria described in methodology.

Herd structure

The herd studied was mixed of pure *Baruwal* sheep and their crossbreds with Kagi sheep. It was learned that the number of ewes dominated the flock (83.3%) followed by rams (9.5%) and the least by the lambs (7.2%). However, the number of Baruwal × Kagi crossbreds were in small proportions than the purebred Baruwal sheep (Table 1). Most of the lambs were sold out for daily subsistence of the herder's family.

The crossbreds were produced at the lower altitudes when the Baruwal herd migrated down in the winter according to the respondents. The rams were all intact used for breeding purposes, although they were in small proportions (Table 1). Among the females; around 80% were parturient annually according to the herders. Few lambs (less than 10%) were raised to replace the male and females within the flock, whilst rests of the lambs were sold out for family needs.

Transhumant movement

The general pathway of flock movement has been sketched in Table 2. Usually the upward migration starts at April and the flocks were allowed to graze on 3 to 5 pastures of varying altitudes ranging from 2000 to 4500 m.Then at around mid- August, the flocks used to move to the high elevation alpine vegetation zones in between 4500 to 4700 m. Again at the start of September, flocks move down to the rangeland/pastures of lower altitudes (3500 to 4000 m) of mixed forest vegetation. In the winter

Table 1. Structure of the flock selected for study purpose in Mauma, Taplejung.

Breed type	Rams	Ewes	Lambs	Total
Baruwal	15	140	10	165 (91.7%)
Baruwal × Kagi	2	10	3	15 (8.3%)
Total	17 (9.5%)	150 (83.3%)	13 (7.2%)	180 (100%)

Table 2. Transhumance migration of selected sheep herds in the study site of the KCA as given by the herders (observation for four sheep herds at 4000 m in 2011).

Variables	Transhumance movement pattern			Ecology	
	Altitude	Months	Direction	Pasture type	Competitive species
Lungthung	1700-2000	April-May	Upward	Subtropical	Buffalo, hill cattle, and goats
Se Lele	2600	May-June	Upward	Subtropical-temperate	-
Bakang/Mauma	3600-4100	June-July	Upward	Subalpine-alpine	Yaks and chauries
Langyong	4100-4500	July-August	Upward	Alpine	Yaks and chauries
Thangje	4200-4700	July-August	Top lying	Alpine	Yaks and chauries
Lumbasamba	4500-4700	August-September	Top lying	Alpine	Yaks and chauries
Lamidanda	4000	September	Downward	Alpine	Yaks and chauries
Hile	3500	September-October	Downward	Subalpine-alpine	Yaks and chauries
Lungthung	1700	October-March	Winter stay	Subtropical	Buffalo, hill cattle, and goats

period, the flocks are confined to the summer crop residues, fodder trees and straws. The duration of stay at pastures depends upon the weather; pasture condition and availability and the herd size. At high altitude pastures, the grazing competition basically occurs with yaks and their hybrids with cattle called chauries (*Bos indicus* × *Bos grunnniens* or *Bos grunniens* × *Bos taurus*) (detail in Table 2).

The vegetation remained different according to the altitude and slope and aspect (authors own observation during field visit). In the lower mountains, the mixed meadow is available for sheep grazing. The main palatable pasture species in the altitudes (3500 to 4000 m) were the *Poa* sp*., Kobresia* sp. and *Bistorta* sp. and *Potentilla* sp*.* In the upper zones (>4000 m) mixed meadow of *Kobresia, Festuca,* and *Agrostis* with other plant species were available rather than *Poa*.

Lambing and herd management

Usually the lambing starts during October when the herds arrive from a long march after utilizing the alpine pasture. Around 50% of the sheep lambs. Amongst them 10% lose their respective lambs due to abortion, whilst next 10% had the kid mortality (unknown causes). The high rate of abortion might be due to the long transhumant route in the grass based natural pasture or due to the diet energy constraints. Feeding of high plane of nutrition (concentrates) to the sheep at pregnancy was almost nil at the same time when the vegetation is tended to be lignified in the lower altitudes at the start of winter in the study site.

Shearing of wool

In the traditional practice, wool was sheared for two times in a year that is, first at spring (April) before being at high altitude (>4000 m a.s.l) and second at autumn (<3000 m a.s.l) in September to October at the start of the downward movement (Table 2). The average annual wool production was about one kg and costs nearly NRs.100/kg (1 USD/kg) for raw wool (according to the herders).

Economics of transhumance sheep farming

Instead of decline of transhumant sheep population, the system is depended on the natural vegetation and costs very little to the unit sheep production. Herders usually give only salt occasionally to the sheep besides grazing. The rate of feeding salt is about 70 kg in winter compared to 60 kg during summer for 120 sheep. Concentrate feeding is not established in the traditional transhumant sheep farming. Sheep herding is depended merely on natural seasonal pasture and has been recognized to be the low input system of rearing. The inputs used to rear the sheep in transhumance is very low as compared to the output gained. One mature sheep (20 to 25 kg) in this area costs around NRs 5000 (USD 75), although the

Table 3. Annual expenses for herding of 180 Baruwal sheep at KCA.

[1]Particulars	Total costs	
	[2]NRs.	[3]USD
Expenses for salt (130 kg) and others for sheep	6500	87.00
Food support for 2 herders during grazing period	25000	333.33
Expenses of shelter to the herders (tents, plastic thatch etc.)	4000	53.30
Food for 2 dogs annually	3000	40.00
Annual payment to KCA community forest users groups	500	6.67
Gross expenses	39,000	520

[1]information based on the combined flock of 4 herders having common share of benefits and moving the herds together, [2]information based on the herder's record during the time of interview in 2011, [3] based on exchange rate of 1USD= 75 Nepalese Rupees (NRs.).

Table 4. Annual net income from transhumant herding of 180 Baruwal sheep at KCA.

[1]Particulars	Total income	
	[2]NRs.	[3]USD
Selling of wool 160 kg @NRs100	16000	213.33
Selling of lambs 60 @NRs2000	120000	1600.00
Selling of mature rams @ NRs 5000 (5 rams)	25000	333.33
Occasional selling of ghee and butter and cheese 30 kg @NRS 350	10500	140
Gross income	171500	2286.66

[1]information is based on the combined flock of 4 herders having common share of benefits and moving the herds together, [2]information based on the herder's record during the time of interview in 2011, [3]based on exchange rate of 1USD= 75 Nepalese Rupees (NRs.)

price might vary depending upon male and female and live weight. The net expenses and net return of the herd has been highlighted in Tables 3 and 4.

From Tables 3 and 4, the net benefit was calculated. Accordingly Net benefit= Gross income- Gross expenses= 2286.66-520 = 1766.67 (USD) value. This value gained from sheep farming is 50% net benefit gained from one ropani (500 m^2) land from cardamom in Lungthung area of KCA (general estimation of production of cardamom as of 200 kg from one ropani and current price of the one kg fresh harvested cardamom is 75 USD) according to the respondents.

Constraints of transhumant sheep farming in KCA

A number of constraints/problems have been identified in the FGD. The major constraints of transhumance herding of sheep have been discussed as follows:

Transhumant livestock species overlap

Domestic species overlapped both yak/chauri grazing at high alpine pastures. However, the intensity of grazing competition and its impact on vegetation and productive performance has not yet been available. According to the shepherds, sheep had blamed for spoiling the yak pasture due to frequent urination and gutter, and hoof damage of yaks (*Bos grunniens*) and yak × cattle hybrids called chauries had been reported by the yak herders when yaks travelled closely at the night camps sites of sheep and or during daytime grazing. The exact scientific reason for hoof damage of yaks and chauries due to sheep dung was not known but however it could be due to the bacterial infections.

Disputes between the shepherds and conservation area management

After the declaration of conservation area in 1996 (Parker and Thapa, 2012), few sanctions had been imposed to the transhumant herders. Every year, the sheep herders had to pay NRs. 500 (6.7 USD / herd) to the local community forest users groups (CFUG) for alpine grazing. But there were no sufficient provisions for training of herders on herd management to promote the livelihood of the sheep herders.

Wildlife-domestic animal species overlap

Predation of sheep and their lambs had also been

Table 5. Ranking of major diseases and parasites of sheep by the respondents at KCA.

[1]Rank	[1]Major problem	[2]Altitude	[2]Months
1	Hoof infection	4000-4500 m	July
2	Eye infection and head swelling	>4500 m	August
3	Cough	4000-4500 m	July-September
4	Leach bite and bleeding, plant poisoning	2000-3000 m	April-June

[1]Information collected during the FGD,[2]Information elaborated together with the sheep herders during field visit.

reported by the local people. However, no satisfactory insurance payment system had been arranged by KCA to the shepherds whose sheep were poached by the wild predators (leopards, jackals and foxes). Annual depredation rate was unpredictable but loss of 1-2 animals was unstoppable according to the shepherds. Ikeda (2004) reported the series of depredation the domestic animals by snow leopards in the eastern part of KCA with having no economic damage on households of larger herd size (37 per households) of yaks and chauries, but this might cost to the households of smaller herd sizes of sheep and the intensity of depredation might have also been increased over time.

Farming systems overlap

The lower altitude sites (<2000 m a.s.l) of Kanchenjunga conservation area had been intensified with the commercial high value crop like cardamom and the grazing land has been reduced annually since few decades.

In discussion, the herders replied that cardamom grown at 500 m^2 could yield three times more earning than the herder having 100 sheep in transhumance. The net return of sheep herding from 180 sheep was quiet low to cardamom farming, although the transhumance sheep farming was very labor extensive but observed with less productive (Tables 3 and 4).

Diseases and parasites

Diseases and pests were also found causing significant reduction in sheep production. The herders had ranked the following diseases and pests related to the decline of sheep productivity. In FGD, it was learned that hoof and eye infection was a major disease of sheep during high altitude stay (Table 5). Likewise, the eye infection and head swelling were reported by the herders for crossbreds when above 4500 during alpine period followed by the cough. Leach bite and bleeding were of less importance for high altitude period although happened during the start of upward movement due to progressing hot and humid environment at low altitude grazing period.

Others

When the rainy season started in June, however leaching remained the main problem when starting the upward movement at low altitude sites in between 2000 to 3000 in those days when the weather is wet and humid. Likewise, trapping in stony caves was being the problem in high alpine pastures due to rough and undulated Himalyan pastures.

DISCUSSION

There were no separate indicators available on productive traits of Baruwal sheep in transhumance for KCA. To the author's knowledge, this paper is the first to report on the basic of transhumant sheep production from KCA of Nepal. The information collected in this study supposed to be helpful for future planning of the nature conservation, decentralization of development activities in KCA for improvement of the livelihood of people and also for the sheep based grazing experiments at high altitude conditions.

In Nepal, the history of mountain development has rather a very short history (Barsila, 2008); likewise, the promotion activities for mountain livestock development are still on shadow due to poor institutional infrastructures (Dong et al., 2009b). Although, the livestock system is the sort of livelihood of peoples of mountains of Nepal (Tulachan and Neupane, 1999) where there are no sudden possibilities of other economic interventions and due to very limited arable land holding (CBS, 2001/02). In KCA, the arable land holding is only 2% of the total area (2035 km^2) of KCA coverage (Parker and Thapa, 2012). Baruwal Sheep are the important native transhumant species raised by the mountain peoples of Nepal since the historic period.

The transhumant cycle is associated with the increasing ambient temperature for which the sheep could not tolerate at low altitude during summer. The year round altitude movement of sheep across the altitudinal gradients in the Himalayan Mountains of KCA was longer in duration and wider in altitude comparable to the migration pattern of of yaks and their hybrids in Nepal. Approximated 6 to 8 months of high altitude sheep grazing observed in this study was higher than that of the

transhumant sheep grazing in the French Alps (Gruner et al., 2006). However, it could be hypothesized that high altitude summer grazing competitions and farming system overlap in winter at low altitude of KCA might have resulted the poor performance of sheep, as there were no provisions of winter feeding except than the common salt. Galanopoulos et al. (2011) reported that smallholder transhumant sheep had the technical efficiency low due to smaller herd size as this might also be applied in this study having only 180 sheep by four herders of different families. The annual wool production per sheep and shearing time reported by the respondents in this study were similar to that reported by Joshi et al. (2004) and Wilson (1997) for Nepalese sheep.

The grazing route in the present study site was comparatively long (more than 30 km estimated by authors) and were physically isolated. Unlike in winter crossbreeding with Kagi sheep available at low altitude the inbreeding would result significant declined sheep productivity in the transhumance. The pedigree and the time since when the inbreeding started were unknown in the traditionally raised flocks, as herders lack most of the information due to poor record keeping system. The data on rainfall variation and its relationships on parasitic infection were also not known and for which the separate indicators yet to be built in future experiments.

The diseases and parasitic infections during high altitude grazing reported by the respondents in this study were similar to the report by Joshi et al. (2004) in the western Nepal for the Baruwal sheep. The crossbred sheep would be less tolerant to move on a long grazing route as it might be tedious to them to walk in hypoxic pastures of high altitude and were usually affected by eye swelling due to high altitude sickness (Joshi et al., 2004) and hoof damage during July to August could also be due to the poor mineral content of mountain pastures (Xin et al., 2011). The high altitude pasture sites had been reported covered by more unpalatable forbs than the palatable grasses and sedges in the recent study (Barshila and Devkota, 2013). The eye infection and head swelling were prominent to the crossbreds (Baruwal × Kagi) as compared to the purebred Baruwal sheep could also be viewed as the high altitude intolerance and due to the reason that one of the parents of crossbreds was the native dwellers of low altitude (Kagi sheep), whilst Baruwal sheep are migrated to high altitude and would have better adaptability than Kagi at high altitude. The change in crossbreeding strategy would be an option to improve the high altitude tolerance of crossbreds. The detailed quantitative genetic studies of transhumant sheep performance further needs to be carried out in KCA particularly for Baruwal sheep.

The major constraints of transhumant sheep farming in KCA was learnt due to poor net return from the herding and further exacerbated by legal, biological and social problems as noted in this study. In spite of these constraints the KCA region has a greater scope of transhumant sheep farming due to the availability of different kinds of vegetation due to its wider ecology from 1200 m that is, mixed forest in lower elevation zones to open mixed meadows in high alpine Himalayan Mountains (more than 3000 m). KCA is also well known for trekking route and usually 600 tourists per year visit KCA (Parker and Thapa, 2012) to the world's third highest peak – Mt. Kanchenjunga (Parker and Thapa, 2011).

The transhumant sheep farming still has the opportunities for the valorization of products as Baruwal is raised as a multipurpose animal (Wilson, 1997). Meat and milk products from animals grazing on the high alpine areas are known to have excellent nutritive quality that is, conjugated linoleic acid is higher in milk fat of naturally grazed animals (Pajor et al., 2009). High altitude milk products are also thought useful in prevention of various human diseases including cancer and diabetes (Mcguire and McGuire, 2000; Singhfield et al., 2012). For further promotion of livelihood of the herders, a number of possible interventions could be suggested in short term example, concentrate feeding before and after lambing, fencing and lighting at night, frequent vaccination and introduction of new rams for control of inbreeding etc. Market promotion and crossbreeding as well could be the better alternatives in long term development strategies.

Conflict of Interest

The authors have not declared any conflict of interest.

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Physiological maturity of seeds and colorimetry of yellow passion fruit (*Passiflora edulis* f. *flavicarpa* Degener)

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The passion fruit is among the major fruit crops grown in Brazil, being propagated by seeds due to ease of implementation. However, obtaining seeds of quality is associated with the proper time of collection. Thus, the objective was to determinate the physiological maturity of seeds of passion fruit (*Passiflora edulis* f. *flavicarpa* Degener) using the external color of the fruit. Seeds from passion fruit were collected from four plants. It was adopted a completely randomized design, separating the fruit into four stages of maturation, based on color of the epicarp and quantified as Munsell color chart and refractive index analysis using digital colorimeter. Seeds of completely green fruit did not germinate. Seeds from fruits with advanced stage of maturation tending to wilt, staining with 5 Y 7/10 according to the Munsell color chart and measures of refractive index falling, showed better physiological potential according to the integrity of their membranes, accumulation of dry matter and performance in tests of vigor. This stage was identified as the stage of physiological maturity of the passion fruit.

Key words: Maturation stage, germination, electric conductivity, vigor.

INTRODUCTION

The passion fruit (*Passiflora edulis*) is among the major fruit crops that are grown in Brazil, making the country the world's largest producer, with production of 776,097 tonnes and 59,246 ha planted in 2012, with sales of the fruit in raw or processed for industrial extraction of pulp (IBGE, 2012). Highlighting the yellow passion fruit (*P. edulis f. flavicarpa* Degener) and purple passion fruit (*P. edulis f. edulis* Sims.) due to a greater acceptance and commercial exploitation (Negreiros et al., 2006).

The propagation of these species can be performed sexually, by seed, or asexually, by budding, cuttings or tissue culture *in vitro*. Propagation by seed is widely used

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due to its greater facility of implementation, reduced time to obtain seedlings and the need of hand labor less specialized when compared to asexual methods (Ferreira, 2000).

In order to obtain good quality seeds, there must be considerations about the time of the fruit's collection. It is necessary to follow the development of the fruit and/or seed through their physical and physiological characteristics (Carvalho and Nakagawa, 2012). According to these authors, the maximum quality of the seed is associated with the accumulation of dry matter, also called mass maturity.

Mass maturity occurs close or at the same time of physiological maturity of the seeds, stopping the transfer of plant dry matter to the seeds, reaching maximum physiological potential or quite near of reaching it (Marcos Filho, 2005). According to Carvalho and Nakagawa (2012), seeds that are not completely mature also possess capacity of germination, however, do not generate plants as vigorous as those collected at the point of physiological maturity. Analyzing the process of physiological maturation in seeds of *P. edulis f. flavicarpa* Degener, Negreiros et al. (2006) found that the extraction of seeds should be done on fruits with epicarp color at least 5 to 50% yellow. Before the production of yellow passion fruit's seedlings, it is recommended to keep the fruits for 3 to 6 days in storage before the seed extraction.

However, the visual qualification of fruit's coloration even when based in diagrammatic scale is subjective and with limitation of precision, and when done by softwares demand time and deep training (Effendi et al., 2009). This way, standardized methods with fast execution to determinate physiological maturity can be elaborated. Effendi et al. (2009) and Dranski et al. (2010) presented the use of electronic colorimetry in cultures of jatropha (*Jatropha curcas* L.) to quantify the visual stage of maturity groups. Rodríguez-Pulido et al. (2012) also found a relation between the color of the fruit and the processes of grape maturation, finding reliable correlation between the disposition of phenolic compounds and berry epicarp color, indicating the harvest time to processing.

In general, the propagation of passion fruit by the seeds presents difficulties due to the low percentage of germination and disuniformity on seedling formation (Pádua et al., 2011). Therefore, knowing the aspects that affect seed germination, as those related to pre and post-harvest, morphological and physiological, assist in the solution of several problems related to the seminiferous propagation of the specie for seedling. In addition, stays evident the need of studies that determinate standard conditions and ideal harvest period of yellow passion fruit, to obtainment of seeds with maximum physiological potential, in order to obtain larger uniformity in germination and production of seedlings. Thus, the objective of the present study was to determinate the physiological maturity and ideal time of harvest of yellow passion-fruit's seeds (*P. edulis* f. *flavicarpa* Degener), using the extern coloration of the fruit through digital colorimeter as the indicator of maturity.

MATERIALS AND METHODS

The experiment was conduced in the Seed and Seedling Laboratory and Plant Physiology Laboratory, both located in State University of West Paraná – Unioeste, Campus Marechal Cândido Rondon, in 2013. The seeds were collected from fruits of yellow passion-fruit, specie *P. edulis* f. *flavicarpa* Degener, that came from four plants in rural propriety, situated in 24°33'20,68"S and 54° 02'36,32"W, in different stages of maturity, collected in one single moment. The experiment was conducted in entirely casualized lineation with four treatments. In the harvest process, occurred the separation of the fruits in four visual states of maturation (Figure 1). For each stage, were used five fruits, totalizing 20 experimental plots. For the segregation of the stages of maturity, were applied Munsell's color chart (Munsell, 1976) and analysis of the refractive index of the epicarp in the red, green and blue spectrum, with the assistance of digital electronic colorimeter (Instrutherm® ACR 1023). The values were obtained from three measurements in each fruit, taken at equidistant points in the equatorial region. After classification, the fruits were opened, with the aid of a scalpel, in the equatorial region of fruit and, supported by a spatula, had its seeds extracted along with the placental membranes.

In relation to the seeds, as stated in Figure 1, in the first stage of maturation of the fruits, the absence of yellow color was observed, as well as thin integument, without formed pulp, indicating the immature stage of the fruit. On stage two, the seeds presented in transition phase, with seeds both dark and yellowish, and has been verified the presence of characteristic pulp, although in low quantity. On stages three and four, all the seeds presented dark coloration of the integument and the fruits had high quantity of pulp. On stage four, the fruits presented external wilt. The seeds obtained from the fruits were washed in running water, on steel sieve, to remove the aryl, as described by Carvalho et al. (2012). Then, were accommodated in trays above paper towel and under shade for four hours. After processing the seeds, the electrical conductivity, dry matter, moisture content and seed vigor and germination were determined.

The electrical conductivity test occurred in five replicates of 25 seeds, weighed on an analytical balance and placed in polypropylene cups of 200 ml. The seeds were immersed in a volume of 50 ml, with distilled and deionized water, and kept at constant temperature of 25°C, for 24 h. With the assistance of a bench conductivity meter (Engineering® BEL W120), and according to the methodology of Vieira and Krzyzanowski (1999), the reading of conductivity were performed, and the results were expressed in $\mu S\ cm^{-1}\ g^{-1}$. Five samples of 100 seeds were used to quantify the mass of dry matter and moisture content of seed for each group, quantifying the fresh mass of the samples, placed in kraft paper bags bags properly identified and taken to forced circulation air oven, with a temperature of 105 ± 3°C for 24 h. After this period, the samples were weighed again on analytical balance, measuring the dry mass of the sample. Simultaneously, it was determined the moisture content of seeds based on the mass of wet material, as described by Marcos Filho (2005), consisting on the ratio between the mass of water present in the seeds (Wm -Dm) and dry mass (Dm) of the sample, expressed in percentage.

The germination test was conducted under vermiculite substrate, accommodated in plastic polypropylene trays. The trays were placed in a germinator, type Biochemical Oxygen Demand (BOD), set at 25°C, as recommended by Brasil (2009), photoperiod 12:12 (light: dark). Were used five replicates of 20 seeds for each group,

Stage of maturation	Coloration	Visual characterization	Munsell's chart
1		Green fruit	7,5 GY 4/4
2		Fruit in pigmentation transition green/yellow	2,5 GY 6/8
3		Predominantly yellow fruit	5 Y 8/6
4		Yellowish fruit starting wilt	5 Y 7/10

Figure 1. Coloration of yellow passion fruit [*Passiflora edulis* f. *flavicarpa* (Degener)] in different stages of maturation. Marechal Cândido Rondon/PR, 2013.

separated by the color of the fruit. The count of normal emerged seedlings occurred daily from the test installation and finished at 50 days from sowing. With data from seedling emergence, determined the number of days until the start of emergence (EI), according to Labouriau (1983). Also, computed the Emergence Speed Index (ESI) proposed by Maguire (1962). After 50 days, it was determined the Percentage of Germination (PG) by counting the emerging seedlings.

Using the methodology proposed by Czabator (1962), it was stipulated the Peak Value of Germination (PV), corresponding to the ratio between the maximum value of the germination accumulated percentage and the number of days to obtain it. It was also determined the Daily Germination Average (DGA), which expresses the ratio between the total germinated seeds and the time (in days) of the duration of the test, and the Germinal Value (GV), obtained by the product between PV and DGA.

The data were subjected to analysis of variance. The treatment means were compared by Tukey's test at 5% probability of error, with the aid of SISVAR version 5.1 (Ferreira, 2011). Based on the average of nine characteristics: electrical conductivity, dry matter weight of 100 seeds, moisture content, EI, ESI, PG, PV, GV and DGA, in each maturation stage, the dissimilarity matrix was calculated using the Euclidean mean distance standardized, by the method of UPGMA (unweighted pair group method with arithmetic mean). Next, applied to the analysis of hierarchical agglomerative clustering (HAC) concocting dendrogram with the aid of χLstat program®.

RESULTS AND DISCUSSION

According to the data shown in Table 1, there was a decrease in the electrical conductivity, accompanying the ripening of the fruits, of 51.61, 75.76 and 83.25%, comparing stage one to two, three and four, respectively. Thus, according to the advance in development and fruit ripening process, there is greater organization of the seed's membranes, and therefore less loss of electrolytes through these (Araújo et al., 2007), resulting in increased seed vigor when compared to seeds that have high electrical conductivity.

Studying the correlation between electrical conductivity test and vigor and seed viability tests of yellow passion fruit, Barbosa et al. (2012) verified that the conductivity test is effective to segregate seed lots at different levels of vigor, when correlated with seedling emergence. Evaluating different stages of maturation and storage conditions of the yellow passion fruit, Araújo et al. (2007) also observed that the seeds that presented lower values for the electrical conductivity showed highest germination, recommended parameter for the determination of seed vigor of this specie.

In the analysis of dry matter weight of 100 seeds, were also found differences between the stages of fruit maturation. Stage three of maturation showed higher absolute dry matter accumulation (0.486 g), however, being statistically similar to stage four (0.466 g). Stage one had the lowest values (0.266 g) differing significantly of the others, while stage two presented an intermediate value (0.394 g), also showing significant differences. This increase in dry matter accumulation may have correlation with seed vigor, since the greater the accumulation of reserves, the better the intrinsic ability to properly develop (Marcos Filho, 2005).

Table 1. Electrical conductivity, dry matter weight of 100 seeds and moisture content calculated on a wet basis of seeds of yellow passionfruit [*Passiflora edulis* f. *flavicarpa* (Degener)] at different stages of fruit ripening. Marechal Cândido Rondon/PR, 2013.

Stage of maturation[(1)]	Electrical conductivity (μS cm^{-1} g^{-1})	Dry matter weight of 100 seeds (g)	Moisture content (%)
1	388.898[a]	0.226[c]	36.03[a]
2	188.156[b]	0.394[b]	24.16[b]
3	94.265[c]	0.486[a]	26.04[b]
4	65.136[d]	0.466[a]	16.72[c]
Mean	184.114	0.393	25.74
CV (%)	9.55	3.32	10.15

Different letters in the column indicate significant differences by Tukey test at 5% probability of error. (1) 1: Green Fruit; 2: Fruit in pigmentation transition green/yellow; 3: predominantly yellowish fruit; 4: yellowish fruit starting wilt.

Table 2. Days until the start of emergence (EI), emergence speed index (ESI), percentage of germination (PG), peak value (PV), daily germination average (DGA) and germinative value (GV) of seeds of yellow passion fruit [*Passiflora edulis* f. *flavicarpa* (Degener)] at different stages maturation. Marechal Cândido Rondon /PR, 2013.

Stage of maturation [(1)]	EI	ESI	PG (%)	PV	DGA	GV
1	50.00[a]	0.000[c]	0.00[c]	0.000[b]	0.00[c]	0.000[b]
2	39.00[b]	0.593[b]	25.50[ab]	0.054[a]	5.10[ab]	0.289[ab]
3	40.80[b]	0.441[b]	20.00[b]	0.048[a]	4.00[b]	0.249[ab]
4	22.60[c]	1.312[a]	37.50[a]	0.078[a]	7.50[a]	0.611[a]
Mean	37.95	0.668	20.75	0.045	4.15	0.287
CV (%)	4.91	12.44	18.93	1.58	16.66	11.81

Different letters in the column indicate significant differences by Tukey test at 5% probability of error. (1) 1: Green Fruit; 2: Fruit in pigmentation transition green/yellow; 3: predominantly yellowish fruit; 4: yellowish fruit starting wilt.

Similarly, Araújo et al. (2007) find no significant differences in dry matter accumulation in seeds derived from passion fruit harvested at intervals of 10 days, at 55, 60 and 65 days after anthesis (DAA). Also stated that treatment with higher average of dry matter accumulation showed a low percentage of germination when subjected to the accelerated aging test, indicating that larger seeds are more subject to degradation. The same authors also found that the decrease of the water content of seeds occurs at 55 and 60 DAA.

The moisture content of the seeds suffered a decrease, following the trend of electrical conductivity, due to the organization of the membranes during the process of seed and fruit maturation (Table 1). The stage four showed lower moisture percentage (16.72%) when compared to others; the second (26.04%) and third (24.16%) showed no significant differences, showing lower moisture content when compared to stage one (36.03%). These results indicate the superiority of the group four when compared to the other stages, showing greater amount of reserves in relation to water, being beneficial to the storage, germination and development process of the seed. According to Padua et al. (2011), seeds of *Passiflora setacea* are resistant to desiccation in levels close to 4% water, and low humidities and storage temperatures induce dormancy in seeds of this species. Fonseca and Silva (2005) found that 7% of moisture reveled favoring the maintenance of the physiological potential of *P. edulis* f. *flavicarpa* Degener in between 31 and 7% moisture in seeds.

In Table 2, it was observed a difference between the phases of maturation to EI, in stage four showed shorter period for early emergence (22.60 days), while the first stage showed no seed emerged during the study period. Stages two and three showed an intermediary behavior when compared to similar treatments and to each other, 39 and 40.80 days, respectively. These responses follow the progress of the maturation process by reducing the number of days for the beginning of germination according with the course of the senescence process of the fruit. It can be observed that stage four also showed better performance for ISE and PG variables, resulting in the stage with greater speed emergency (1.312) and higher germination (37.50%). These results complement those obtained previously in the electrical conductivity and dry matter tests, giving these answers to a greater integrity of their membranes and higher accumulation of dry matter of stage four, providing better conditions for germination and survival of seeds. Highest germination of seeds of passion fruit are found in seeds from shriveled fruit (Lopes et al., 2007). Similarly, seeds derived from fruits harvested at 65 DAA showed higher germination percentage compared to fruit harvested at 55 and 60 DAA (Araújo et al., 2007).

Table 3. Refractive index mean values of the fruits of yellow passion fruit [*Passiflora edulis* f. *flavicarpa* (Degener)], obtained through digital colorimeter and qualitative characterization of the Munsell chart. Marechal Cândido Rondon/PR, 2013.

Stage of maturation(1)	Refractive Index Scale (nm)			Munsell
	Red	Green	Blue	
1	170.133[d]	157.400[c]	98.866[b]	7.5 GY 4/4
2	238.933[c]	206.000[b]	113.000[b]	2.5 GY 6/8
3	491.933[a]	346.600[a]	192.333[a]	5 Y 8/6
4	386.066[b]	246.600[b]	116.533[b]	5 Y 7/10
Mean	321.766	239.150	130.183	-
CV (%)	9.81	9.54	14.27	-

Different letters in the column indicate significant differences by Tukey test at 5% probability of error. (1) 1: Green Fruit; 2: Fruit in pigmentation transition green/yellow; 3: predominantly yellowish fruit; 4: yellowish fruit starting wilt.

Stages two and three presented similar results and intermediary among treatments for the ESI and PG variables. Corroborating with the results found, Negreiros et al. (2006) did not find significant differences in the ESI in three stages of maturation (unripe fruit starting to yellow, 5 to 50% yellowish and more than 50% yellowing) of yellow passion fruit. So when contrasted his description of stages of maturation with the present work, the green fruit starting to yellow equates to stage two, while the group with more than 50% yellowish equivalent to stage three. For the PV variable in Table 2, stages two (0.054), three (0.048) and four (0.078) were superior to stage one. For the analysis of DGA, the early stages showed lower values compared to the others, again being the fourth stage (7.50) the one that was successful for the variable, equivalent to stage two (5.10) and differing from stage three (4.00). Finally, to the GV test, only the fourth stage differed from the first, being equivalent to the others. These data are derived from mathematical relations that utilize directly the germination percentage and the test duration in days as the basis of calculation, being variables used to express the vigor, speed and uniformity of germination (Czabator, 1962), having high correlation and similarities between them (Santos et al., 2009). There is the possibility of pointing stage four as the best point to harvest the fruits for seed production.

Refractive index measures made with digital colorimeter showed differences between the maturation phases of passion fruit for the three measurements made (Red, Green and Blue) as shown in Table 3. The readings showed increase in values following the maturation, reaching its highest values in stage three of maturation (491.933, 346.600 and 192.333 for red, green and blue, respectively), and coinciding with the scale 5 Y 8/6 of the Munsell chart (Munsell, 1976). After this stage, there was a decrease in the three evaluated colors.

Several studies have succeeded in relating the coloring, either of the fruit or seed, as an indicator of physiological maturity of seeds from many different cultures. Ragagnin et al. (1994) in seeds of maritime pine (*Podocarpus lambertii* Klotzsch), Gemaque et al. (2002) in seeds of purple ipe (*Tabebuia impetiginosa* (Mart.) Standl.), Fonseca et al. (2005) with seeds of yellow ipe (*Tabebuia chrysotricha* (Mart. Ex DC.) Standl.) and Mendes et al. (2005) studied the process of maturation in annatto (*Bixa orellana* L).

However, evaluations of tone require trained personnel and are mostly subjective, causing a reduction in the accuracy on practical application. Thus, it denotes the importance of a standardized method such as digital colorimeter, exemplified in studies conducted with jatropha (*Jatropha curcas* L.). Effendi et al. (2009), who developed a classification system to indicate the level of ripeness of the fruit system, helping to define the ideal harvest point for production of oil crop. Likewise, Dranski et al. (2010) related the epicarp color of the fruit to the physiological maturation of the seeds of Jatropha. The use of colorimetry is also applied to other cultures and with different purposes, as in grape (*Vitis vinifera* L. cv. Graciano), where Rodríguez-Pulido et al. (2012) related the epicarp color to the contents of phenolic compounds in the culture, making colorimetry an alternative, fast, and inexpensive tool for indicating the point of harvest for wine production. The passion fruit seeds presents greater germination power and dry matter accumulation when the fruit is fully yellowish (Negreiros et al., 2006), following the highest values of refractive index obtained in the study (Table 3). Thus, the use of digital colorimeter shown as an excellent tool to help with the identification of suitable harvest time, in order to obtain seeds for propagation purposes.

Finally, it is observed by the dendrogram (Figure 2) the formation of three distinct groups. It can be identified similarity between the stages three and four of passion fruit maturation, which although differ in most vigor tests, are grouped due to proximity to the mass of dry matter accumulated in the seeds and the integrity of their membranes (electrical conductivity). Stage two showed intermediary dissimilarity, while stage one, presented farthest of the other stages of maturation. Thus, stage four, followed by three, presented better characteristics to

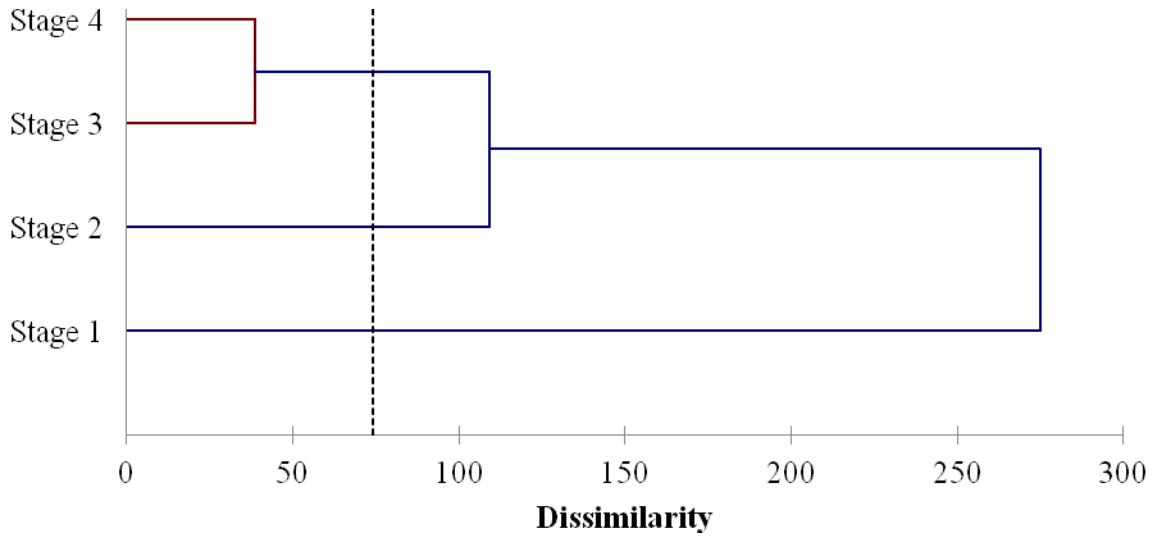

Figure 2. Dendogram made using cluster analysis of stages of maturation of yellow passion fruit based on the standardized Euclidean mean distances matrix calculated by the method of UPGMA (unweighted pair group method with arithmetic mean), using nine characteristics related to seed vigor and viability.

form vigorous plants and performance in vigor and seed viability tests, while stage two showed intermediate behavior, being stage one smaller than the others due to the inability of the seeds to produce viable seedlings.

Conclusions

Seeds of yellow passion fruit (*P. edulis* f. *flavicarpa* Degener) obtained from yellowish fruits tending to wilt (stage four) showed better performances in tests of vigor, having higher speed, uniformity and emergence percentage. In the moment that refractive index values reach its peak (491.933, 346.600 and 192.333 nm for the red spectrum, green and blue, respectively) and go into decline, coincides with the higher physiological potential of the seeds. It is accompanied by the maximum accumulation of dry matter, lower water content and higher membrane integrity, indicating that the refractive index of the fruit's epicarp is a valid and easy method to use for determining the physiological maturity of seeds of passion fruit.

Conflict of Interest

The authors have not declared any conflict of interest.

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Full Length Research Paper

Herbaspirillum seropedicae inoculation and nitrogen fertilization on nitrogen use efficiency of different corn genotypes

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The biological N fixation provided by diazotrophic bacteria may be an alternative for corn production in more sustainable or low-input agricultural systems. Therefore, this study aimed to evaluate the interactive effect of seed inoculation with *Herbaspirillum seropedicae* and nitrogen fertilization on the nitrogen use efficiency of different corn genotypes, through the technique of isotopic dilution of ^{15}N. The experiment was carried out under controlled conditions in a greenhouse using a completely randomized design in a 3 × 2 × 2 factorial scheme with three corn hybrids: Maximus, P3646H and BRS3035, using inoculated and non-inoculated plants with two nitrogen doses (0.0 and 80 kg ha^{-1}) and four replicates. At 35 days after emergence plants were collected, divided into root and shoots and analyzed with regard to root and shoot dry matter production, accumulated N in plant, N percentage and content coming from the fertilizer and use efficiency of N applied as a fertilizer. Results showed that there is a difference between corn genotypes concerning shoot dry matter production and percentage of N shoot coming from the fertilizer. Shoot dry matter production is influenced by *H. seropedicae* inoculation. This inoculation coupled with nitrogen fertilization promotes shoot N increments of about 32 and 62% for the hybrids P3646H and BRS3035, respectively. These hybrids showed increases of 34.3 and 64.4%, respectively, in N use efficiency when inoculated with *H. seropedicae* without the addition of N, what allows inferring that the BNF behaves as an important source of N to the system.

Key words: *Zea mays* L., diazotrophic bacteria, ^{15}N, nitrogen fertilization.

INTRODUCTION

Corn is a crop that demands soil fertility especially nitrogen (N) whose deficiency may reduce grain yield from 10 to 22% (Subedi and Ma, 2009). Thus, inadequate management of nitrogen fertilization is still one of the obstacles for the increase in productivity. Currently nitrogen fertilizers represent more than 70% of corn

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fertilization which greatly influences the economic viability of the crop (Machado et al., 1998). The obtention of cultivars adapted to low-N soils and capable of associating with diazotrophic bacteria might represent an economically viable alternative for corn production in agricultural systems with low supply of inputs (Reis Junior et al., 2008).

The use of relatively high nitrogen doses during the growth stage of hybrid corn cultivars may lead to the selection of genotypes with low luxury consumption of this nutrient and/or requiring high nitrogen fertilization to express their productive potential (Carlone and Russel, 1987). On the other hand, low nitrogen doses may naturally contribute to the selection of genotypes efficient at associating with nitrogen-fixing bacteria which can provide reduction in the consumption of synthetic nitrogen fertilizers (Roesch et al., 2005) and selection and/or development of cultivars with higher N use efficiency (Carvalho et al., 2012).

The obtention of higher efficiency in N use has been the goal of both capitalized and low-input agriculture. This occurs due to the waste and scarcity of nitrogen which can cause economic, environmental, public health and food safety problems (Carvalho et al., 2012). However, the ways to obtain genotypes that are more efficient in N use are rather complex because nitrogen metabolism is influenced by various environmental factors (Hirel et al., 2001; Majerowicz et al., 2002).

The positive interaction between diazotrophic bacteria and corn has been reported by many authors (Dobbelaere et al., 2001) and a survey of various experiments carried out in many countries shows that the inoculation with bacteria from the genus *Herbaspirillum* resulted in most cases in increase of dry matter, productivity and nitrogen accumulation (Reis, 2007). Therefore, considering that the genetic range can influence the interaction between corn plants and endophytic diazotrophic bacteria, this study aimed to evaluate the interactive effect of seed inoculation with *Herbaspirillum seropedicae* and nitrogen fertilization on the N use efficiency of different corn genotypes determined through the ^{15}N-isotope dilution technique.

MATERIALS AND METHODS

The experiment was carried out in a greenhouse (22°12' S; 54°56'W; 452 m) at the College of Agricultural Sciences of the Federal University of Grande Dourados, MS, Brazil, from December 2012 to January 2013. The climate in the region is Cwa according to Köppen's classification. The soils used in this study classified as Distroferric Red Latosol with very clayey texture (Embrapa, 2013) was collected in the layer of 0 to 20 cm. The chemical analysis of the soil before the experiment is as follows: pH ($CaCl_2$): 4.15; P: 26 mg dm^{-3}; K: 5.0 mmolc dm^{-3}; Ca: 9.0 mmolc dm^{-3}; Mg 2.0 mmolc dm^{-3}; Al: 3.3 mmolc dm^{-3}; H+Al: 41.6 mmolc dm^{-3}; Sum of Bases: 115.1 mmolc dm^{-3}; CEC: 531.1 mmolc dm^{-3} and Base Saturation: 21.7%. Granulometric analysis showed 225 g kg^{-1} of sand, 125 g kg^{-1} of silt and 650 g kg^{-1} of clay.

Soil pH correction was performed 30 days before sowing in order To increase base saturation to 50% using finely ground dolomitic limestone (RNV 100%) considering the results of soil chemical analysis. Due to the low soil fertility, base fertilization was also performed in order to guarantee the establishment of the crop. 100 kg ha^{-1} of P_2O_5 (270 mg dm^{-3}) and 60 kg ha^{-1} of K_2O (51 mg dm^{-3}) were applied (mixed to the soil) in the forms of single super phosphate and potassium chloride, respectively. Micronutrients were applied according to the crop demand in solution using deionized water and salts (p.a.) according to Epstein and Bloom (2006). Nitrogen fertilization was performed using 80 kg ha^{-1} of N (54 mg dm^{-3}) as urea (45%) is divided into two applications of 40 kg ha^{-1} of N (27 mg dm^{-3}). The first one was applied at sowing and the second in covering 15 days after plant emergence. The isotopic enrichment used was equal to 0.7% of ^{15}N atoms in excess for the dose of 80 kg ha^{-1}. In order to guarantee a uniform application, the nitrogen fertilizer was diluted in 50 mL of deionized water and applied using a pipet.

The adopted experimental design was completely randomized in a 3 × 2 × 2 factorial scheme with three corn hybrids: Maximus, P3646H and BRS3035 using inoculated and non-inoculated plants; with two nitrogen doses (0.0 and 80 kg ha^{-1}) and four replicates. The experimental units consisted of 10-dm^{-3} plastic pots daily irrigated with deionized water in a controlled way to replenish the water lost through evapotranspiration in order to maintain soils at 60% of the field capacity.

Corn seeds of the simple hybrid P3646H (Pioneer), double hybrid Maximus (Syngenta) and triple hybrid BRS3035 (Embrapa) were used in the sowing and previously inoculated with the Z-94 strain of *H. seropedicae* (inoculant cell concentration of about 10^9) in the peat-based formulation produced by Embrapa Agrobiology, Seropédica - RJ. The applied dose was 250 g of peat inoculant for each 10 kg of corn seeds. For inoculation, 60 mL of a sugary solution at 10% (m/v) were added for each 10 kg of seeds, aiming to increase the adhesion of the peat inoculant to the seeds. Seeds were left to germinate directly in the pots and eight days after emergence thinning was performed leaving only one plant per experimental unit.

At 35 days after emergence, plants were collected and divided into root and shoot. All collected material was sequentially washed in running water, 0.1 mol L^{-1} HCl solution and deionized water. Then, samples were accommodated in paper bags and dried in a forced-air oven at 65°C for 72 h. After drying the material, dry matter was weighed and ground in a Wiley-type grinder for analyses of N total content and N isotopic composition.

Total N in different parts of the plant (root and shoot) was determined through the Kjeldahl method according to the methodology described in Embrapa (2009). As for the ^{15}N isotopic composition analyses, samples were processed according to the method of Rittenberg (1946); from the final distillate obtained in the analysis of % of Total N, the extracts were again acidified with 0.5 mol L^{-1} H_2SO_4 and concentrated through evaporation; $N\text{-}NH_4^+$ was converted into N_2 through oxidation with lithium hypobromite (LiOBr) (Porter and O'Deen, 1977). The analyses of the ^{15}N isotopic composition were performed using a mass spectrophotometer Delta Plus from the John M. Day Stable Isotope Laboratory at Embrapa Agrobiology. With the results of nitrogen isotopic composition (% in ^{15}N atoms) of the samples was calculated as:

a) Total N content accumulated in the plant (TN, mg/plant)

$$TN = \frac{DMY \times N}{100}$$

Where DMY is the dry matter yield and N is the N content in the plant (g kg^{-1}).

b) Percentage of N in the plant coming from the fertilizer (%PNF)

$$\%PNF = \left(\frac{\%\text{ in 15N atoms excess plant test}}{\%\text{ in 15N excess fertilizer}}\right) \times 100$$

Table 1. Analysis of variance of shoot dry matter (SDM), root dry matter (RDM), shoot nitrogen percentage (%SN) and total shoot nitrogen (TSN) of three corn hybrids subjected to different nitrogen levels and inoculated with *Herbaspirillum seropedicae*. Dourados, MS, Brazil (2013).

Source of variation	DF	Mean Square			
		SDM	RDM	%SN	TSN
Hybrid (H)	2	51.30*	3.94	0.58*	8357.16*
Nitrogen (N)	1	224.42*	41.45*	8.50*	193080.79*
Inoculation (I)	1	33.41*	12.19	0.01	11041.70*
H*N	2	7.23	0.48	0.07	6130.75*
H*I	2	2.55	12.53	0.32*	2703.31
N*I	1	4.30	3.49	0.15*	1692.00
H*N*I	2	0.77	0.72	0.41*	5262.89*
Residue	36	3.33	5.01	0.02	916.63
CV (%)		24.07	9.18	11.32	25.95

* – significant by Tukey test to 5% probability. CV- coefficient of variation.

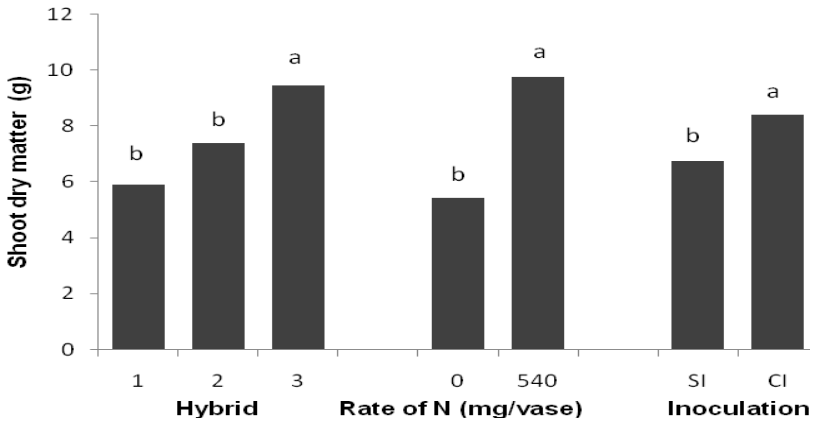

Figure 1. Shoot dry matter (SDM) of three corn genotypes inoculated or not with *Herbaspirillum seropedicae* under different nitrogen levels. Hybrid 1 (Maximus), Hybrid 2 (P3646H) and Hybrid 3 (BRS 3035); NI (no inoculation); WI (with inoculation).

c) N in the plant coming from the fertilizer (PNF)

$$\mathrm{PNF} = \frac{\%\mathrm{PNF}\ \mathrm{x}\ \mathrm{DMY}}{100}$$

d) Use efficiency of N applied as a fertilizer as a function of the applied quantity (NAQ)

$$\mathrm{NUE} = \frac{\mathrm{PNF}}{\mathrm{NAQ}} \mathrm{x}\ 100$$

The obtained results were subjected to the analysis of variance and the means were compared by Tukey test at 5% of probability using the statistical software Sisvar (Ferreira, 2000).

RESULTS AND DISCUSSION

Significant negative effects (p≤0.05) were observed in the interaction hybrid x inoculation x nitrogen for shoot N percentage (%SN) and total shoot N (TSN). Shoot and root dry matters (SDM and RDM) did not show significant effect of the interaction and are independently shown for each hybrid, inoculation and nitrogen in Table 1.

The corn hybrids significantly differed (p≤0.05) with respect to SDM (Table 1) which was 60.5% higher for the BRS 3035 compared to Maximus and 28.3% compared to P3646H (Figure 1). In 2008, Reis Junior and coworkers also observed difference in dry matter accumulation between the studied corn hybrids.

In this study, nitrogen fertilization promoted increase of 79.5% in shoot dry matter production of corn plants compared to the control treatment which was not fertilized (Figure 1). Similar results were found by Gava et al. (2010); these authors found that the increase in the dose of N fertilizer caused increase of dry matter and dry matter production rate in corn. Root dry matter in the fertilized treatment was 7.92% higher than in the non-

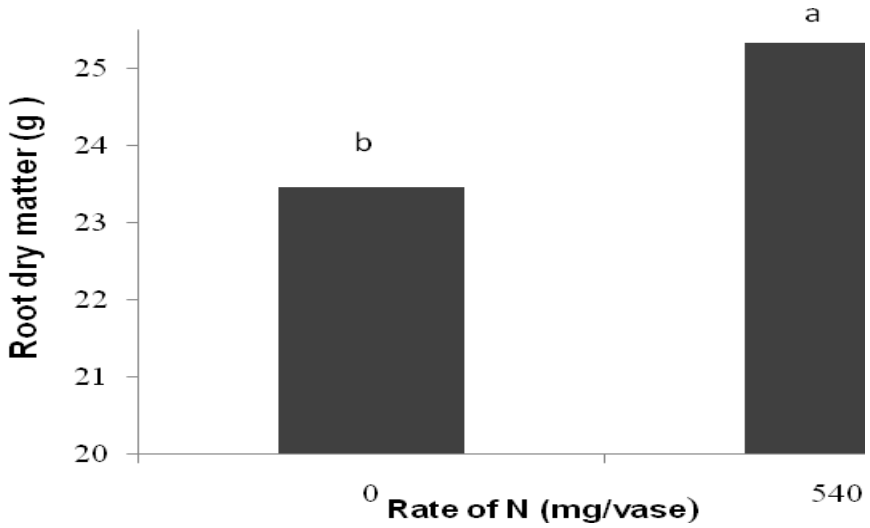

Figure 2. Root dry matter (RDM) as a function of nitrogen fertilization.

fertilized (Figure 2) corroborating the results found by Taylor and Arkin (1981). Glass (1990) reported change in root growth as a result of soil fertility. Moreover, nitrogen favors root system growth providing plants with the conditions for greater water and nutrient absorption (Rao et al., 1992).

It is observed that the inoculation with the Z-94 strain of *H. seropedicae* promoted an increase of 24.74% in the SDM production compared to the control. The values ranged from 6.75 g/plant in the control to 8.52 g/plant in the inoculated treatment (Figure 1). This greater SDM production in inoculated plants may have been favored by the production of growth-promoting substances by bacteria. Ferreira et al. (2011), Ferreira et al. (2010) and Guimarães et al. (2010) also observed significant effect of the inoculation with *H. seropedicae* on the SDM accumulation of rice plants.

For %SN of corn plants, there was significant interaction between corn genotypes, seed inoculation with *H. seropedicae* and nitrogen doses (Table 2). There was no significant difference between corn genotypes without inoculation and nitrogen fertilization (Table 2). In the presence of inoculation with *H. seropedicae* and no nitrogen fertilization, there was lower %SN in the hybrid BRS 3035 compared to the others (Table 2). In the study of Sala et al. (2007), for one of the two tested wheat genotypes without the addition of N fertilizer, plants not inoculated with diazotrophic bacteria had higher N accumulation than those in treatments with inoculation. Araujo et al. (2013) reported that, in the absence of nitrogen fertilizer, inoculation may reduce shoot dry matter of plants. This negative effect may also occur as a result of an increase of the root system at the expense of the shoot development. An explanation for this is that one of the benefits provided by diazotrophic bacteria for grass crops is the capacity to produce phytohormones (Dobbelaere et al., 2001, 2002; Mendonça et al., 2006; Sala et al., 2007).

In the presence of nitrogen fertilization and inoculation with *H. seropedicae*, increases of 26.8% and 36.2% in %SN of the hybrids P3646H and BRS 3035 was verified, respectively, when fertilized and inoculated with *H. seropedicae* (Table 2). However, opposite results were obtained for the Maximus hybrid for both %SN and TSN (Table 2).

The TSN did not differ statistically between corn hybrids for the treatment without nitrogen fertilization with and without inoculation. Nevertheless, in the presence of nitrogen fertilization, the inoculation with *H. seropedicae* caused an increase of approximately 32% in the TSN of P3646H and 62% of BRS 3035 compared to the treatment with fertilization and without inoculation with *H. seropedicae* (Table 2). Some reports reinforce that variable responses of inoculation with endophytic diazotrophic bacteria are common in grass crops which has justified investments to improve this technology (Sala et al., 2007). Even in controlled environments as in the greenhouse, discrepant responses of the association between diazotrophic bacteria and corn cultivars have been frequent which has probably limited the consolidation of commercial inoculants (Dobbelaere et al., 2002).

The percentage of nitrogen in the shoot coming from the fertilizer (%SNF) differed only between corn hybrids while the shoot nitrogen content coming from the fertilizer (SNF) and shoot N use efficiency (SNUE) were influenced by the interaction hybrid x inoculation with *H. seropedicae* (Table 3).

The lowercase letters match the hybrid effect, capital letters compared the effect of inoculation and the letters in italic compared the effect of nitrogen. Same letters do not differ by Tukey test at 5% probability. Hybrid 1

Table 2. Shoot nitrogen percentage (%SN) and total shoot nitrogen (TSN) of three corn genotypes inoculated or not with *Herbaspirillum seropedicae* under different nitrogen levels. Dourados, MS, Brazil (2013).

Nitrogen levels		%SN											
		H1				H2				H3			
N_0	SI	1.10	a	A	*b*	1.14	a	A	*b*	0.90	a	A	*b*
	CI	1.07	a	A	*b*	1.09	a	A	*b*	0.76	b	A	*b*
N_{540}	SI	2.09	a	A	*a*	1.86	a	B	*a*	1.38	b	B	*a*
	CI	1.54	c	B	*a*	2.36	a	A	*a*	1.88	b	A	*a*
		TSN (mg plant^{-1})											
		H1				H2				H3			
N_0	SI	33.76	a	A	*b*	48.20	a	A	*b*	50.12	a	A	*b*
	CI	64.78	a	A	*b*	60.19	a	A	*b*	62.49	a	A	*b*
N_{540}	SI	144.60	a	A	*a*	181.40	a	B	*a*	151.00	a	B	*a*
	CI	118.54	b	A	*a*	238.59	a	A	*a*	246.55	a	A	*a*

Table 3. Analysis of variance of shoot nitrogen percentage coming from the fertilizer (%SNF), shoot nitrogen coming from the fertilizer (SNF) and shoot N use efficiency (SNUE) of three corn hybrids subjected to different nitrogen levels and inoculated with *Herbaspirillum seropedicae.* Dourados, MS, Brazil (2013).

Source of variation	GL	Mean square		
		%SNF	SNF	SNUE
Hybrid (H)	2	18.32*	12129.35*	415.95*
Inoculation (I)	1	3.81	9100.66	312.09*
H*I	2	1.67	6157.04*	211.14*
Residue	18	2.44	1350.96	46.32
CV (%)		1.77	23.11	23.11

* – significant by Tukey test to 5% probability. CV- coefficient of variation.

(Maximus), Hybrid 2 (P3646H) and Hybrid 3 (BRS 3035); SI (without inoculation) CI (inoculation).

The %SNF was higher for P3646H with percent increases of 2.8 and 3.1% compared to the hybrids Maximus and BRS3035, respectively (Figure 3). In average, 89.8% of total N accumulated in the shoot of P3646H comes from the fertilizer. On the other hand, SNF differed between corn hybrids and in the presence and absence of inoculation with *H. seropedicae*. There was no significant difference (p≤0.05) between corn hybrids without inoculation with *H. seropedicae* while the hybrids P3646H and BRS3035 showed higher SNF in the presence of inoculation with *H. seropedicae*, both differing from the treatment without inoculation (Table 4). This increase in SNF when inoculated with *H. seropedicae* may have occurred due to the production of auxins by the bacteria which stimulates root growth and increases the explored volume of soil contributing to the increase in the amount of nutrient absorbed.

Shoot N use efficiency (SNUE) which refers to the use of N fertilizer in relation to the amount of N applied as a fertilizer differed between corn hybrids and in the presence and absence of inoculation with *H. seropedicae* (Table 4). There was no significant difference (p≤0.05) for SNUE between corn hybrids without inoculation with *H. seropedicae* while the hybrids P3646H and BRS 3035 showed higher SNUE in the presence of inoculation with *H. seropedicae*, with percentage increases of about 34.3 and 64.4% compared to the treatment without inoculation (Table 4). Opposite results were found for the hybrid Maximus regarding SNF and SNUE. This hybrid showed decrease of 22.11% in SNF and SNUE when compared to the treatment inoculated with *H. seropedicae* (Table 4). Most studies show that there is great variation in the use of N fertilizers by plants, rarely exceeding 50% of the applied. In this study, the relatively high use by the corn hybrids compared to the treatments with and without inoculation with *H. seropedicae* is probably related to the conditions in the pot, where the root system remains confined, exploring the entire volume of soil and also

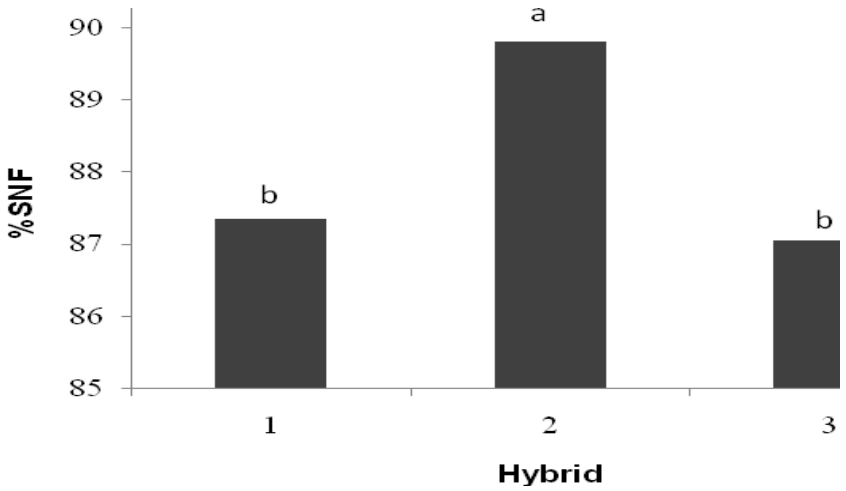

Figure 3. Shoot nitrogen percentage coming from the fertilizer (%SNF) of three corn genotypes. Hybrid 1(Maximus), Hybrid 2 (P3646H) and Hybrid 3 (BRS 3035).

Table 4. Shoot nitrogen coming from the fertilizer (SNF) and shoot nitrogen use efficiency (SNUE) of three corn genotypes inoculated or not with *Herbaspirillum seropedicae*. Dourados, MS, Brazil (2013).

	SNF (mg plant^{-1})						SNUE (%)					
	H1		H2		H3		H1		H2		H3	
SI	126.46	aA	161.25	aB	131.03	aB	23.41	aA	29.86	aB	24.26	aB
CI	103.57	bA	216.59	aA	215.41	aA	19.17	bA	40.11	aA	39.89	aA

without N leaching losses to other layers out of roots' reach. Similar results were obtained by Brito et al. (2011) evaluating the contribution of nitrogen biological fixation, nitrogen fertilizer and soil nitrogen to the development of cowpea and common beans.

The lowercase letters match the hybrid effect and capital letters compared the effect of inoculation. Same letters do not differ by Tukey test at 5% probability. Hybrid 1 (Maximus), Hybrid 2 (P3646H) and Hybrid 3 (BRS 3035); SI (without inoculation) CI (inoculation).

Conclusions

There is a distinction between corn genotypes regarding shoot dry matter production and shoot nitrogen percentage coming from the fertilizer. The shoot dry matter production is influenced by the inoculation with *H. seropedicae*. The inoculation with *H. seropedicae* in the presence of nitrogen fertilization promotes increases of approximately 32% in the amount of nitrogen in the shoot of the hybrid P3646H and 62% of the hybrid BRS 3035. The hybrids P3646H and BRS3035 showed increases of 34.3 and 64.4%, respectively in nitrogen use efficiency when inoculated with *H. seropedicae* with no addition of nitrogen, what allows to infer is that the BNF behaves as an important source of N to the system.

Conflict of Interest

The author(s) have not declared any conflict of interest.

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Technical efficiency and productivity growth in Egypt's cotton production

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The production of cotton has great economic importance in Egypt. However, in the last years an important decline was observed both in cotton production and cultivated area. One of the suggested causes of this decline is the low productivity growth. In this paper we employ the stochastic frontier analysis to analyze the evolution of total factor productivity (TFP) from its determinants (including the efficiency). The empirical results indicate that technical efficiency improves substantially during the period considered, but TFP growth is small. This low growth is due primarily to the unfavorable evolution of technical progress. Policy implications are offered in light of the results.

Key words: Cotton, Egypt, stochastic frontier, technical efficiency, technical progress, total productivity.

INTRODUCTION

The Egyptian cotton is a peculiar type of cotton that is characterized by high quality. From the botanical point of view, it belongs to the species *Gossypium barbadense.* The production of cotton has great economic importance in Egypt (Abdel-Salam and Negm, 2009). Cotton production is important in Egypt because it helps to guarantee the food security for population (Global, 2009). This contribution to the food security takes several forms. To begin with, cotton production is a source of income for rural families, who can then afford the basic products (milk, fruits, vegetables, clothes, etc). Also the exports of cotton enable the import of food. In addition, the oil extracted from the cotton seeds is used for culinary purposes, and the chaff (oilcake) is used to feed animals, since it is rich in proteins. Furthermore, cotton production is important because it is a very intensive activity in the use of labor and it is carried out principally by small family exploitations. Additionally, it generates new opportunities of employment in the subsequent processes (e.g. ginning, transportation, commercialization and the local textile industry). Finally, the production of cotton is important as a raw material to impel the textile industry in Egypt.

Despite the importance of cotton production in Egypt, a substantial decline was observed in cotton production in the last years, while the production of other agricultural products, such as sugarbeet, fruits, vegetables, rice and wheat, increased in Egypt in a remarkable way (Global, 2009). In addition, the decline of cotton production in Egypt took place in a context in which the Egyptian economy was growing and so was the Egyptian cotton textile industry, while the production, consumption,

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exports, and imports of cotton did not decrease at theglobal level but remained stable (Karvy, 2009).

Some studies on cotton in Egypt (such as Abdel-Salam and Negm, 2009) enumerate multiple causes (at the national and international level) that can explain the evolution of cotton production. One of the suggested causes is the low productivity growth, given that productivity is an important determinant of the evolution of product competitiveness. And competitiveness is one of the principal factors that determine increases in the demand of products and, as a consequence, increases in the possibilities of production.

There are many descriptive studies that analyze cotton production in Egypt (RATES, 2005; Abdel-Salam et al., 2009; Global, 2009; Karvy, 2009), but there are not empirical studies on this sector that analyze total factor productivity from its determinants. The main objectives of this study are: (a) to estimate the technical efficiency of cotton production in Egypt through two stochastic frontier production models; (b) to investigate the evolution of TFP from its determinants -technical efficiency (TE), technical progress (TP) and the scale component (SC), and (c) to compare differences in TE, TP, SC and TFP among major cotton-producing provinces.

The results provide evidence that technical efficiency improves substantially during the period considered, but TFP growth is small. This low growth is due primarily to the unfavorable evolution of technical progress.

METHODOLOGY

Efficiency and productivity are generally measured using either parametric or non-parametric methods. There exist two main types of approaches that can be applied to estimate efficiency and decomposing productivity growth: Data envelopment analysis (DEA) and stochastic frontier analysis (SFA). Since the advent of SFA, it has been widely used to analyze efficiency and productivity (Si and Wang, 2011). In general, the SFA approach has been preferred in the agricultural economics literature (Coelli and Battese, 1996; Hasnah et al., 2004; Headey et al., 2010). The most important potential advantage of SFA is that it can separate noise in data from variations in efficiency. Given the inherent variability of agricultural production, the assumption that all deviations from the frontier are associated with inefficiency (as assumed in DEA approach) is difficult to accept in this sector.

Following previous literature in the agricultural field (Rae et al., 2006; Lambarraa et al., 2007; Jin et al., 2010; Si and Wang, 2011; Yoo et al., 20012), a measure of productivity in cotton production can start with a stochastic frontier production function:

$$Q_{it} = f(A_{it}, t; \alpha) e^{v_{it} - u_{it}} \quad (1)$$

Where Q_{it} is the cotton production of *i-th* province (*i* = 1, 2 . . . *N*) in the *t-th* time period (*t* = 1, 2 . . . *T*), $f(.)$ is the production tecnology, A_{it} is the cotton area of *i-th* province in the *t-th* time period, *t* is the time trend index, α is a vector of unknown parameters to be estimated; v_{it} is a vector of random errors which are assumed to be iid. $N(0, \sigma_v^2)$ and independent of u_{it}; and u_{it} is a vector of non-negative random variables which are assumed to account for technical inefficiency in cotton production. There are several specifications that make the technical inefficiency term u_{it} time-varying. In this paper we use the Error Components Model (ECM) specification proposed by Battese and Coelli (1992), and the Technical Efficiency Effects Model (TEEM) specification proposed by Battese and Coelli (1995).

In ECM, the inefficiency term u_{it} is defined as:

$$u_{it} = u_i \exp(-\eta[t - T]) \quad (2)$$

Where the distribution of u_i is taken to be the non-negative truncation of the normal distribution $N(\mu, \sigma_u^2)$, and η is a parameter that represents the rate of change in technical inefficiency. The positive (negative) value of η is associated with improvements (deterioration) in cotton production technical efficiency over time.

In TEEM, the non-negative random variables (u_{it}) are assumed to be independently distributed as truncations at zero of the $N(\mathrm{m}_{it}, \sigma_u^2)$ distribution; where:

$$m_{it} = z_{it}\delta \quad (3)$$

Where z_{it} is a vector of variables which may influence the efficiency of a firm and δ is a vector of parameters to be estimated. Differentiating the production technology in Equation (1) with respect to time and rearranging terms we have that:

$$\frac{d \ln f(.)}{dt} = TP + \varepsilon \dot{A} \quad (4)$$

Where $TP = \frac{\partial \ln f(.)}{\partial t}$ represents the technical progress or technical change, $\varepsilon = \partial \ln f(.) / \ln A$ is the elasticity of output with respect to the cotton area input, and a dot over A indicates its rate of change over time.

Totally differentiating the logarithm of *Q* in equation (1) with respect to time, the change in production can be represented as:

$$\dot{Q} = TP + \varepsilon \dot{A} - \frac{du}{dt} \quad (5)$$

In Equation (5) the overall productivity change is affected not only by TP and changes in cotton area, but also by the change in technical efficiency $(-du/dt)$; thus if du/dt is positive (negative), technical efficiency deteriorates (improves) over time.

To examine the effects of technical progress and changes in efficiency on TFP growth, the traditional definition for productivity total growth is used; that is, TFP is taken as the residual resulting from the output growth unexplained by input growth:

$$T\dot{F}P = \dot{Q} - s\dot{A} \quad (6)$$

Table 1. Summary statistics for cotton production and cotton area.

Governorates	Cotton production (Thousand ton)		Cotton area (Thousand hectare)	
	1990	2008	1990	2008
Dakahlia	123.01	44.45(-5.5)	53.14	18.25(-5.76)
Sharkia	88.05	25.79(-6.59)	45.78	11.51(-7.38)
Kafr Elshikh	92.1	66.96(-1.81)	42.85	29.66(-2.02)
Gharbia	85.15	22.4(-7.15)	36.29	7.09(-8.67)
Menoufia	41.36	3.01(-13.54)	16.05	1.54(-12.21)
Behairah	150.02	63.79(-4.64)	55.16	25.06(-4.29)
Beni Suef	41.31	9.17(-8.02)	20.11	4.49(-7.99)
Fayoum	30.89	15.05(-3.92)	13.86	7.3(-3.50)
Menia	61.34	4.71(-13.28)	29.5	1.73(-14.58)
Total	713.23	255.33(-5.55)	312.74	106.63(-5.80)

Source: Own elaboration from the sample data (Ministry of Agriculture and Land Reclamation, Egypt). Figures in parentheses are average annual growth rates (1990-2008).

Where S is the share of the input of cotton area in production costs.

By substituting Equation (5) in (6), equation (6) is rewritten as:

$$T\dot{F}P = TP - \frac{du}{dt} + (RS - 1)\dot{A} \quad (7)$$

Where RS denotes the measurement of returns to scale.

In Equation (7) the total factor productivity growth is decomposed into three components: Technical progress TP), changes in technical efficiency $(CTE = -du/dt)$ and changes in the scale component $[CSC = (RS - 1)\dot{A}]$.

The interpretation of the three components in equation (7) is the usual: TP measures the change in frontier output, TP is positive (negative) if exogenous technical change shifts the production frontier upward (downward) for a given level of inputs. CTE can be interpreted as the rate at which an inefficient province catches up to the production frontier. Depending on whether *RS* > 1, *RS* < 1 or *RS* = 1, positive scale effects, negative effects or non-scale effects, respectively, will exist.

RESULTS AND DISCUSSION

In this study we use panel data at the province level covering the period 1990-2008. The dataset was obtained from "Agricultural Statistics", a publication by the Ministry of Agriculture and Land Reclamation (MALR, 1990-2008) which contains information on the main nine cotton-producing provinces. One of the limitations of this publication is that it offers too scarce data for a detailed study of total productivity.

The most important problem from the empirical point of view is that, in this publication, the only real and direct data provided are the production (Q) and the cotton area (A). Therefore, there is no direct data for others inputs such as labor input or capital input. Additionally, there is a lack of province-level statistical information on specific variables such as education, credit use, infrastructures, land quality, average size of plantations, composition of labor or characteristics of machinery, which may have a significant impact on the behavior of efficiency and productivity. There is also no data at the farm level, such as age, farm income, off-farm income, womens' participation, experience or farm size.

Table 1 presents the summary statistics for production and cotton area by provinces. This table shows that, for the total sample, the average annual growth rates for the period 1990-2008 were negative for output and for the cotton area input, the decrease of output (-5.55%) is slightly lower than that of the cotton area input (-5.80%). At the province level, the two variables also show an unfavorable evolution, but the average annual growth rates are not homogeneous across provinces.

In order to decompose TFP growth (Equation 7) it is necessary to choose a functional form for the stochastic frontier production function (Equation 1). Taking into account the related literature (Hasnah et al., 2004; Rae et al., 2006; Lambarraa et al., 2007; Jin et al., 2010), and that the translog form is a flexible functional form, in this paper we propose a translog stochastic frontier production function:

$$\ln Q_{it} = \alpha_0 + \alpha_a \ln A_{it} + \alpha_t t + \frac{1}{2}\alpha_{aa}(\ln A_{it})^2 + \frac{1}{2}\alpha_{tt}t^2 + \alpha_{at}(\ln A_{it})(t) + v_{it} - u_{it} \quad (8)$$

Since in this study the statistical information on province-specific variables is not available, we create dummy variables (D_i) for the different provinces to identify the possible heterogeneous behavior across them, with D_i being equal to 1 if the province is *i* and zero otherwise. Additionally, we incorporate a time variable to verify if inefficiency has increased or decreased in the analyzed period. Consequently, we specify the technical

Table 2. Maximum Likelihood estimates of the stochastic frontier production models.

Variable	ECM		TEEM	
	Coefficient	Std. error	Coefficient	Std. error
Frontier production function				
Constant	0.249289	(0.114530)**	0.289005	(0.068000)***
LnA	0.563986	(0.072296)***	0.699767	(0.037490)***
(t)	-0.051286	(0.014198)***	-0.056645	(0.009873)***
½[lnA]²	0.056628	(0.065249)	0.107206	(0.050583)**
½[t]²	0.009530	(0.002870)***	0.010833	(0.002081)***
(lnA)(t)	0.062670	(0.008544)***	0.054666	(0.005415)***
Inefficiency effects				
Constant			-0.370452	(0.375941)
D2			-0.298510	(0.342997)
D3			-0.386331	(0.365466)
D4			-0.285047	(0.350853)
D5			0.310457	(0.240579)
D6			-0.338407	(0.298452)
D7			0.149254	(0.271432)
D8			0.421313	(0.225923)*
D9			-0.077183	(0.321359)
(t)			-0.120472	(0.026703)***
Sigma-squared	0.0705182	(0.009734)***	0.154840	(0.040440)***
Gamma	0.062285	(0.088807)	0.888835	(0.037669)***
Mu	0.038352	(0.093079)		
Eta	0.134596	(0.030282)***		
Log likelihood function	-23.082432		-0.725175	
LR test of the one-sided error	24.227818		68.942331	
Total number of observations	171		171	

***, ** and * indicate significance at 1, 5 and 10% level, respectively.

inefficiency effects (Equation 3) as:

$$m_{it} = \delta_0 + \sum_{i=2}^{N} \delta_i D_{it} + \beta t \quad (9)$$

From the estimation of the translog stochastic frontier production function (Equation 8), the components of productivity change can be calculated. The technical efficiency levels (*TE*) and changes in technical efficiency (*CTE*), technical progress (*TP*), and returns to scale (*RS*) and changes in the scale component (CSC) are obtained as:

$$TE = \frac{Q_{it}}{f(.)} = \exp(-u_{it}) \ , CTE = \exp(-u_{it}) - \exp(-u_{it-1}) \quad (10)$$

$$TP = \frac{\partial \ln Q_{it}}{\partial t} = \alpha_t + \alpha_{tt} t + \alpha_{at} \ln A_{it} \quad (11)$$

$$RS = \frac{\partial \ln Q_{it}}{\partial \ln A_{it}} = \alpha_a + \alpha_{aa} \ln A_{it} + \alpha_{at}(t), \ [CSC = (RS-1)\dot{A}] \quad (12)$$

The maximum-likelihood estimates for the parameters of models, defined by Equations (8) and (2), and equations (8) and (9), can be obtained by using the FRONTIER 4.1 program (Coelli, 1996), in which variance parameters are expressed in terms of $\sigma^2_s = \sigma^2_u + \sigma^2_v$ *and* $\gamma = \frac{\sigma^2_u}{\sigma^2_s}$, and γ is an unknown p

arameter to be estimated. The adequacy of the models proposed and the different variants with regard to the type of distribution for inefficiency and the possible mu (μ) and eta (η) values have been tested. From the various alternatives, we selected (following the criterion of the likelihood ratio test) the results shown in Table 2. The results of the two models are similar. Taking into account the values obtained for the log likelihood function, TEEM offer a better specification than ECM.

Table 3. Technical efficiency by year (total sample).

Years	ECM	TEEM	Mean
1990	0.519	0.378	0.449
1991	0.557	0.420	0.489
1992	0.594	0.575	0.585
1993	0.629	0.710	0.670
1994	0.663	0.478	0.571
1995	0.695	0.491	0.593
1996	0.725	0.721	0.723
1997	0.752	0.767	0.760
1998	0.778	0.634	0.706
1999	0.802	0.749	0.776
2000	0.823	0.788	0.806
2001	0.843	0.801	0.822
2002	0.860	0.879	0.870
2003	0.876	0.903	0.890
2004	0.891	0.882	0.887
2005	0.903	0.905	0.904
2006	0.915	0.900	0.908
2007	0.925	0.879	0.902
2008	0.934	0.895	0.915
Mean (1990-2008)	0.773	0.724	0.749
Rate[a]	3.318	4.905	4.112

[a]Average annual growth rate (1990-2008).

All variables appearing in natural logarithms were divided by their geometric mean prior to estimation. The time trend was at zero in 1999. As a result, the coefficients of the first-order terms of the variables in natural logarithms can be interpreted as production elasticities in that year evaluated at the geometric mean of the explanatory variables.

The first-order coefficient, α_a, has the anticipated positive sign and it is statistically significant. The coefficient of the first order term of the time trend, α_t, is statistically significant in the two models, but with negative sign; that is, technical progress shifts the production frontier downward for a given level of inputs. This result must be considered in light of the definition of the time trend variable that serves as a proxy for technical progress.

The dummy variable coefficients of the different governorates are not statistically significant for TEEM except for one governorate (Fayoum), which is statistically significant at 10% level of significance. However, the coefficient for the time variable (β) is statistically significant in this model. The negative and statistically significant coefficient for the time variable (β) suggests that technical inefficiency in cotton production in Egypt tended to decrease during the studied period. As the eta (η) value is statistically different from zero (it takes a positive value), this implies that technical inefficiency in these provinces is not time-invariant; the level of technical efficiency improves in the analyzed period (Table 3). The positive and statistically significant coefficient of η is compatible with the negative and statistically significant coefficient β (as we explained before).The variance parameter, gamma (γ), is statistically significant in TEEM and close to one, which suggests the relevance of technical inefficiency in explaining output variability. This estimate is consistent with the results of previous works (Battese and Broca, 1997; Lambarraa et al., 2007).

Table 3 shows the annual levels of the estimated technical efficiency, using the two proposed models in this study (results of the total sample). By comparing the results obtained for the two models, it is possible to emphasize that the estimated annual levels of technical efficiency are similar in both of them. With respect to the average level of efficiency of the period 1990-2008, the average of the two models is 0.749. The two models consistently show that technical efficiency improves during the period considered. The average annual growth rate is also similar in the two models, with the average across the two models being 4.112%.

Results of TFP growth decomposition by provinces are reported in Table 4. Changes for the two models and

Table 4. Results of TFP change decomposition by provinces[a].

Provinces	ECM				TEEM			
	TP	CSC	CTE	CTFP	TP	CSC	CTE	CTFP
Dakahlia	-0.0182	0.0243	0.0131	0.0192	-0.0265	0.0148	0.0212	0.0095
Sharkia	-0.0403	0.0329	0.0191	0.0117	-0.0458	0.0219	0.0277	0.0039
Kafr Elshikh	-0.0026	0.0081	0.0148	0.0203	-0.0129	0.0046	0.0229	0.0146
Gharbia	-0.0518	0.0398	0.0214	0.0094	-0.0558	0.0278	0.0303	0.0023
Menoufia	-0.1007	0.0629	0.0362	-0.0016	-0.0985	0.0508	0.0327	-0.0151
Behairah	0.0121	0.0168	0.0049	0.0338	-0.0001	0.0088	0.0157	0.0244
Beni Suef	-0.0711	0.0381	0.0330	0.0000	-0.0727	0.0284	0.0357	-0.0086
Fayoum	-0.0851	0.0168	0.0372	-0.0311	-0.0848	0.0130	0.0373	-0.0346
Menia	-0.0712	0.0719	0.0276	0.0282	-0.0728	0.0534	0.0348	0.0154
Total Sample	-0.0477	0.0346	0.0230	0.0100	-0.0522	0.0248	0.0287	0.0013

[a]Mean changes for the period1990-2008.

the mean for every period are shown. As explained above, the decomposition of changes in total factor productivity (CTFP) is calculated as a sum of technical progress (TP), changes in scale component (CSC) and changes in technical efficiency (CTE). The two models offer similar results for the total sample in the sense that both indicate the negative contribution of the technical progress component and the positive contributions of technical efficiency and scale components. In addition, for total sample, the two models have similar results for the magnitude of the average contribution of the different components. As a consequence of the above, a slight average productivity growth is estimated (0.13% per year for TEEM and 1% per year for ECM).

The negative contribution of the technical progress component must be considered in light of its definition as a residual. A negative value of this component does not mean that in average there was no technical progress in cotton production; what this result probably indicates is that its contribution was small, and that there are possibly other factors (which have not been explicitly considered in the model) whose negative effect on production outweighs the small positive effect of the possible technical advances (e.g. land quality, climate changes, plant diseases, plant viruses, etc).

The positive value for the changes in the scale component means that the cotton production sector took advantage of the economies of scale. With the statistical information available it is not possible to identify the specific factors responsible for this improvement in scale economies.

The positive value for the changes in technical efficiency shows that the gap between the production frontier and the actual cotton production was squeezed throughout the analyzed period.

The changes in productivity and in its components are not homogeneous across the provinces. In three provinces (Menoufia, Beni Suef and Fayoum) the average changes in productivity are not positive. The components CSC and CTE maintain the sign of their contribution across provinces, although with varying magnitude. The TP component has a positive contribution in Behairah (ECM), and for the other provinces has a negative contribution but also with different size in each of them.

CONCLUSIONS AND POLICY IMPLICATIONS

Despite the economic importance that cotton production has in Egypt, a remarkable decline was observed in the last years. Considering the nine main governorates of cotton production in Egypt (and the period 1990-2008), in the year 1990 the cotton production exceeded 700 thousand tons, and in the year 2008 the level of production was reduced to approximately 255 thousand tons (it declined at an annual average growth rate of -5.55%).

Two stochastic frontier models were estimated with the purpose of providing evidence on the possible problems affecting the cotton production system in Egypt. The results of the estimation for the two models are consistent. The two models provide similar results for the technical efficiency (considering the total sample), and they indicate the negative contribution of technical progress and the positive contributions of technical efficiency and scale components to total productivity growth.

These results provide evidence that the cotton production system in Egypt offers some deficiencies. Although the levels of technical efficiency improve substantially during the period considered (with an average annual growth rate across the two models of around 4%), in this production system the increase in total productivity is low -varying from 0.13% per year (TEEM) to a 1% per year (ECM). Therefore, this low

productivity growth can be one of the causes of cotton production decline. This low productivity growth is due primarily to the negative contribution of technical progress to total productivity growth. The negative contribution of technical progress does not mean that in average there was no technical progress in cotton production; what this result probably indicates is that its contribution was small, and that there are possibly other factors whose negative effect on production outweighs the small positive effect of the possible technical progress. Another possible reason is that the existent technology might not be used appropriately.

This production system leads to varying results in the different cotton-producing provinces of Egypt, since the results for the changes in productivity and in its components are not homogeneous across them. This might suggest that some provinces have worse conditions for the production of cotton. Although the situation of scarce statistical information limits the scope of the results obtained in this study, some recommendations can be made: (a) Policy makers should improve the database of the cotton production sector (at provincial and farm levels). With this kind of statistical information it would be possible to carry out a detailed study of total productivity and identify the specific factors that influence technical progress; (b) Although these factors are not precisely identified due to the lack of statistical information, following previous literature in the agricultural field (Iráizoz et al., 2003; Bozoglu and Ceyhan, 2007) some effective policy measures (such as providing better extension services and farmer training programs) can be proposed to improve the capacity of farmers; (c) policy makers should take into account that the behavior of productivity and its components is not homogeneous across provinces. Therefore, it seems reasonable to propose specific measures for each of them, orientating production towards the provinces where productivity shows a relatively better behavior.

Conflict of Interest

The authors have not declared any conflict of interest.

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