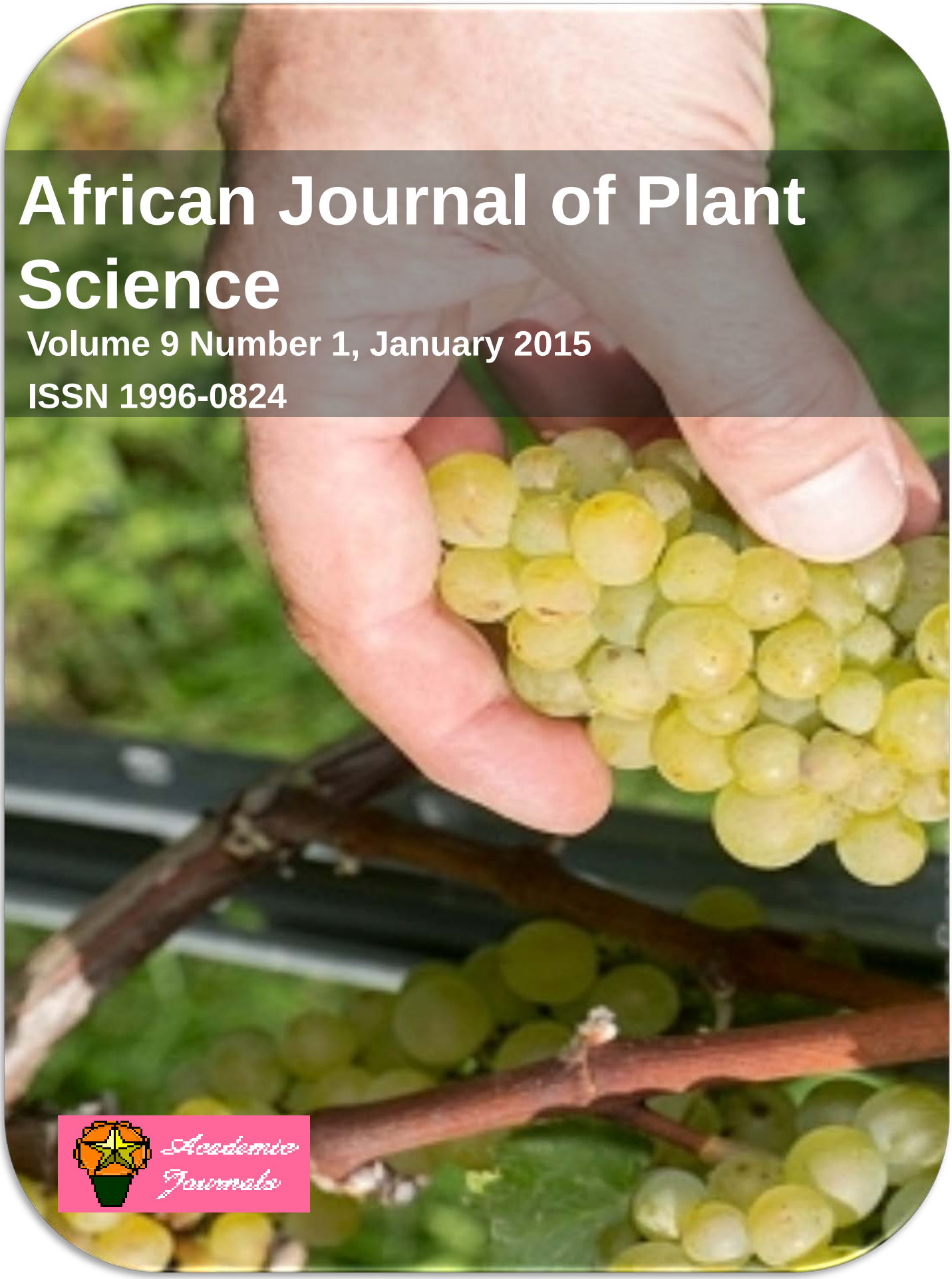

# African Journal of Plant Science

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Pitout JDD, Church DL, Gregson DB, Chow BL, McCracken M, Mulvey M, Laupland KB (2007). Molecular epidemiology of CTXM-producing Escherichia coli in the Calgary Health Region: emergence of CTX-M-15-producing isolates. Antimicrob. Agents Chemother. 51: 1281-1286.

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*Full Length Research Paper*

# Development of elite medium staple cotton (*G. Hirsutum*) genotypes for production in middleveld upland ecologies

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**Ten (10) medium staple cotton genotypes comprising of five commercial varieties and five experimental lines were evaluated for field performance, genetic and environmental variability. The trials were laid out in a randomized complete block design (RCBD) with three replications. Analysis of variance was done for total seed cotton yield, lint yield, boll weight, earliness and gin out turn (GOT %) using Genstat 14th edition while stability and adaptability analysis was done using the AMMI model and the GGE biplot software. Significant differences (P<0.05) in genotype performances were observed in all the traits except for boll weight and earliness. The environment (E) effect was significant (P<0.05) for seed cotton yield and gin out-turn percentage (GOT %). The genotype (G) effect significantly (P<0.05) accounted for differences in boll weight and earliness index. The genetic x environment (GEI) interaction was not significant across the two seasons. SZ 9314 showed wide adaptation to all environments, a well-known and recommended characteristic of the commercial variety. These results show that 644-98-11, 917-05-7 and SZ-95-7 are promising genotypes that can be registered for production in upland cotton ecologies worldwide and they can be incorporated in future cotton improvement program. It is suggested that fibre quality traits for these experimental lines should be assessed.**

**Key words:** Stability, genetic variation, environmental variability, environment interaction (GEI), additive main effect and multiplicative interaction (AMMI), genetic by environment (GGE), gin out turn (GOT %).

## INTRODUCTION

Cotton (*Gossypium hirsutum* L.) commonly referred to as the “king of fibres’ has global importance in the industrial production of fibre for the cloth industry, vegetable oil for human consumption and stock feed for domesticated

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animals (Sibanda, 2012). The crop is grown on commercial basis in both developed and developing nations by smallholder and large scale commercial farmers. Global production is on commercial basis by both smallholder and large scale commercial farmers. China, India, Brazil, United States of America and Pakistan in descending order form the top five cotton producing countries in the world (ICAC, 2013). However, Zimbabwe is among the top ten cotton producing countries in sub Saharan Africa and was, until recently, the regional standard bearer for quality (Gono, 2012).

The production of cotton in Zimbabwe is mainly clustered into two mega environments on the basis of altitude, levels of production and on the nature/types of varieties grown however, new areas in natural regions one and two of the country have also witnessed an upsurge in cotton growers; a feat that can be attributed to the effects of climate change. Expansion in new areas of production coupled with climatic pattern changes calls for the revision and redefinition of the breeding objectives as well as taking stock of germplasm which can adapt to the challenges associated with the emerging production environments. The target populations of environments (TPE) for cotton production have expanded prompting the need to increase our research sites, more rigorous variety testing schemes; minimize the effects of genetics by environment interaction (GEI) and to recommend adapted varieties for these new areas based on realistic research findings. Furthermore, the effects of climate change require a thorough and arduous variety testing scheme which ensures precise variety recommendation (Martin et al., 2013) to all cotton growing farmers. It is against this backdrop that the cotton breeding program continues to develop and evaluate a wide range of germplasm basically divided into medium staple middleveld, medium staple lowveld and the long staple cotton genotypes (Cotton Research Institute, 2012). Evaluation of varieties under different conditions representing different production environments is recommended to avoid selecting varieties which are only adaptable to a few environments. Proper GEI analysis groups with similar environments together (Ceccarelli et al., 2009) defines mega production environments as well. Classification of environments helps in culling similar testing environments which facilitate effective utilization of scare resources by testing germplasm in only different environments (Gono, 2012).

Usually, breeding programs focus on the development and selection of superior genotypes for seed cotton yield, lint yield and superior fibre quality (Feiyu and Weujun, 2013). In tandem with this focus, breeding objectives are meant to satisfy the needs of all stakeholders in the cotton value chain. The farmer is concerned with high seed cotton yield, the ginner requires high lint yield while the spinner considers fibre quality as important at their interface. Using the bulk pedigree breeding method with minor modifications, the cotton breeding programs generates new every season from the diverse gene pool available. The progeny (F1) are allowed to self in the first generation before selections are initiated in the succeeding season. Selections of the progenies continue for 2 - 3 generations and at F4 or F5 selected progenies will constitute the strains. Evaluations for either specific or broad adaptations are usually done.

Matching cotton variety selection with its production environment is often challenged by the occurrence of significant genotype x environment interaction in the variety development programmes. The objectives of this paper were to identify the most stable and adaptable cotton varieties combining the best on seed cotton yield, lint yield, ginning out turn, boll weights and earliness index. The study would also identify ideal test that are either representative of the target population of environments and also those that are discriminating in carrying out cotton research trials.

## MATERIALS AND METHODS

A total of 10 cotton genotypes were evaluated at eight locations which represent the major cotton growing areas in Zimbabwe. The genotypes consisted of five registered varieties which were used as checks and the other five genotypes which were elite cotton experimental lines. Genotypes used in the current project were selected based on merits (Table 1).

### Testing locations and seasons

The genotypes were evaluated in eight locations which represented all major cotton growing areas of Zimbabwe. Characteristics of each location are presented in Table 2.

### Trial design

The trials were laid out in a randomized complete block design (RCBD) with 3 replications at all the locations. Randomization was done at each individual site. Plot sizes measured 5 rows x 6 m x 1 m (30 $m^2$).

### Agronomic field management practices

#### *Planting*

The crop was planted on ridges after opening the planting furrows using a ridger. The seed was hand placed in the plots at a rate of 3-5 seeds per. The crop was thinned to one plant per station to achieve a desired plant population of about 33 333 plants per hectare.

#### *Fertilizer application*

Fertilizer application followed guidelines and recommendations from the cotton agronomy annual (C.T.C, 2008). A basal application of compound L (N: P: K: S = 5:18:10:8: {0.25B}) was manually banded at a rate of 250 kg per hectare to the planting furrows.

**Table 1.** Names and characteristics of genotypes evaluated during the 2012-2013 and 2013-2014 cropping seasons.

| Code | Genotype | Status | Type | Growth habit | Maturity |
|---|---|---|---|---|---|
| 1 | CRI MS 1 | Check | Medium Staple | determinate | Long |
| 2 | 917-05-7 | Elite line | Long Staple | Indeterminate | Long |
| 3. | 648-01-4 | Check | Long Staple | Semi-determinate | Long |
| 4. | SZ 9314 | Check | Medium Staple | Indeterminate | Medium |
| 5. | CRI MS 2 | Check | Medium Staple | Semi determinate | Medium |
| 6. | QM 301 | Check | Medium Staple | Semi-determinate | Short |
| 7. | SZ 95-7 | Elite line | Medium Staple | Determinate | Short |
| 8. | BC 853 | Elite line | Medium Staple | Semi determinate | Medium |
| 9 | 644-98-11 | Elite line | Medium Staple | Determinate | Short |
| 10 | 280-94-10 | Elite line | Long Staple | Indeterminate | Long |

**Table 2.** Testing locations and season characterization for genotype evaluation

| Location | Natural region | Rainfall (mm) 2011 - 2012 | Altitude masl | Rainfall 2012-2013 | Max Temperature (°C) | Soil type |
|---|---|---|---|---|---|---|
| Shamva | 11a | 807.20 | 1149 | 699.00 | 38 | Basaltic, loamy |
| Kadoma | 11b | 729.60 | 1156 | 800.13 | 38 | Red clay loamy |
| Wozhele | 111 | 867.88 | 1245 | 722.34 | 37 | Alluvial |
| Kuwirirana | 1V | 669.82 | 996 | 440.00 | 38 | Black |
| Chitekete | 1V | 867.45 | 914 | 799.00 | 42 | Black vertisols |
| CC Mollen | 11b | 805.46 | 1089 | 864.90 | 37 | Black loamy |
| Muzarabani | V | 706.54 | 600 | 635.50 | 40 | Clay alluvial |
| Chisumbanje | V | 872.12 | 300 | 744.35 | 41 | Black Alluvial vertisols |

Ammonium nitrate (34.5% N) was applied at a rate of 150 kg per hectare at the ninth week after crop emergence.

***Pest control***

A uniform cotton management regime was applied at all the trials to control all cotton pests. The general recommended cotton pest scouting and control protocol developed at Cotton Research Institute (C.R.I, 2012) was used. Pests were kept at below the economic thresholds levels following weekly scouting.

***Weeding***

An average of three weedings was done manually across all the test sites using hoes and ox drawn cultivators. No chemical weed control was ever applied.

**Data management**

Days to flowering and physiological maturity, plant height, seed cotton yield, yield components such as boll weight, boll mass and the number of bolls per plant were also recorded in the field.

***Seed cotton yield***

The total seed cotton yield was weighed using a digital scale after picking. The total yield is computed from the sum of weight of boll samples plus the seed cotton weights at picks 1, 2 or 3. Hence the total seed cotton yield is a collection of all the split bolls picked from each plot at each picking period.

***Lint index or gin out turn ratios***

Gin out turn ratios or lint ratios, refers to the amount of fibre that is produced from any given sample of seed cotton after removing the seeds. For the gin out turn percentage, 100 boll samples from each plot were weighed and ginned. The resulting lint was packed and all the seeds from the ginned 100 boll samples were weighed. The percentage of lint from each sample was then calculated using simple proportion to determine the gin out turn percentage. GOT = (total seed cotton sample – total ginned seed weights) x 100% / (total weight of ginned sample).

**Data analysis**

Data was analyzed using Genstat software 14$^{th}$ edition. Analysis of variance (ANOVA) for seed cotton yield and GOT % was done for

**Table 3.** Overall performance in field characteristics of medium staple middle veld variety trial over two seasons (2011-12 & 2012 – 2013) in Zimbabwe.

| Genotype | Seed cotton yield (kg/ha) | Lint yield (kg/ha) | Gin out turn (%) | Boll wt (g) | Earliness index (%) |
|---|---|---|---|---|---|
| 644-98-11 | 1384 | 694.5 | 43.7 | 5.7 | 85.65 |
| SZ-95-7 | 1566 | 628.21 | 44.6 | 6.0 | 78.74 |
| CRI MSI | 1233 | 514.6 | 43.1 | 5.6 | 77.26 |
| CRI MS2 | 1282 | 435.5 | 43.5 | 6.0 | 74.84 |
| SZ-95-23 | 1337 | 567.6 | 42.4 | 6.2 | 77.34 |
| SZ 9314 | 1850 | 532.9 | 42.9 | 7.7 | 72.85 |
| 280-94-10 | 1305 | 549.6 | 42.3 | 6.3 | 86.42 |
| 648-01-4 | 1342 | 486.9 | 41.4 | 5.8 | 73.96 |
| QM 301 | 1218 | 580.1 | 43.7 | 6.3 | 70.28 |
| BC 853 | 1171 | 544.7 | 41.6 | 5.7 | 78.90 |
| Mean | 1299 | 553.38 | 43.2 | 6.0 | 77.39 |
| F Prob | 0.0241 | 0.329 | 0.94 | <0.001 | <0.001 |
| CV | 12.7 | 16.8 | 11.1 | 4.9 | 20.4 |
| LSD | 213.7 | 106.7 | 3.47 | - | - |
| SED | 101.7 | 53.592 | 1.75 | 0.11 | 3.709 |

each individual site in each season followed by a combined ANOVA across sites and across seasons in order to estimate the magnitude of the variance components on genotype performance. Significance of GEI necessitated the application of the additive main effect and multiplicative interaction (AMMI) model and the principal component analysis (PCA). Suitability and stability analysis for each variety on each production environment was estimated through the use of the genetic and genetic by environment (GGE) biplots (Gabriel, 1971).

## RESULTS

The combined analysis of variance for yield, yield components and lint yield of cotton over two years indicated that there were significant differences ($P<0.05$) among the genotypes and genetic by environment interaction for all traits except on boll weight and earliness index. There were significant differences ($P<0.05$) in total seed cotton yield (TSC), lint yield and gin out turn (GOT %) among the test genotypes while traits such as boll weight (BWT), earliness and seed weight (SWT) were not significant (Table 3). Mean seed cotton yield ranged from 1171 to 1850 kg/ha with SZ 9314 as the best yielder (1850 kg/ha) and its yield was significantly different from the other genotypes.

Across the two seasons, across all the testing sites SZ 9314 and 95-7 were the best performing genotypes in terms of mean seed cotton yield (1850 and 1566 kg/ha, respectively) (Table 3). GOT % values were generally high averaging at 43.2%. Genotype SZ 95-7 had the highest GOT % value of 44.6% while 648-01-4 had the least GOT % value (Table 3).

Earliness indices were high with genotypes 280-94-10 and 644-98-11 having 86.42 and 85.65%, respectively. Test site comparisons shows that Chisumbanje had the best mean seed cotton yield of 2006 kg/ha followed by Save Valley Experiment station with a seed cotton yield of 1905 kg/ha while Chitekete produced the least mean seed cotton yield of 789 kg/ha (Table 4). This can be attributed to the quality of the seasons in terms of annual rainfalls received at each site during the cropping period (Table 2).

The environment (E), genotype (G) and genetic x environment interaction (GEI) effects were all statistically significant ($P<0.05$) for seed cotton yield, gin out-turn percentage (GOT %), boll weight (BWT), earliness index (ERL) and lint yield across the sites (Table 5).

BC 853 and 917-05-7 had the least seed cotton yield but portrayed high stability for seed cotton yield. CRI MS1, CRI MS2, SZ-95-7, QM 301, 280-94-10 and 644-98-11 were good performers, giving seed cotton yields which were above the mean (Table 3). However, Figure 1 shows that CRI MS 2, BC 853 and 644-98-11 were highly unstable whilst CRI MS1, SZ-95-7, 917-05-7 and QM301 were relatively stable (PC1 scores near 0).

Lower yielding sites included Shamva and CRI and high yielding sites were Chizvirizvi, Chitekete, Wozhele, Save and Chisumbanje. The biplot shows genotypes 280-94-10 being nearest to the average environmental coordinate (AEC) followed by 919-05-7 hence revealing minimal sensitivity to environmental and interactive forces. CRI, Chitekete, Shamva and Wozhele also showed lesser interactive forces than Save Valley Experiment station, Chisumbanje and Kuwirirana. Genotypes CRI MS1, 280-94-10, SZ 9314, and QM 301 showed large interactive effects and were adaptable at CC Moleen, CRI and Chitekete (Figure 1).

All environments that are close to the centre are ideal

**Table 4.** Overall performance in field characteristics of medium staple middleveld variety trial in 2011/12 and 2012/13 seasons.

| Site | Seed cotton yield (kg/ha) | Lint yield (kg/ha) | Gin out turn (%) | Boll weight (g) | Earliness index (%) |
|---|---|---|---|---|---|
| Chisumbanje | 2006 | 845.8 | 42.4 | 5.6 | 70.09 |
| Chitekete | 789 | 349.85 | 44.2 | 5.2 | 88.44 |
| Dande | 988 | 398.45 | 40.4 | 5.1 | 83.22 |
| Shamva | 1209 | 489.96 | 43.7 | 5.1 | 78.70 |
| Kuwirirana | 829 | 378.05 | 45.6 | 5.2 | 73.64 |
| CRI | 1105 | 482.80 | 43.7 | 6.4 | 80.45 |
| Wozhele | 1765 | 710.71 | 40.2 | 6.4 | 66.87 |
| Save Valley | 1905 | 856.97 | 43.2 | 5.9 | 76.53 |
| Mean | 1324.1 | 564.1 | 42.9 | 5.6 | 77.24 |
| F Prob | 0.576 | <0.001 | 0.340 | 0.221 | <0.001 |
| CV(%) | 22.6 | 13.1 | 8.4 | 17.5 | 18.7 |

**Table 5.** AMMI analysis of variance for seed cotton yield of 10 cotton genotypes grown at 8 environments across two seasons in Zimbabwe.

| Source | df | SS | Total variation (%) | MS | F | F_prob |
|---|---|---|---|---|---|---|
| Total | 149 | 38875441 | | 260909 | | |
| Treatments | 49 | 16677953 | | 340366 | 2.15$^{ns}$ | 0.00081 |
| Genotypes | 9 | 2319466 | 6.0 | 257718 | 1.63* | 0.01819 |
| Environments | 7 | 7741801 | 19.9 | 1935450 | 2.43* | 0.05370 |
| Block | 10 | 7979672 | | 797967 | 5.05* | 0.00001 |
| Interactions | 36 | 6616686 | 17.0 | 183797 | 1.16* | 0.027890 |
| IPCA | 12 | 4136151 | 62.5 | 344679 | 2.18* | 0.001901 |
| IPCA | 10 | 1299524 | 19.6 | 129952 | 0.82* | 0.000783 |
| Residuals | 14 | 1181011 | | 84358 | 0.53 | 0.90673 |
| Error | 90 | 14217816 | | 157976 | | |

Ns = Not significant; * = Significant at 0.05, df = degree of freedom, SS = sum of squares, MS = mean square, F = frequency, F Prob = F probability.

test environments. Chisumbanje is an ideal test environment while Wozhele and CRI are fairly good. Chitekete and Kuwirirana show that they are diverse environments because they are furthest apart. Figure 2 show that Chisumbanje, CRI, Wozhele and Chitekete are ideal test environments in discriminating and representativeness. AMMI partitioned the variance components into two principal components (PC 1 and PC 2). The two principle components explained 76.39% of the total variation (Figure 2) and they were both significant (Table 5).

## DISCUSSION

Analysis of variance using the AMMI model was done to indicate the relative magnitude of genetic (G), environment (E) and genetic by environment (GE) interaction for seed cotton yield. The results show that the source of variation due to genotype (G), environment (E) and genotype x environment interaction (GEI) were significant at P<0.05. Genotype accounted for 4%, (E) for 35.8% and GEI for 23.8% of the total sum of squares (SS) respectively. The environment effect was responsible for the bigger part of the variation followed by GEI and lastly G. This large yield variation explained by environments indicated that the environments were highly discriminating and very diverse (Gabriel, 1971) and this is shown by the differences among the environmental means.

CRI MS2, 648-01-4 and 280-94-10 were closely associated and hence more adaptable at Wozhele whilst SZ-95-7, 644-98-11 and SZ-95-23 were adapted to Chisumbanje. Therefore, CC Moleen, CRI and Chitekete constitute a mega environment for the cultivation of CR1 MS 1, 280-94-10, SZ 9314, and QM 301.

These results indicate that traits with low heritability (seed cotton yield, lint yield, boll sizes) can be affected by environmental conditions more than traits with high heritability values.

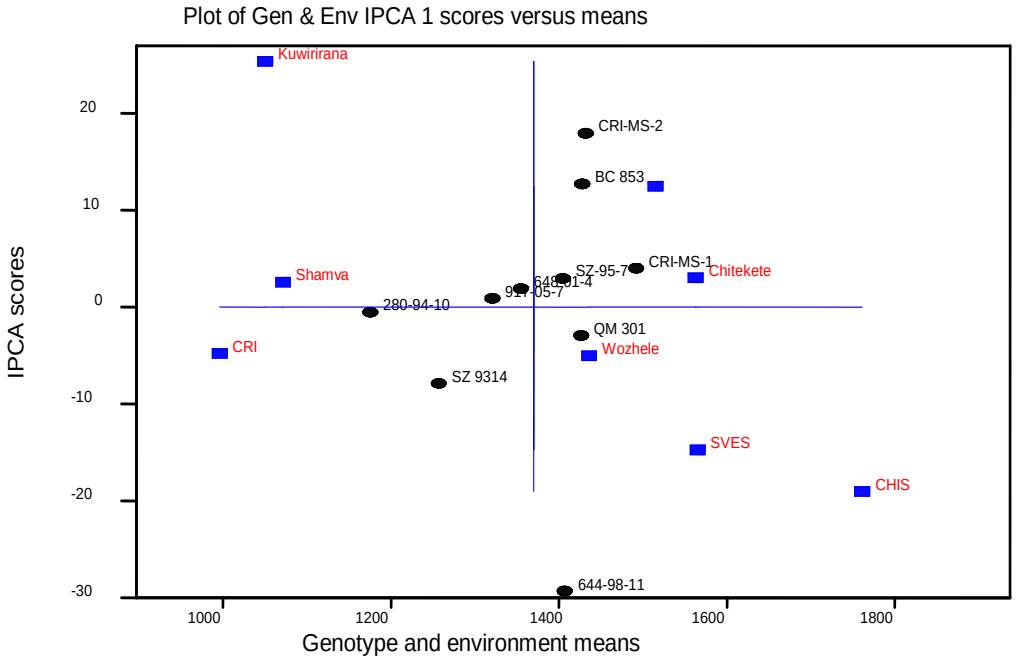


**Figure 1.** AMMI biplot showing the main and interaction effects of genotypes and environments on seed cotton.

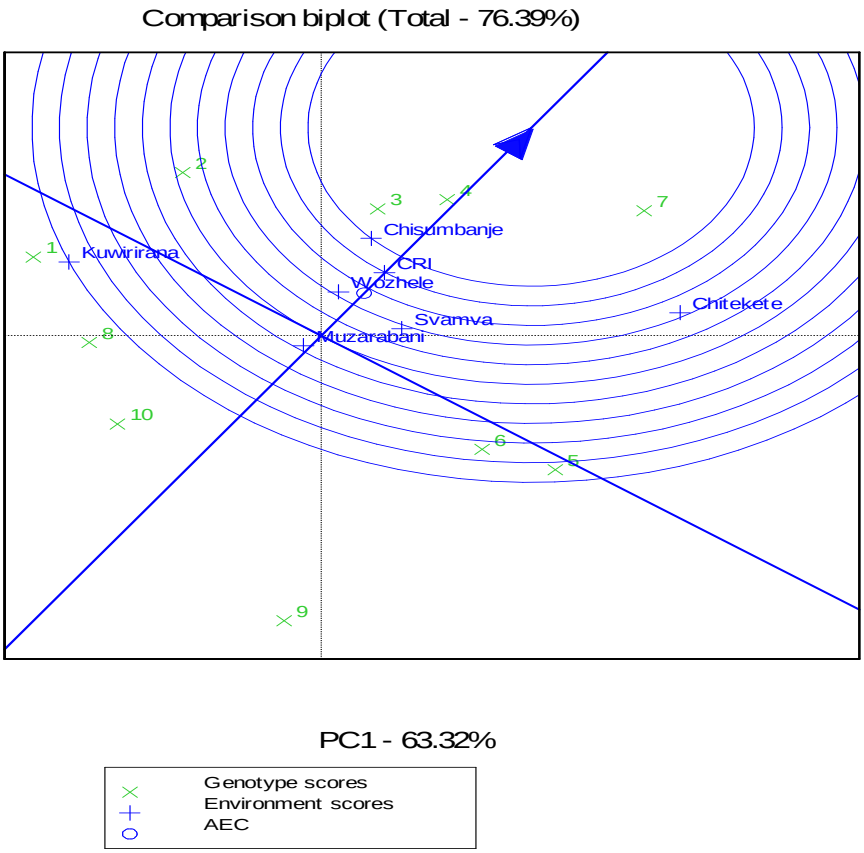


**Figure 2.** GGE biplot showing relationships among the test environments.

The results further confirm that improvement of the characteristics having low heritability can be done through pedigree selection and or through progeny testing (DeLacy et al., 2001). The results agree with work done by Shankar and Dhayal (2013) where it was noted that the pedigree selection method can be used to improve traits with low heritabilities.

Chenu et al. (2011) used this phenomenon creatively in their study of cotton test locations and they managed to identify a minimum set of test locations for cotton research in India. An ideal test environment is one which should be both discriminating of the genotypes and representative of the mega environment (Chisumbanje, CRI and Wozhele are ideal test sites because of their near proximity to the inner concentric circle) (Campbell and Jones, 2005). Such sites can be used for early generation screening of experimental lines while discriminating sites can be used for selecting specifically adapted varieties in this mega environment. Kuwirirana on the other hand is highly discriminating and it can be used for culling of genotypes.

## Conclusion

The study indicated that both genotypes and GE interaction were significant for both seed cotton yield and GOT%. This allowed easier selection of genotypes for these traits. CRI MS1, 644-98-11, SZ-95-7, QM 301 and SZ 9314 had seed cotton yields above the grand mean. SZ-95-7 (1466 kg/ha) and 644-98-11(1384 kg/ha) gave highest yields and had good GOT % as well. 644-98-11 had a high earliness index indicating early maturity. 644-98-11 and SZ-95-7 have since been tested for distinctiveness, uniformity and stability (DUS) by the seed certifying authority in Zimbabwe before release of proposal considerations.

Ideal test sites observed are Chisumbanje, CRI and Wozhele while discriminating and environments observed are Chitekete, Kuwirirana and Muzarabani. Kuwirirana is highly discriminating but not representative.

## Recommendations

1. The study shows outstanding genotypes that can be registered for upland cotton cultivation although future work can focus on testing their performances across a wider region.
2. There is significant G x E interaction in cotton yield trials and hence this should be exploited by ensuring that more METs trials are conducted to see the effect of the different seasons and locations.
3. High yielding and stable genotypes identified are 644-98-11and SZ 95-7 and these should be used to improve seed cotton yield in other cultivars through incorporating them in hybridization and or backcross programs. The genotypes can also be used in cotton improvement programs in future as well.
4. Fibre testing should be conducted so as to assess their fibre quality parameters.

## Conflict of interests

The authors have declared that there is no conflict of interests.

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*Full Length Research Paper*

# Effect of weed control methods on weed density and maize (*Zea mays* L.) yield in west Shewa Orimia, Ethiopia

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**Field experiments were conducted during 2013-2014 crop seasons at Ambo and Guder to study the effect of weed control methods on weed dynamics in maize (*Zea mays* L.) variety BH-660 in randomized complete block design with three replications. Five treatments, including Nicosulfuron (Arrow 75 WDG) at 0.09 kg ha$^{-1}$ + silwet gold (adjuvant) at 0.10%, s-metolachlor 290 + Atrazine (Primagram) at 3.00 kg ha$^{-1}$, s-metolachlor (dual gold) 1.5 kg ha$^{-1}$, and hand weeding and weedy check (control) were used. Effect of different herbicides on weed density was significant. The lowest weed density (0.71 and 4.99 m$^{-2}$) was recorded in plot treated with hand weeding followed by Nicosulfuron at 0.09 kg ha$^{-1}$ (3.68 and 5.92 m$^{-2}$) whereas the maximum was recorded in weedy check (14.16 and 24.24 m$^{-2}$) in Guder and Ambo, respectively. Like density and dry weight of weeds, the minimum was observed in hand weeding and hoeing followed by Nicosulfuron at 0.09 kg ha$^{-1}$ which is not significantly different from s-metolachlor at 1.50 kg ha$^{-1}$ and the lowest dry weight of weeds (0.0 and 26.67 gm$^{-2}$) was recorded in plot treated with hand weeding followed by Nicosulfuron at 0.90 kg ha$^{-1}$ (2.13 and 65.60 gm$^{-2}$), however, non-significant difference existed among them in Guder, whereas the highest was observed in weedy check (170.93, 382.13 gm$^{-2}$) in Guder and Ambo, respectively. Moreover, those treatments also significantly increased the yield and yield component of maize in both locations.**

**Key words:** Weed, weed control methods, herbicides, maize yield.

## INTRODUCTION

In Ethiopia, maize has been selected as one of the national commodity crops to satisfy the food self-sufficiency program of the country, to feed the alarmingly increasing population because maize has a great promise for higher yield and easier cultivation than any other cereal crop and if managed properly can go a long way in increasing food production in Ethiopia. Unfortunately and despite its great yield potential, the average maize grain yield (2.29 tons ha$^{-1}$)

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**Table 1.** Description of treatment used in the experiment.

| Chemical name | Trade name | Dosage | Time of application |
|---|---|---|---|
| Nicosulfuron + silwet gold (adjuvant) at 0.10 % | Arrow 75WDG | 0.90 kg $ha^{-1}$ | Post emergence |
| s-Metolachlor | Dual Gold | 1.50 kg $ha^{-1}$ | Pre emergence |
| Primagram | Primagram Gold 660EC | 3.00 kg $ha^{-1}$ | Pre emergence |
| Hand weeding and hoeing | - | - | Post emergence |
| Weedy check | - | - | - |

in Ethiopia (CSA, 2010) is still less than that of the yield (5.14 tons $ha^{-1}$) in other important maize-growing countries of the world (http://faostat.fao.org/site). Weed infestation is of supreme importance among biotic factors that are respon-sible for low maize grain yield. Worldwide maize production is hampered up to 40% by competition from weeds which are the most important pest group of this crop (Oerke and Dehne, 2004). Generally, weeds reduce crop yields by competing for light, nutrients, water and carbon dioxide as well as interfering with harvesting and increasing the cost involved in crop production. Kebede (2000) reported that most farmers in Ethiopia commonly lose up to 40% of yield in maize due to weed infestations. Weeds not only cause severe crop losses but also require farmers and their families to spend a considerable amount of their time on weeding. More than 50% of labor time is devoted to weeding, and is mainly done by the women and children in the farmer's family (Ellis-Jones et al., 1993; Akobundu, 1996).

Control of weeds in maize is, therefore, very essential for obtaining good harvest. Weed control practices in maize resulted in 77 to 96.7% higher grain yield than the weedy control (Khan et al., 2003). Different weed control methods have been used to manage the weeds but mechanical and chemical methods are more frequently used for the control of weeds than any other control methods. Mechanical methods including hand weeding are still useful but are getting expensive, laborious and time-consuming. Chemical control is a better alternative to manual weeding because it is cheaper, faster and gives better control (Chikoye et al., 2002, 2004). Weed control in maize with herbicides has been suggested by researchers (Correa et al., 1990; Owen et al., 1993). Ali et al. (2003) also reported that herbicides significantly increased maize yield and decreased the weed density. Therefore, the present research work was carried out to evaluate the effect of different weed control methods on weeds and yield and yield components of maize and to assess economics of herbicides under field conditions at Guder of Toke Kutaye and Ambo district, West Shoa, Ethiopia.

## MATERIALS AND METHODS

The field experiment was conducted at two different areas, Guder and Ambo in West Showa, Ethiopia during the main cropping season of 2013. Guder and Ambo district has total geographical area of 78887 $km^2$ and are located at 8° 57' North latitude and 38° 07' East longitude at an average elevation of 1800-2300 m. a. s. l. The annual rainfall ranges from 1000 -1588.06 mm and the temperature of the district ranged between 9.4 and 21.9°C with average of 15.7°C. The soil of the experimental site is light red in color (Guder), clay loam (Ambo) in texture and with pH value of 6.8.

The field experiment consisted of five treatments, S-metolachlor 290 + Atrazine (Primagram) at 3 kg $ha^{-1}$, s-metolachlor (dual gold) at 1.5 kg $ha^{-1}$, Nicosulfuron (Arrow 750 WDG) at 0.09 kg $ha^{-1}$ + silwet gold (adjuvant) at 0.10%, hand weeding and hoeing at 30 days after sowing and weedy check (no weed management) plot were carried out and arranged in a randomized complete block design with three replications. Herbicides were applied at 2 days after sowing as pre-emergence and 30 days after planting for post emergence with backpack sprayer with the spray volume of 600 L of water per hectare (Table 1). The size of each plot was 1.5 x 2.4 m. The distance between adjacent replications (blocks) and plots were 1 and 0.5 m, respectively.

The experimental plots were ploughed twice by oxen to prepare and plots were leveled manually before the field layout was made. Variety BH-660 was used as a planting material. The maize seeds were planted manually in the month of May. At planting, two maize seeds were placed in each hole, at approximately 5 cm depth. The plants were thinned to one plant per hill 20 days after sowing. The recommended amount of 100 kg $ha^{-1}$ urea and 100 kg $ha^{-1}$ DAP as source of nitrogen and phosphorus was applied. Half of nitrogen and of all the phosphorus were drilled in rows at the time of sowing. The remaining half of the N was applied at knee high growth stage of the plant (30 days after planting).

Weed population was counted with the help of quadrate thrown randomly at three places in each plot at 45 days after planting. The weeds were categorized/classified into broadleaved, grasses and sedges and converted in to area of $m^2$. The total aboveground weed dry matter was also recorded from the above thrown quadrates after cutting weeds from the ground level and then oven dried at 70°C temperature till a constant weight and was converted to $m^2$. Weed control efficiency (WCE) was determined using the following formula:

$$WCE = \frac{WDC - WDT}{WDC} \text{X} 100$$

Where, WDC = weed dry matter in weedy check, WDT = weed dry matter in a treatment

Plant height (cm), ear length (cm), ear diameter and number of cobs per plant were measured from eight randomly selected (pre tagged) plants in the middle four rows of each plot. Thousand kernels were counted from each plot and their weight was recorded. The final gran yield was measured and adjusted to 12.5% moisture content using the formula:

$$Adjusted grain yield\left(kgha^{-1}\right) = \frac{Actual yield \text{X} 100 - M}{100 - D}$$

**Table 2.** Weed floral composition of at Guder and Ambo experimental site.

| Guder | | Ambo | |
|---|---|---|---|
| **Botanical name** | **Family name** | **Botanical name** | **Family name** |
| *Amaranthus hybridus* L. | Amaranthaceae | *Amaranthus hybridus* L. | Amaranthaceae |
| *Commelina banghalensis* L. | Commelineae | *Bidens biternate* | Asteraceae |
| *Corrigiola capensis* L. | Caryophyllaceae | *Canyz aboniersis* | Asteraceae |
| *Cynodon dactylon* L. | Poaceae | *Datura stramorium* | Solanaceae |
| *Cyperus esculentus* L. | Cyperaceae | *Digitaria abysinca*. | Poaceae |
| *Cyperus rotundus* L. | Cyperaceae | *Erucastrum arabicum* Fisch and May | Brassicaceae |
| *Erucastrum arabicum* Fisch and May | Brassicaceae | *Galinsoga parviflora* cav. | Asteraceae |
| *Galinsoga parviflora* cav. | Asteraceae | *Ipomoea ariocarpa* | Convolvulaceae |
| *Oxalis comiculate*L. | Oxalidaceae | *Launaea cornuta* | Asteraceae |
| *Oxalis latifolia* L. | Oxalidaceae | *Oxalis comiculate*L. | Oxalidaceae |
| *Polygonum nepalense* Meisn | Polygonaceae | *Polygonum nepalense* Meisn | Polygonaceae |
| | | *Tribulu sterrestris* | Convolvulaceae |

**Table 3.** Effect of different herbicides on density and dry weight of weeds.

| Treatment | Density of weeds (weeds $m^{-2}$) | | Dry weight of weeds($gm^{-2}$) | |
|---|---|---|---|---|
| | **Guder** | **Ambo** | **Guder** | **Ambo** |
| Nicosulfuron at 0.09 kg $ha^{-1}$ | 3.68(13.33)$^{d}$ | 5.92(34.67)$^{c}$ | 2.13$^{bc}$ | 65.60$^{c}$ |
| s-metolachlor 1.50 kg $ha^{-1}$ | 5.45(29.33)$^{b}$ | 12.87(168.00)$^{b}$ | 21.33$^{bc}$ | 105.07$^{b}$ |
| Primagram 3.00 kg $ha^{-1}$ | 4.65(21.33)$^{c}$ | 11.99(144.00)$^{b}$ | 26.67$^{bc}$ | 93.33$^{b}$ |
| Hand weeding and hoeing | 0.71 (0.00)$^{e}$ | 4.90(24.00)$^{c}$ | 0.00$^{c}$ | 26.67$^{d}$ |
| Weedy check | 14.16(200.00)$^{a}$ | 24.24(589.33)$^{a}$ | 170.93$^{a}$ | 382.13$^{a}$ |
| LSD (0.05) | 0.5 | 2.8 | 25.9 | 26.2 |
| CV | 4.6 | 12.4 | 31.1 | 10.3 |

Figures or numbers in the parenthesis are original value, LSD = least significant difference, CV = coefficient of variation.

Where, M is the measured moisture content in grain and D is the designated moisture content. Relative crop yield loss was calculated using:

$relativeYieldloss=\frac{MY-YT}{MY}\text{X}100$, Where, MY = maximum yield from a treatment, YT = yield from a particular treatment.

Weed density was subjected to square root transformation $\left(\sqrt{(X+0.5)}\right)$to have normal distribution. Data were subjected to the analysis of variance. Mean separation was conducted for significant treatment means using least significance differences (LSD) at 5% probability level using SAS computer software version 9.1.

## RESULTS AND DISCUSSION

### Weed floral composition of the experimental sites

The experimental site at Ambo was infested with 12 different weed species belonging to 8 different families. Out of the total weeds, 91.7% were broadleaved weeds whereas the remaining 8.3% were grasses weeds (Table 2). This indicated that indicating a species-rich weed community in the experimental field. Similarly at Guder, 10 weeds species belonging to 9 families were identified. Out of the total weeds 70% were broadleaved weeds whereas the remaining 10 and 20% were grasses and sedges weeds, respectively (Table 2).

### Density and dry weight of weeds

Effect of different weed control methods on weed density both at Ambo and Guder were significant ($p<0.05$). As shown in Table 3, the lowest weed density (0.71 and 4.99 $m^{-2}$) was recorded in plot treated with hand weeding followed by Nicosulfuron at 0.09 kg $ha^{-1}$ (3.68, 5.92 $m^{-2}$) whereas the maximum was recorded in weedy check (14.16 and 24.24 $m^{-2}$) in Guder and Ambo, respectively. Similar finding was reported by Mehmeti et al. (2012) who found highest weed density in weedy check.

The weed control methods significantly affected the dry weight of weeds at both locations ($p<0.05$). The lowest

**Table 4.** Effect of different herbicides on weed control efficiency.

| Treatment | WCE (%) | |
|---|---|---|
| | Guder | Ambo |
| Nicosulfuron at 0.09 kgha$^{-1}$ | 98.8$^{a}$ | 83.0$^{b}$ |
| s-metolachlor 1.50 kgha$^{-1}$ | 87.1$^{b}$ | 72.5$^{c}$ |
| Primagram 3.00 kgha$^{-1}$ | 83.9$^{b}$ | 75.5$^{c}$ |
| Hand weeding and hoeing | 100.0$^{a}$ | 93.0$^{a}$ |
| Weedy check | 0.0$^{c}$ | 0.00$^{d}$ |
| LSD ( 0.05) | 7.9 | 4.1 |
| CV | 5.7 | 3.4 |

LSD = Least significant difference, CV = coefficient of variation.

dry weight of weeds (0.0, 26.67 gm$^{-2}$) was recorded in plot treated with hand weeding followed by Nicosulfuron at 0.90 kg ha$^{-1}$ (2.13 and 65.60 gm$^{-2}$). No significant differences existed among treatments in Guder, whereas the highest was observed in weedy check (170.93, 382.13 gm$^{-2}$) in Guder and Ambo, respectively. These results are in agreement with those reported by Hassan et al. (2010) who reported reduced weed biomass due to use of selective pre-emergence and post emergences herbicides for controlling different maize weed species.

## Weed control efficiency

Weed control efficiency at both locations was also significantly affected (p< 0.05). As described in Table 4 in Guder, the minimum weed control efficiency was observed in weedy check (0.00%) whereas the highest (100.0%) was recorded in a plot treated with hand weeding and hoeing which was not significantly different from Nicosulfuron at 0.90 kg ha$^{-1}$ (98.8). Similarly, in Ambo the maximum weed control efficiency (93.0%) was recorded in hand weeding and hoeing followed by Nicosulfuron at 0.90 kg ha$^{-1}$ (82.0), whereas the minimum was in weedy check (0.0%) This result further indicated that herbicides are more effective in reducing density and dry weights of weeds when compared with hand weeding and hoeing which are more effective than weedy check. This result was in accordance with Mehmeti et al. (2012) who reported that herbicides reduced the weed infestation in maize in comparison with the control plots.

## Yield and yield components

All the weed control treatments proved significantly superior to weedy check with respect to yield attributes and yield of maize. At Guder, cob number per plant, ear length and diameter were significantly affected by weed control methods, whereas plant height was not (p< 0.05). The maximum number of cobs per plant (1.9) was observed in hand weeding and hoeing followed by Nicosulfuron at 0.90 kgha$^{-1}$ (1.8); the lowest was recorded in weedy check (0.47). Similarly at Ambo site, weed control methods significantly affected the yield component of maize (p< 0.05).

Weed control methods also significantly affected the ear length and ear diameter of maize at both locations. The highest ear length (16.3, 19.2 cm) was in hand weeding and hoeing which was not statistically different from Nicosulfuron at 0.90 kg ha$^{-1}$, s-metolachlor 1.50 kg ha$^{-1}$ and Primagram 3.00 kg ha$^{-1}$, whereas the lowest was recorded from weedy check (12.1, 12.9cm) in Guder and Ambo, respectively. Hundred kernel weigh, grain yield and relative yield losses were significantly affected by weed control methods. The highest thousand kernel weight was recorded with hand weeding (45.33, 49.7 g) whereas the lowest was recorded with weedy check (33.8, 29.8 g) in Guder and Ambo, respectively. These results are in accordance with work of Patel et al. (2006) who stated that all the weed control treatments proved significantly superior to weedy check with respect to yield attributes and yield of maize.

Maximum grain yield (6989.8, 7223.1 kg ha$^{-1}$) was recorded in plots treated with hand weeding and hoeing and Nicosulfuron at 0.90 kg ha$^{-1}$ (6883.3, 6883.3 kg ha$^{-1}$). The lowest was recorded in weedy check (2312.4, 2612.4 kg ha$^{-1}$) in Guder and Ambo, respectively (Table 5). The efficiencies of various chemicals and other weed control practices in enhancing grain yield have previously been observed by Toloraya et al. (2001). The highest relative yield loss (63.7 and 75.7%) was recorded from weedy check whereas the lowest relative yield losses was observed from hand weeding and hoeing (0.0, 0.0%,) followed by Nicosulfuron at 0.90 kg ha$^{-1}$ (4.7, 6.3%) in Guder and Ambo, respectively (Table 6). All yield and yield parameter of maize were best in weed control methods as compared to weed control (check), this may be due to lowest weed density and dry weight.

## Conclusion

The results from both locations suggest that the density and dry weight of weeds was lower in hand weeding and hoeing followed by Nicosulfuron at 0.09 kg ha$^{-1}$, whereas the maximum was recorded in weedy check in both locations. Weed control efficiency was also high in these treatments. Like density, the minimum dry weight of weeds was observed in hand weeding and hoeing followed by Nicosulfuron at 0.09 kg ha$^{-1}$. Moreover, those treatments also increased the yield and yield component of maize in both locations.

## Conflict of interests

The author(s) have declared that there is no conflict of interests.

**Table 5.** Effect of different herbicides on plant height, ear length and diameter in Guder and Ambo.

| Treatments | Guder | | | | Ambo | | | |
|---|---|---|---|---|---|---|---|---|
| | pH (cm) | Cobs /plant | EL (cm) | ED (cm) | pH (cm) | Cobs / plant | EL (cm) | ED (cm) |
| Nicosulfuron at 0.90kgha$^{-1}$ | 150.5$^{a}$ | 1.87$^{a}$ | 18.0$^{a}$ | 7.1$^{b}$ | 175.5$^{ab}$ | 1.9$^{a}$ | 19.5$^{a}$ | 7.1$^{b}$ |
| s-metolachlor 1.50 kgha$^{-1}$ | 148.0$^{a}$ | 1.20$^{b}$ | 17.1$^{ab}$ | 7.1$^{b}$ | 160.7$^{ab}$ | 1.4$^{b}$ | 18.8$^{b}$ | 7.2$^{b}$ |
| Primagram 3.00 kgha$^{-1}$ | 157.0$^{a}$ | 1.33$^{b}$ | 16.8$^{ab}$ | 7.2$^{b}$ | 175.5$^{ab}$ | 1.5$^{ab}$ | 19.2$^{a}$ | 7.1$^{b}$ |
| Hand weeding and hoeing | 152.7$^{a}$ | 1.93$^{a}$ | 16.3$^{ab}$ | 8.2$^{a}$ | 179.1$^{a}$ | 1.9$^{a}$ | 19.7$^{a}$ | 8.1$^{a}$ |
| Weedy check | 147.9$^{a}$ | 0.47$^{c}$ | 12.2$^{c}$ | 6.5$^{b}$ | 144.3$^{b}$ | 0.8$^{c}$ | 12.9$^{a}$ | 6.1$^{c}$ |
| LSD ( 0.05) | NS | 0.3 | 2.3 | 0.8 | 31.4 | 0.4 | 1.9 | 0.8 |
| CV | 3.4 | 11.7 | 7.2 | 5.9 | 10.0 | 15.1 | 5.6 | 5.8 |

LSD = Least significant difference, CV = coefficient of variation, EL = ear length, ED = ear diameter, PH = plant height.

**Table 6.** Effect of herbicides on 100 kernel weight, grain yield and relative yield loss.

| Treatments | Guder | | | Ambo | | |
|---|---|---|---|---|---|---|
| | HSW (g) | GY (kg ha$^{-1}$) | RYL (%) | HSW (g) | GY (kg ha$^{-1}$) | RYL (%) |
| Nicosulfuron at 0.90 kgha$^{-1}$ | 41.53$^{a}$ | 6883.3$^{a}$ | 4.737$^{cd}$ | 44.667$^{b}$ | 6883.3$^{ab}$ | 6.314$^{d}$ |
| s-metolachlor 1.50 kgha$^{-1}$ | 42.633$^{a}$ | 5026.4$^{b}$ | 30.15$^{b}$ | 41.167$^{c}$ | 5026.4$^{c}$ | 29.368$^{b}$ |
| Primagram 3.00 kgha$^{-1}$ | 42.833$^{a}$ | 6159.2$^{a}$ | 14.519$^{c}$ | 41.30$^{c}$ | 6159.2$^{b}$ | 11.803$^{c}$ |
| Hand weeding and hoeing | 45.333$^{a}$ | 6989.8$^{a}$ | 0.000$^{cd}$ | 49.667$^{a}$ | 7223.1$^{a}$ | 0.00$^{e}$ |
| Weedy check | 33.80$^{b}$ | 2312.4$^{c}$ | 63.655$^{a}$ | 29.80$^{d}$ | 2612.4$^{d}$ | 75.712$^{a}$ |
| LSD (0.05) | 5.19 | 921.28 | 9.79 | 3.29 | 812.36 | 5.32 |
| CV | 6.68 | 8.84 | 23.01 | 4.24 | 7.73 | 11.47 |

LSD = Least significant difference, CV = coefficient of variation, HSW = hundred seed weight, GY = grain yield, RYL = relative yield loss.

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*Full Length Research Paper*

# Distribution of kolanut weevil (*Balanogastris kolae*) (Coleoptera:Curculionidae) in *Cola nitida* stored in baskets

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**The kolanut weevil, *Balanogastris kolae* is usually referred to as a field-to-store pest as their infestation starts in the field and continues in storage. The distribution preferences of the weevil were investigated with a view of determining their vertical and horizontal distribution in storage baskets. The kola pods used for this experiment were obtained from kola groves at the Cocoa Research Institute of Nigeria (CRIN), Ibadan. After harvesting the pods, the nuts were extracted, skinned, cured and stored in baskets lined with banana leaves for 2 weeks (according to the traditional methods) before transferring them to experimental baskets. The weevils recorded in basket A were 74% which was significantly different ($p < 0.05$) from the 16.5% in basket B and 9.5% in C. The horizontal distribution at the peripheral section (D) of the basket was 70.1%. This was significantly ($p < 0.05$) higher than the distribution at the core section (E), which was 29.9%. Therefore in the storage baskets, 74.0% of adult weevils exhibited positive geotaxis while 70.1% had affinity for lateral distribution. Farmers should therefore concentrate more at the bottom section of the basket during regular sorting and removal of weevils and infested nuts from kolanuts in storage baskets.**

**Key words:** Kolanuts, weevil, exposure, distribution, baskets.

## INTRODUCTION

The genus *Cola*, especially *Cola nitida* (Vent.) Scott and Endl. (1832), and *Cola acuminata* Scott and Endl. (1832) are important economic crops in West and Central Africa, Carribean Islands, Mauritius, Sri Lanka and Malaysia (Eijnatten, 1969; Oladokun, 1982). Both are the only edible species of kola grown on a commercial scale.

*C. nitida* is considered to be indigenous to the forest area of Cote d' Ivoire and Ghana, where it was originally distributed along Africa's West Coast from Sierra Leone to Dahomey (Nzekwu, 1961). Cultivation of *C. nitida* in Nigeria must have started long before 1900 because by 1904, Bernegau (1908) reported established plantations at Agege and villages between Abeokuta and Lagos with trees which were estimated to be up to 30 years old. The

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**Figure 1.** Baskets used for the determination of vertical distribution of *Balanogastris kolae* in stored kolanuts.

original distribution area of *C. acuminata* stretches from Nigeria to Gabon. It is still cultivated in South Eastern Nigeria, the South South states and in the Middle Belt states. However, in Southwestern Nigeria, its cultivation has been replaced by that of *C. nitida* (Eijnatten, 1969).

Kola fruits usually mature 4-5 months after pollination; at this stage the fruit is inconspicuously brown and changes in colour from deep green to a paler tint. At this time they should be harvested, because soon afterwards the follicles will begin to dehisce. When exposed, the seeds become more prone to insect attack. It is important that harvesting should be carried out before the follicles begin to split and fall to the ground. This will guard against infestation by weevils. Harvesting should be carried out once or twice a month during the fruiting season beginning from September to the end of January. Sporadic fruiting often occurs in July and August. The peak period of production is from October to December for *C. nitida* (Ndubuaku, 2014).

The adults of *Balanogastris kolae*, (which is the most common and important of the kola weevils), are dark brown; 3 to 4 mm long and 1.5 to 2 mm wide. The female lays egg 1 cm deep in the nuts and in other parts of the fruit through wounds and holes made by other insects such as *Ceratitis colae* Silv. or through cracks on the husk created when the follicles dehisce before harvest. Incubation lasts for about 4 to 6 days. Larva stage takes 17 to 20 days and the larva feeds extensively reducing the kola to brown powdery mass. Pupation lasts for about 5 to 6 days. The larval and pupal periods take place inside the nut. The average period from oviposition to the emergency of the adult of *B. kolae* adult is 29 days. The average life span of *B. kolae* adult is 53 days with oviposition starting on the third day (Daramola, 1973, 1978). Breeding continues throughout the year on left over nuts and nuts produced in-between the main harvest seasons (Alibert and Mallamaire, 1955; Daramola, 1974). The weevil is usually referred to as a field-to-store pest as their infestation starts in the field and continues in storage.

The objective of this study therefore was to study the distribution preferences of kolanut weevil (*Balanogastris kolae*) (Coleoptera: Curculionidae) in *Cola nitida* stored in baskets.

## MATERIALS AND METHODS

The kolanuts used for this experiment were obtained from kola groves at the Cocoa Research Institute of Nigeria (CRIN), Ibadan. After harvesting the pods, the nuts were extracted, skinned, cured and stored in baskets lined with banana leaves for 2 weeks (according to the traditional methods) before transferring them to the experimental baskets.

### Vertical distribution of *B. kolae*

For the study of vertical distribution of kolanut weevils in storage baskets, the experimental set up consisted of 3 baskets, A, B, C, of the same size, each measuring 26 cm in length, 48 cm as the diameter of the mouth and 45 cm as the diameter of the bottom. Baskets B and C had their bottoms removed and replaced with false bottoms consisting of fish nets of 3.5 cm mesh. The baskets were lined with banana leaves but the bottom side of B and C were not lined to allow the weevils have free movement from one basket compartment to another. Each basket was half filled with 500 kolanuts. The baskets were stacked on each other with basket A at the base, B at the middle and C on top, such that the bottom of C fitted into the mouth of B to rest on the surface of the kolanuts in B while the bottom of B fitted into the mouth of A to rest on the surface of the kolanuts in A (Figure 1).

Twenty (20) weevils were distributed at the top of each basket just before stacking the baskets. The weevils were then left in the baskets for an exposure period of 24 h. At the end of each period of exposure, the baskets were separated and the insects in each were collected and counted to determine their distribution. The treatment was replicated four times. Results obtained were subjected to analysis of variance. Treatment means which differed significantly at $p = 0.05$ were separated using least significant difference (LSD).


**Figure 2:** Baskets used for the determination of horinzontal distribution of Balanogastris kolae in stored kolanuts.

**Horizontal distribution of *B. kolae***

The study of horizontal distribution of the weevils in storage baskets was carried out using 2 baskets D and E. Basket D is a traditional kolanuts storage weaker basket, cylindrical in shape, measuring 26 cm in height and 48 cm in diameter. Basket E is also cylindrically shaped but made of 1mm binding wire frame fitted on the side and bottom with a 35 cm mesh fish net to allow unhindered horizontal movement of the weevils. It is half the volume of basket D, measuring 26 cm in height and 23 cm in diameter. Basket D was lined with banana leaves and Basket E was placed in the middle of basket D before filling both baskets with a total of 500 kolanuts (Figure 2).

Sixty (60) weevils were evenly distributed on the kolanuts at the concentric mouths of the baskets before the baskets were covered with banana leaves for an exposure period of 24 h. At the end of each exposure period, basket E was carefully pulled out from basket D and the adult weevils in each were collected and counted to determine their distribution. The treatment was replicated 4 times. The difference between the numbers of weevils found in the different sections of the basket was compared with the 't' distribution.

**Statistical analysis**

The treatment means obtained for the vertical distribution of *B. kolae* were separated using least significant difference (LSD), while the difference between the numbers of weevils found in the different sections of the basket was compared with the 't' distribution.

## RESULTS

### Vertical distribution *B. kolae*

Table 1 shows the vertical distribution of kola weevils in the different basket compartments. The weevils recorded in basket A were 74% which was significantly different ($p < 0.05$) from the 16.5% in basket B and 9.5% in C. There was no significant difference between the mean number of the weevils in basket B and C. The calculated t was 2.276, while the observed t at $p < 0.05$ was 3.182.

### Horizontal distribution *B. kolae*

Table 2 shows the distribution of kola weevils in the two sections of the basket. The horizontal distribution at the peripheral section (D) of the basket was 70.1%. This was significantly ($p < 0.05$) higher than the distribution at the core section (E), which was 29.9%.

**Table 1.** Vertical distribution of adult kola weevils, *Balanogastris kolae* in storage baskets

| Section of basket | Number of weevils in different sections of storage baskets after exposure for 24 h | | |
|---|---|---|---|
| | Mean | Range | Percentage of total (%) |
| Top Section (C) | 6.0 | 0 – 14 | 9.5 |
| Middle section (B) | 10 | 2 - 26 | 16.5 |
| *Bottom section (A)* | 46.25 | 34 - 51 | 74.0 |

LSD (P=0.05)13.611.

**Table 2.** Horizontal distribution of adult kola weevils, *Balanogastris kolae* in storage baskets.

| Sections of basket | Number of weevils in different sections of storage baskets after exposure for 24 h | | |
|---|---|---|---|
| | Range | Mean | Percentage of total (%) |
| Outer section (D) | 30 - 51 | 39.75 | 70.1 |
| Inner section (E) | 8 - 26 | 17.0 | 29.9 |

Calculated t=2.276; Observed t (P=0.05) = 3.182

## DISCUSSION

Studies on the distribution of kola weevils in storage baskets showed that kola weevils exhibit positive geotaxis by their tendency to move downward. Youdeowei (1977) observed that the nymphs of *Zonocerus variegatus* exhibited negative phototaxis because of their habit of crawling up the branches of their host plants or a rod or piece of stick held vertically. He however observed that the reactions of insects to gravity were, however, far less known than their reactions to the other environmental stimuli. The presence of more weevils at the periphery of the basket could be due to the tendency of the adult weevils to move and fly out of the habitat (kola nut storage container) probably for dispersal purposes.

The crevices at the bottom of the storage baskets should be thoroughly inspected during the regular replacement of banana leaves or removal of infested nuts to ensure that weevils hiding at the bottom of baskets are not overlooked. Further investigations will be carried out to determine how the positive geotaxis and the tendency to move towards the periphery of the basket can be exploited in baiting kola weevils and in developing an 1PM programme for the control of kolanut weevils in storage. An effective non-insecticidal method of control of kola weevils will reduce the tendency of farmers to use hazardous chemicals for the control of kola weevils in storage.

## CONCLUSION AND RECOMMENDATION

The results obtained in this experiment can be exploited during sorting of kolanuts. It is therefore recommended that during regular sorting and removal of weevils and weeviled nuts from kola nuts in storage baskets, farmers should concentrate more at the bottom section of the basket where most of the adult weevils are located.

## Conflict of interests

The authors did not declare any conflict of interest.

## ACKNOWLEDGMENT

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*Full Length Research Paper*

# Composition and taxonomic similarity of the periphytic algal community in different natural substrates in a neotropical floodplain, Brazil

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**The present study investigated the species composition and taxonomic similarity of periphytic algae on three species of macrophytes (*Eichhornia azurea* Kunth, *Nymphaea amazonum* Martius & Zuccarini and *Oxycaryum cubense* (Poeppig & Kunth) Lye) and also some limnological variables in a lake permanently connected to the Paraná River at the Upper Paraná River floodplain, Brazil, from June 2008 to March 2009. During the study period, the Paraná River showed irregular flood pulses and indistinct hydrological periods. In this same period, 406 taxa of periphytic algae were identified, distributed mainly in the classes Zygnemaphyceae, Bacillariophyceae, Chlorophyceae; and Cyanobacteria. Similarity analysis based on taxonomic composition of sampling periods and substrates showed low values and primarily represented a temporal segregation of periphytic algal community, mainly in June 2008 from others. Secondly, the microespacial segregation occurred to a lesser extent, according to the type of substrate, especially between *O. cubense* and others. It comprises first steps for understating the comparative structure of periphytic algal community in these distinct substrates at the Paraná River floodplain.**

**Key words:** Community structure, epiphyton, macrophyte, periphyton ecology, wetlands.

## INTRODUCTION

Macrophytes consist of important centres for maintenance of the aquatic biodiversity (Mormul et al., 2010), with emphasis on periphyton, since they promote the availability of large surface area for colonization of this attached community (Algarte et al., 2009). Morphoanatomical characteristics of such substrates increase spatial heterogeneity and can determine composition, abundance, biomass and productivity of the periphyton communities(Stevenson, 1997; Hinojosa-Garro et al., 2010).

Previous studies have shown that the taxonomic composition of periphyton communities can differ in distinct

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naturalsubstrates(Jonesetal.,2000).However,thestructure and dynamics of periphyton in floodplains are mainly influenced by flood pulses. Variation of the hydrometric levels, the hydrological periods and other features related to the hydrological regime of the river and its adjacent environments can alter the species composition of periphyton, as previously revealed in studies developed in the Upper Paraná River floodplain (Rodrigues et al., 2003; Rodrigues and Bicudo, 2001; 2004; Algarte et al., 2006; 2014). Nonetheless, these studies have focused on the periphytic communities from a unique natural substrate, the macrophyte *Eichhornia azurea* Kunth.

There are only few comparative studies on the composition and periphyton similarity between different substrates at the Paraná River floodplain. In the low region of this floodplain,TesolínandTell(1996)investigated the periphytic community of four species of floating aquatic macrophytes from a connected lake in Argentina. Richness of taxa in this region is very low, with only 26 taxa recorded by these authors. At the upper portion of the floodplain, Neif et al. (2013) analyzed the periphyton structure from two macrophytes, *E. azurea* and *Egeria najas* Planch - of a lake, both submerged macrophytes. Regarding macrophytes covered in the present study, *Nymphaea amazonum* Martius & Zuccarini and *Oxycaryum cubense* (Poeppig and Kunth) Lye, knowledge of the composition and similarity of periphyton is still scarce, with previous data published related to the specific richness and density of periphytic communities in these macrophytes (Biolo and Rodrigues, 2013) as part of the major project which originated this study.

Therefore, the present study aimed to investigate the composition and taxonomic similarity of periphytic algal communities attached on three aquatic macrophytes with different growth forms (*E. azurea*, emergent; *N. amazonum*, fixed floating; and *O. cubense*, epiphyte) in a permanent connected lake at the Upper Paraná River floodplain. Since diversity of macrophytes was an important factor which influences the periphytic community in this floodplain (Murakami et al., 2009), we expect that the taxonomic composition and similarity of periphytic algae in distinct substrates will be different under similar environmental conditions.

## MATERIALS AND METHODS

### Study area and periphyton sampling

The “Pau Véio” Lake is an open lake with a permanent connection to the Paraná River, located in the Paraná River Floodplain, between the States of Paraná and Mato Grosso do Sul, Brazil (22°44'S - 53°15'W). Sampling of the periphytic community was performed quarterly between June 2008 and March 2009, comprising two hydrological periods (high water, November to May; and low water, June to October).

Natural substrates for collecting periphyton consisted of macrophyte petioles in the adult stage of the following species (and ecological groups), according to Irgang et al. (1984): *E. azurea* Kunth (emerging) and *N. amazonum* Martius & Zuccarini (floating fixed), and the stem of *O. cubense* (Poeppig and Kunth) Lye (epiphyte). In *O. cubense*, the leaf sheath involved in the region of stem was also sampled.

Selection of substrates was done as follows: their presence in a same bank, presence of multi-species under similar environmental conditions, and in all sampling periods. In addition to presenting similar morphostructural characteristics, we attempted to standardize sampling methodologies (which could be equally applied to all substrates according to their morphology). We also aimed to supply the lack of studies of the periphytic community encompassing the last two substrates cited in the Paraná River floodplain.

Substrates collected consisted of replicates (n=2). For removal of the periphytic community of substrates, a steel blade coated on an aluminum sheet with the aid of jets of distilled water was used. Material designated to qualitative analysis was fixed in Transeau solution. Periphytic algae were identified under optical microscope based on classical and regional bibliographies.

### Abiotic variables sampling

Abiotic variables were simultaneously measured during the collection of biological material and corresponded to: water temperature and dissolved oxygen (oximeter YSI model 55 laptop brand), pH (portable pH meter model Digimed DM2), electrical conductivity (Conductivity Digimed laptop model DM2), alkalinity (Carmouze, 1994), transparency of the water column (Secchi disk), turbidity (portable turbidimeter model Lamotte), total solids, organic and inorganic fractions (Wetzel and Likens, 1991), total nitrogen and nitrate (Bergamin et al., 1978; Giné et al., 1980), ammonia nitrogen (Mackereth et al., 1978), and total phosphorus (Mackereth et al., 1978) and phosphate (Mackereth et al., 1978). For analysis of the fraction of dissolved nutrients and suspended solids determination, we filtered samples using Whatman GF-C 52 filters (Golterman et al., 1978). Data of the hydrometric level of Paraná River were obtained by the measurement of the rule relating to the São José Port, Paraná. Abiotic data were ceded by the Laboratory of Limnology, at NUPELIA (“Núcleo de Pesquisas em Limnologia Ictiologia e Aquicultura”) and other details about the sampling methodology are shown in Roberto et al. (2009).

### Data analysis

The species composition of the periphytic algae was evaluated based on the similarity of communities between different natural substrates (*E. azurea, N. amazonum* and *O. cubense*) and sampled months (June, September and November 2008 and March 2009). This attribute was measured by cluster analysis using criterion of presence and absence of species by Jaccard index (determination of index of similarity between communities and the index of species association) consistent with the coefficient of cophenetic correlation (e.g., the Pearson correlation coefficients between the elements of the dissimilarity matrix and the elements of cophenetic matrix). Data was analysed through ANOSIM method with 999 permutations (similarities between two or more groups of sampling units (factors) were compared), resulting in a statistic R which ranges between -1 (similar) and +1 (dissimilar) (Clarke and Gorley, 2001). All analyses were performed by the R statistical software version 3.0.0 (R Core Team, 2013).

## RESULTS

Abiotic data in the Pau Véio Lake analyzed during the study period are shown in Table 1 and the hydrometric

**Table 1.** Abiotic data from the Pau Véio lake, at the Upper Paraná River floodplain, in the period of study June 2008 to March 2009 (Biolo and Rodrigues, 2013).

| Parameter | June | September | November | March |
|---|---|---|---|---|
| Temperature (°C) | 19.4 | 20.9 | 27.1 | 28.5 |
| Dissolved oxygen (mg.$L^{-1}$) | 6.15 | 4.31 | 2.59 | 5.22 |
| pH | 6.83 | 6.55 | 6.62 | 6.91 |
| Conductivity (µS.$cm^{-1}$) | 56.7 | 59.3 | 59.9 | 58.8 |
| Alkalinity (µEq $L^{-1}$) | 468 | 457.5 | 387.2 | 410.4 |
| Mean hydrometric level (m) | 2.95 | 2.55 | 2.39 | 3.16 |
| Transparency (Secchi) (m) | 3.1 | 2.2 | 2 | 2.25 |
| Turbidity (NTU) | 3.33 | - | 2.28 | 3.63 |
| Total solid material (µg.$L^{-1}$) | 2.1 | 0.6 | 0.75 | 1.88 |
| Total nitrogen (µg.$L^{-1}$) | 227.5 | 368.1 | 495.2 | 1000.9 |
| Nitrate (µg.$L^{-1}$) | 135.8 | 97.9 | 45.8 | 120.7 |
| Ammoniacal nitrogen ($NH_4^+$) | 4.9 | 2.6 | 19.3 | 7.26 |
| Total phosphorus (µg.$L^{-1}$) | 13.2 | 12.1 | 18.6 | 20.6 |
| Orthophosphate (µg.$L^{-1}$) | 4.9 | 3.7 | 13.8 | 5.5 |

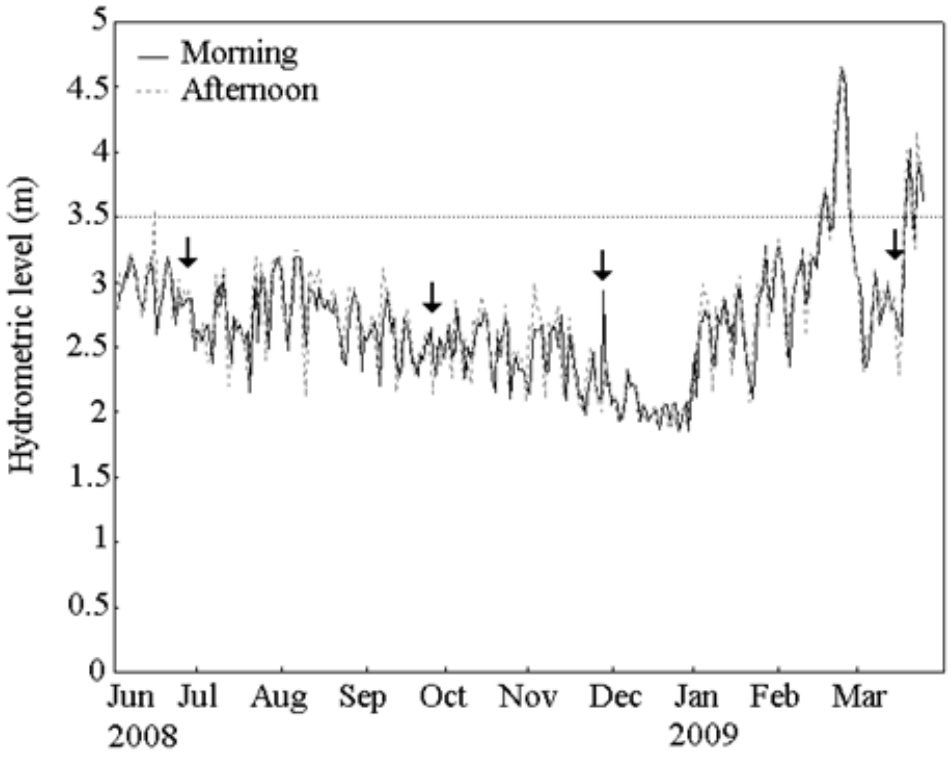


**Figure 1.** Hydrometric level at the Paraná River, at the Upper Paraná River floodplain in the period of study, June 2008 to March 2009 (Biolo and Rodrigues, 2013).

level of the Paraná River floodplain in Figure 1. The year 2008 was irregular in respect of the hydrological periods (high water and low water, with the prevalence of flood pulses with low intensity and low values of hydrometric levels throughout the year (Roberto et al., 2009; Biolo and Rodrigues, 2013). In 2009, floodpulses were more intense and hydrometric levels reached peaks above the level of overflow in February 2009 (between 3.53 and 4.65 m), characterizing the high water period of the floodplain.

The species composition of the periphytic algal community present in the three macrophytes and months

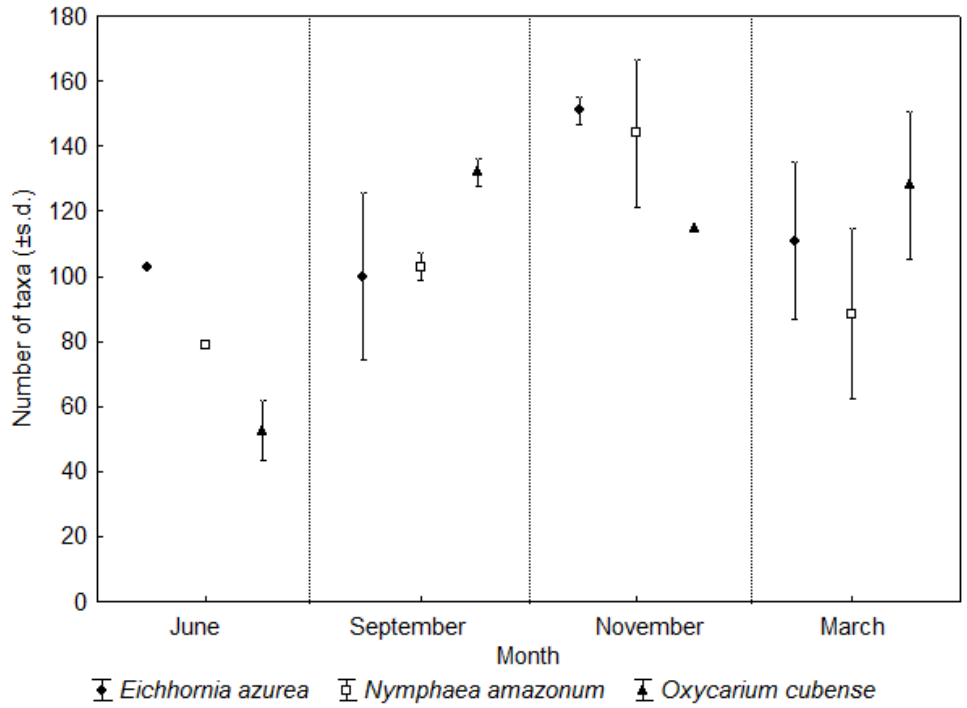


**Figure 2.** Number of periphytic algal taxa for each substrate and sampling period in the "Pau Véio" lake, at the Paraná River floodplain.

totaled 406 taxa belonged to 139 genera and 11 classes: Zygnemaphyceae (114), Bacillariophyceae (86), Chlorophyceae (66), Xanthophyceae (24), Euglenophyceae (23), Chrysophyceae (14), Oedogoniophyceae (9), Cryptophyceae (7), Rhodophyceae (3), Chlamydophyceae (1) and Dinophyceae (1); and Cyanobacteria (58). Figure 2 shows the distribution of main groups over the sampled periods and substrates.

Some species were present in all periods and substrates and corresponded to: *Achnanthidium minutissimum* (Kützing) Czarnecki, *Cymbella affinis* Kützing, *Encyonema mesianum* (Cholnoky) D. G. Mann, *Eunotia intermedia* (Krasske) Nörpel and Lange-Bertalot, *Fragilaria capucina* Desmazières, *Fragilaria tenera* (W.Smith) Lange-Bertalot, *Gomphoneis clevei* (Fricke) Gil, *Gomphonema brasiliense* Grunow, *Gomphonema gracile* Ehrenberg, *Nitzschia linearis* W. Smith, *Nitzschia palea* (Kützing) W. Smith and *Ulnaria ulna* (Nitzsch) P.Compère (Class Bacillariophyceae), *Desmodesmus brasiliensis* (Bohlin) E. Hegewald (Class Chlorophyceae), *Aphanocapsa parasitica* (Kützing) Komárek and Anagnostidis, *Leibleinia epiphytica* (Hieronymus) Compère and *Leptolyngbya perelegans* (Lemmermann) Anagnostidis & Komárek (Cyanobacteria), *Oedogonium* sp. (Class Oedogoniophyceae) and *Tetraedriella* cf. *jovetii* (Bourrelly) Bourrelly (Class Xanthophyceae).

Differences in composition of the periphytic algal communities from substrates and sampled periods were summarized by similarity dendrogram based on the Jaccard Similarity Index (r = 0.787, coefficient of cophenetic correlation) (Figure 3) and ANOSIM (Figure 4). The similarity coefficients ranged from 0.50 to 0.65, indicating a low similarity flora; similarity differences showed by ANOSIM suggest substantial dissimilarities in composition of the periphytic algal community between periods (R = 0.574, p = 0.002), but not between substrates (R = -0.183, p = 0.935). Firstly, a temporal division of periphytic communities in two large clusters was observed (Figure 3, Group I), related to the sampled period (June 2008 from the other). Segregation of periphytic communities mainly between September 2008 (Figure 3, Group II) and the months November 2008 and March 2009 (Figure 3, Group III) were observed. There was an apparent separation of periphytic algal communities related to the type of substrate, mainly from *O. cubense* and the other.

## DISCUSSION

The "Pau Véio" lake was richly represented by periphytic algae in all sampled periods and substrates. The species composition of periphytic algae can indicate the abiotic conditions and the spatial and temporal heterogeneity in each environment (Rodrigues et al., 2003). Despite the fact that some dominant taxa were registered in all periods and substrates, the majority contributed to the

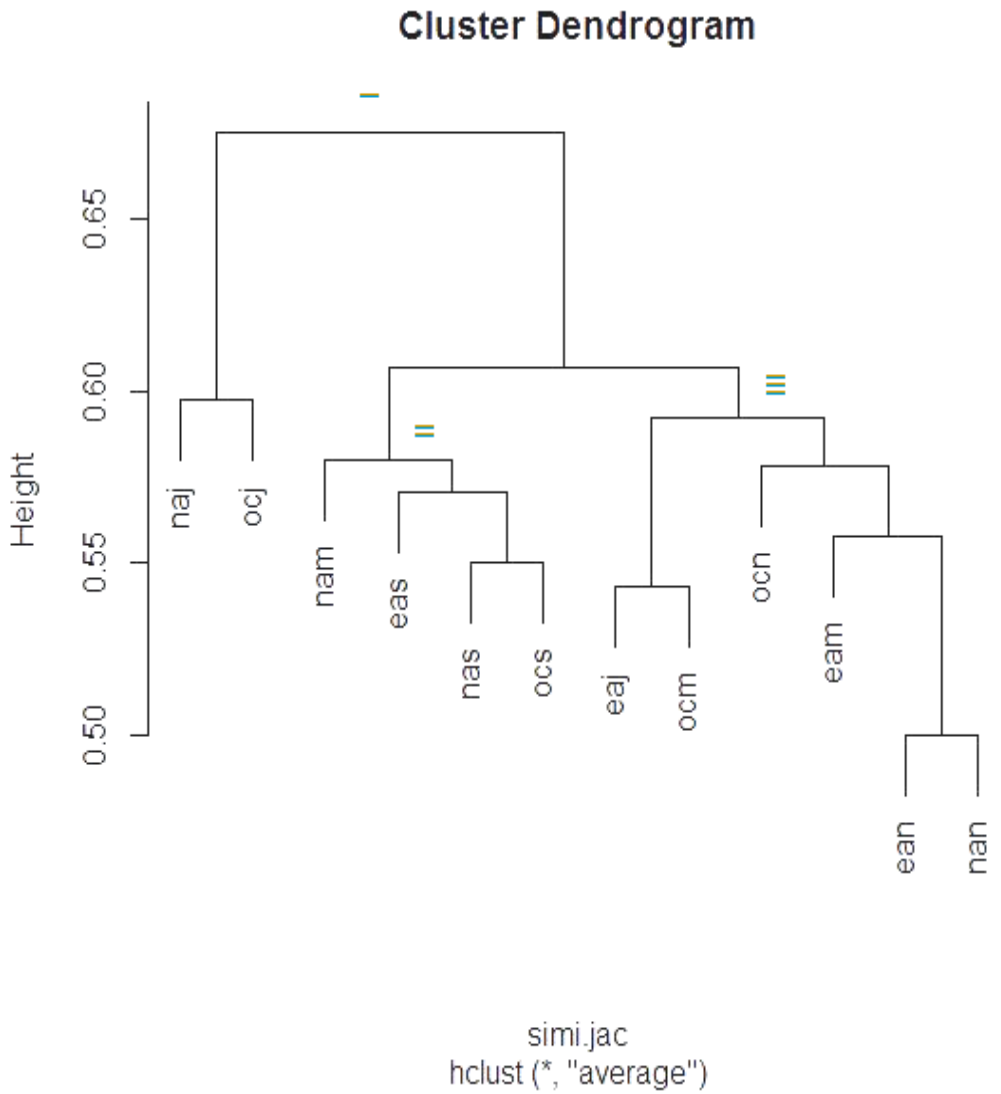


**Figure 3.** Similarity dendrogram (Jaccard Index; r = 0.787, coefficient of cophenetic correlation) from the periphytic algal community in distinct substrates (ea = *Eichhornia azurea;* na = *Nymphaea amazonum*; oc = *Oxycaryum cubense*) in the four months analysed (j = June 2008; s = September 2008; n = November 2008; m = March 2009), in the "Pau Véio" lake, at the Paraná River floodplain.

dissimilarity taxonomic between periphytic algal communities. Differences in taxonomic similarity were mainly temporal, related to different periods. Periphytic algal communities developed in June 2008 were more divergent between other (60% of dissimilarity). In June 2008, hydrometric level and temperature reached their lowest values (Biolo and Rodrigues, 2013), which were probably crucial for structuring the taxonomic composition of periphytic algal community (Wetzel, 1983; Murakami et al., 2009), by increasing dominance of r-strategists algae, as Bacillariophyceae and Cyanobacteria (Biggs, 1996). According to Leandrini et al. (2008), the absence of periods of flooding and the prevalence of low hydrometric levels are important factors that influence distribution, abundance, and biomass of organisms, especially for periphytic algae. In June 2008, when community were more divergent, intensity of floodpulses were very low and pulses were almost absent (Roberto et al., 2009; Biolo and Rodrigues, 2013).

High temperatures supported a rich periphytic flora in the Upper Paraná River floodplain (Murakami et al., 2009; Biolo and Rodrigues, 2013) and could also affect the species composition of this community after June 2008. Furthermore, increase in hydrometric levels and in degree of connectivity of lake with the main river, with the improvement of limnological conditions toward the year 2009 should have promoted greater entry of algal propagules in the environment and their establishment in the periphytic community (Biolo and Rodrigues, 2013). In consequence, composition of periphytic algal community

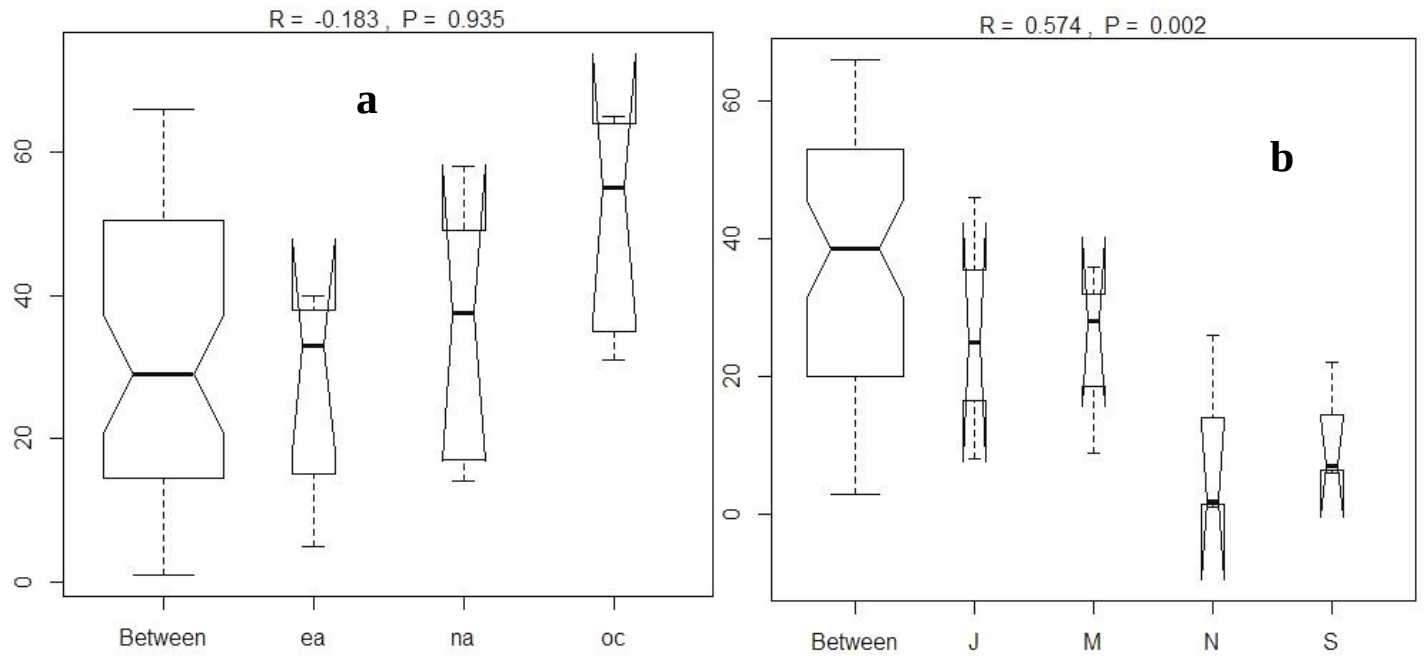


**Figure 4.** Analysis of similarity data (ANOSIM) from the periphytic algal community in (a) distinct substrates (ea = *Eichhornia azurea;* na = *Nymphaea amazonum*; oc = *Oxycaryum cubense*) and (b) in the four months analysed (J = June 2008; S = September 2008; N = November 2008; M = March 2009), in the "Pau Véio" lake, at the Paraná River floodplain.

may also have been affected.

Dissimilarities in the species composition of the periphytic algae in different substrates were less pronounced. Physical factors can structure epiphytic algal communities. Morphology and architecture of macrophytes, in addition to surface microstructure of the plant and the density of macrophyte hosts, are reflected particularly in their associated organisms (Pip and Robinson, 1981). Moreover, these conditions can favor selectivity between habitats and associated organisms (Messyasz and Kuczynska-Kippen, 2006). However, the present study showed that the type of substrate was not a critical factor for influencing segregation of periphytic algal communities on *E. azurea*, *N. amazonum*, and *O. cubense* under similar limnological conditions (in the same sampled period).

Indeed a large temporal influence that may have affected more strongly the *O. cubense* than the other two species of macrophytes was observed, which promoted less pronounced – but not less important - dissimilarities between communities in distinct substrates. Surface microtopography and petioles of macrophytes act similarly as substrates in emergent plants, as previously discussed by Laugaste and Reunanen (2005). This fact could be observed in the present study, which substrate provided by *O. cubense* presented more dissimilar community between other substrates (Figure 2). Its more complex morphology and life habit differs from other macrophytes (e.g., *E. azurea* and *N. amazonum*), because both apparently present more similar morphology of the petioles. Furthermore, leaves of *O. cubense* present a parallel innervation that can increase the spatial heterogeneity providing distinctive microhabitat for algal colonization (Souza and Ferragut, 2012).

These results are similar to those reported for the structural attribute density, however, contrary to the attribute species richness of periphytic algae on *E. azurea*, *N. amazonum* e *O. cubense* (Biolo and Rodrigues, 2013). In this study, Biolo and Rodrigues (2013) showed no significant differences reported in the mean values of specific richness between the same substrates, only for sampled periods and for interaction "time x substrate"; and significant differences between periphytic algae from distinct substrates were registered only for average values of density. Furthermore, Neif et al. (2013) also found no significant differences in the species composition and richness of periphytic algae of different submerged macrophytes at the same floodplain, including the attribute density, but only between periods. Our study provides first steps for understanding comparative structure of periphytic algal communities in *E. azurea*, *N. amazonum* and *O. cubense*.

According to discussions presented by Cattaneo and Kalff (1979) and Jones et al. (2000), there is a considerable controversy about factors that determine the species composition of the periphytic algal communities, especially with respect to the selective influence of the type and shape of substrate. For the present study, results suggest that composition and taxonomic similarity was probably temporally related to the influence of

environmental variables (abiotic variables, flood pulses and hydrometric levels), with influence in a lesser degree of the type of substrate.

Furthermore, we recommend natural substrate provided by the macrophyte *E. azurea* for future studies in periphyton ecology in this floodplain, since it could be easily found and collected in any period in the Paraná River floodplain. In latter case, this substrate may be replaced by *N. amazonum*, with morphology and attached algae more similar to the *E. azurea*.

## Conflict of interests

The authors have not declared any conflict of interest.

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