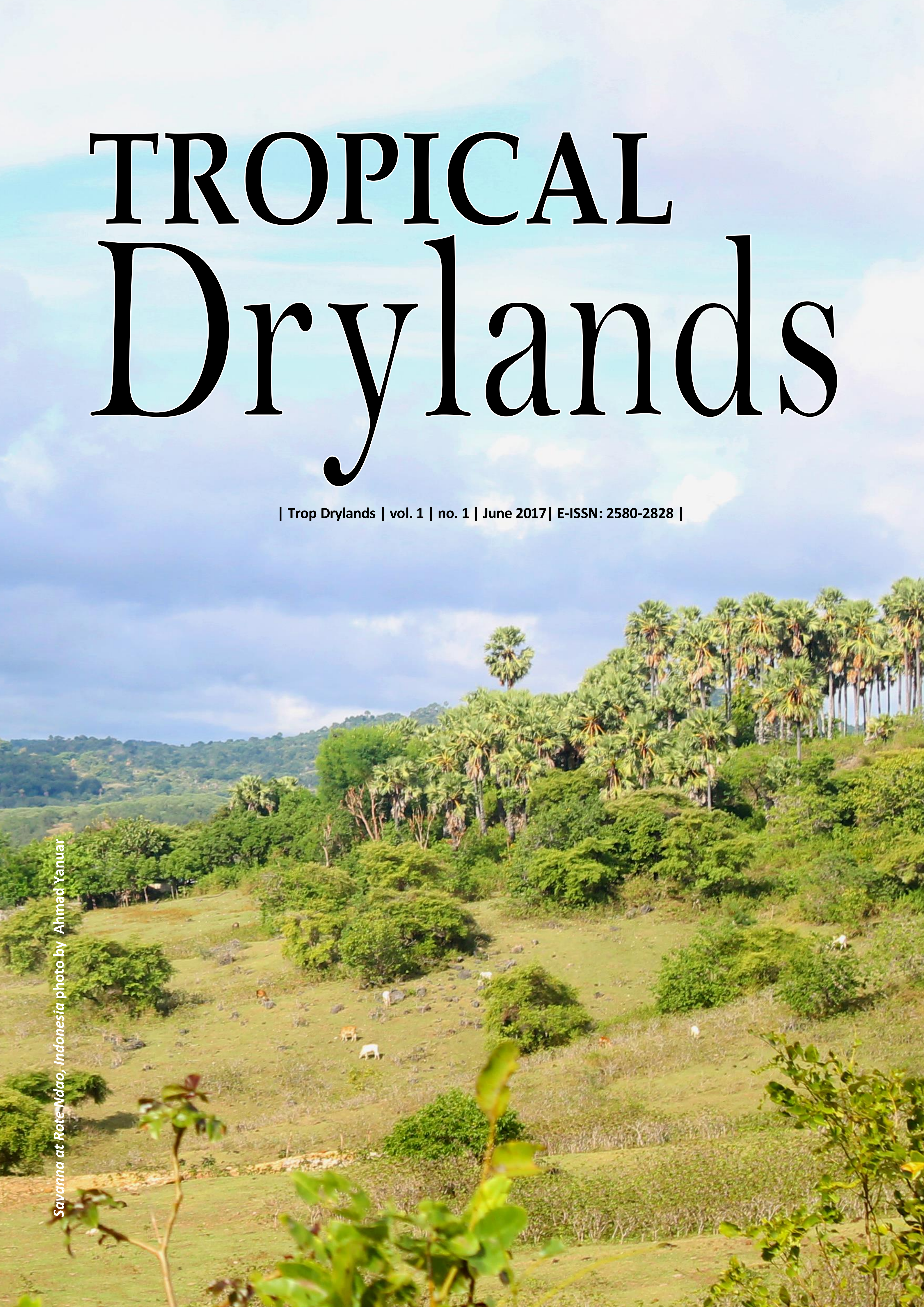

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Savanna at Rote-Ndao, Indonesia photo by Ahmad Yanuar

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Book:

Rai MK, Carpinella C. 2006. Naturally Occurring Bioactive Compounds. Elsevier, Amsterdam.

Chapter in book:

Webb CO, Cannon CH, Davies SJ. 2008. Ecological organization, biogeography, and the phylogenetic structure of rainforest tree communities. In: Carson W, Schnitzer S (eds) Tropical Forest Community Ecology. Wiley-Blackwell, New York.

Abstract:

Assaeed AM. 2007. Seed production and dispersal of *Rhazya stricta*. 50th Annual Symposium of the International Association for Vegetation Science, Swansea, UK, 23-27 July 2007.

Proceeding:

Alikodra HS. 2000. Biodiversity for development of local autonomous government. In: Setyawan AD, Sutarno (eds.) Toward Mount Lawu National Park; Proceeding of National Seminary and Workshop on Biodiversity Conservation to Protect and Save Germplasm in Java Island. Universitas Sebelas Maret, Surakarta, 17-20 July 2000. [Indonesian]

Thesis, Dissertation:

Sugiyarto. 2004. Soil Macro-invertebrates Diversity and Inter-Cropping Plants Productivity in Agroforestry System based on Sengon. [Dissertation]. Universitas Brawijaya, Malang. [Indonesian]

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On-farm phenotypic characterization of indigenous sheep and its crossbreed with exotic Awassi in selected district of South Wollo, Amhara, Ethiopia

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Abstract. *Mohammed N, Alemayehu K, Getachew T. 2017. On-farm phenotypic characterization of indigenous sheep and its crossbreed with exotic Awassi in selected district of South Wollo, Amhara, Ethiopia. Trop Drylands 1: 1-12.* Selection of genetic resources is important to select desirable traits in breeding programs to improve the quality and production of livestock including sheep. This study aimed to investigate the phenotypic diversity of indigenous sheep breeds in selected districts of south Wollo highlands and to assess the effect of crossbreeding of Wollo sheep with exotic Awassi sheep on growth and morphological characters under stallholder management of eastern Amhara, Ethiopia. Three districts were selected purposely based on their sheep flock size, sheep production potential and their contribution to the farmers. Bodyweight and linear body measurements were taken from 525 mature indigenous sheep and Local x Awassi crossbred. Body weight and most of the linear body measurements of indigenous sheep population were significantly affected by the district. Crossbred had significantly higher body weight and body measurements than indigenous sheep in Legambo district. Sex had a significant effect on body weight, wither height, and rump height for Awassi crossbred. Body weight and all linear body measurements were significantly influenced by age group. Sex and age group interaction significantly affected body weight and most linear measurements of indigenous sheep, and significantly affected wither height and rump height of local x Awassi crossbred. There was a positive and strong correlation between body weight and chest girth of indigenous female sheep as well as between rump height, body length, chest girth, wither height, and body weight of indigenous male sheep. Positive and strong correlation between body weight and chest girth was observed for Awassi crossbred females. Chest girth and body length for indigenous and local x Awassi crossbred sheep could be used for the prediction of body weight. The body weight of local x Awassi crossbred (28.75±0.49 kg) was higher than the indigenous sheep (22.19±0.41 kg) in Legambo district. In conclusion, indigenous sheep have potential for improving livelihood and economy of smallholder farmers using crossbred in the study areas.

Keywords: Awassi, characterization, crossbred, indigenous sheep

INTRODUCTION

Genetic diversity provides raw materials for breed improvement in livestock rearing to adapt to changing environments and demands. Information on the origin and history of animal genetic resources (AnGR) is essential to design strategies for sustainable livestock management (Ajmone-Marsan et al. 2010; Felius et al. 2015). Therefore, the aspect of genetic resources is important particularly for the countries with large production of livestock, including sheep. The top five countries by sheep population from the world are China, Australia, India, Iran and Nigeria by 202 million, 72.6 million, 63 million, 50.2 million and 40.6 million sheep population, respectively (FAOSTAT 2014). According to FAOSTAT (2014) Africa has 340.5 million sheep population and the 2nd largest continent next to Asia by sheep population.

Ethiopia is home to at least 14 traditional sheep populations and nine breeds (Gizaw et al. 2007). From 28.89 million sheep population in the country, about 72.84% are females, and about 27.16% are males. From the total number of sheep, 99.78% are indigenous breeds and the rest are improved sheep (CSA 2016). At the national level, small ruminant accounts for about 90% of the live animal/meat and 92% of skin and hide export trade value (Asmare 2010). Sheep rearing in Ethiopia serves as a means of risk mitigation during crop failures, property security, monetary saving and investment, in addition to many other socio-economic and cultural functions. Sheep have special features like efficient utilization of marginal and small plots of land, short generation length, high reproductive rate, low risk of investment and more production per unit of investment as compared with cattle.

Urbanization and growing human population in Ethiopia resulted in increased domestic demand for sheep meat, which also offers a significant incentive for market-oriented production. It is very vital to improve sheep productivity in order to satisfy the large population of the country estimated at 81 million with 2.7% annual growth rate (AMFI 2010). The transfer of successful animal breeding schemes from developed countries also proved to be difficult or impossible in many instances because such schemes are high-tech operations involving sophisticated methods of measuring and evaluating animals, biotechnologies, very high level of organization and high level of input of capital and labor (Kosgey et al. 2006; Kosgey and Okeyo 2007).

The Ethiopian Sheep and Goat Productivity Improvement Program (ESGPIP) took the responsibility for importation of improved genotypes, multiplication of pure-bred, crossing with indigenous sheep and distribution of both crossbred and pure exotic animals to sheep producers especially for Dorper sheep and Boer goat. The sheep crossbreeding project started with various exotic wool breeds. Government bodies were responsible for importing; multiplication and dissemination of Hampshire, Corriedale, Dorper, Merino and Awassi. The two ranches of Debre Birhan and Amedguya were the main actors, particularly for Awassi sheep multiplication and dissemination. Initially, 50 Corriedale, 6 Hampshire and 6 Romney exotic rams were introduced. However, the exotic breeds were later abandoned because they were not accepted by the farmers as the breeds did not meet farmers' phenotypic preferences for horns and tails. In 1980, the Awassi breed, which has a similar phenotypic appearance to the local sheep, was introduced from Israel and has been well accepted by producers (Gizaw and Getachew 2009). In 1980, Awassi sheep were introduced from Israel and kept at Debre Birhan Sheep Breeding and Multiplication Center (DBSBMC) and Amed Guya Sheep Breeding and Multiplication Center (AGSBMC). There were also continuous importations of pure-bred Awassi sheep totaling 45 (ram and ewe lambs). The two government farms have been engaged in multiplication and distribution of crossbred rams to farmers at a subsidized price. In 2011, about 170 pure Awassi sheep were imported from Israel to start again crossbreeding in the farms (Getachew et al. 2016).

The usefulness of characterization of the phenotypic and the genotype of the indigenous sheep breeds (types) is not in doubt to overcome the abovementioned problem. Classification of the breed is the basis for different sheep breed improvement strategies and sheep productivity schemes. Breed diversity is high in peripheral and remote areas of the country (Gizaw et al. 2008) due to less human intervention for crossing. Similarly, sheep are adapted to various ecological niches and the differing needs and preferences of their breeders who belong to different ethnic communities. High phenotypic diversity was observed for morphological characters on sheep found in the country (Gizaw et al. 2007) and the significance within and between breeds.

Animal live body weight is an important feature, but can hardly still be measured in rural areas due to lack of reasonable accurate scales. Therefore, farmers have to rely on questionable estimates of the body weights of their sheep, leading to inaccuracies in decision making. The most important method of weighing animals without scale is to regress body weight to body characteristics, which can be measured readily. Body measurements have been used to predict body weight by several authors in many breeds of sheep (Sowande and Sobola 2007; Cankaya 2008; Abera et al. 2016).

Ethiopia has a varied sheep population of about 14 sheep types in four major groups, i.e. sub-alpine short fat-tailed, highland long fat-tailed, lowland fat-rumped/tailed and lowland thin-tailed (Gizaw et al. 2008). The sub-alpine short fat-tailed group which consists of Menz, Tikur, and Wollo and Simien sheep types is predominantly found in the central and northern highlands at an altitude of above 2500 m. In this area, sheep are mainly reared for income generation from the sale of lambs at market age although they are also important as a source of food, manure and socio-cultural benefits (Gizaw et al. 2008; Getachew et al. 2010).

On regional scale of Ethiopia, Amhara National Regional State has 9.80 million sheep (CSA 2016) with South Wollo zone alone having 1.86 million sheep, making it the zone with the largest sheep population in Amhara region (CSA 2016). This study aimed to investigate the phenotypic diversity of indigenous sheep breeds in selected districts of South Wollo highlands and to assess the effect of crossbreeding of Wollo sheep with Awassi on growth and morphological characters under stakeholder management.

MATERIALS AND METHODS

Study area

The study was conducted in South Wollo zone which is one of 10 zones and one special district in the Amhara Region of Ethiopia. South Wollo is bordered on the south by North Shoa and Oromia Region, on the west by East Gojjam, on the northwest by South Gonder, on the north by North Wollo, on the northeast by Afar Region, and on the east by the Oromia zone and Argobba special district. Its highest point is Mount Amba Ferit. Dessie is the capital city of South Wollo. Based on the 2007 Census conducted by the Central Statistical Agency of Ethiopia (CSA 2008), this zone has a total population of 2,518,862, with an area of 17,067.45 square kilometers. South Wollo zone has 22 districts; and three districts from these were selected for this study, namely Wogide (Wegde), Borena (Debresina), and Legambo (Figure 1; Table 1).

Sampling techniques

Three districts in South Wollo were selected purposely based on their sheep flock size, sheep production potential and their contribution to the farmers. To improve body weight and coarse wool production of indigenous Ethiopian sheep, crossbreeding with the Awassi breed imported from Israel is a common practice at Debre Birhan and Amed Guya sheep improvement stations (Lemma et al. 2014). On-farm Awassi x local crossbreeding was started in 1997 by Debre Birhan Agriculture Research Center (DBARC) aiming to evaluate the performance of crossbred sheep under farmer management. The project was carried out in three villages; Serity, Negasi-Amba and Chiro. Chiro was located in South Wollo administrative zone of the Amhara regional state (Getachew et al. 2015). There were 29 participating farmers in Legambo district of Chiro village. Kebele, the lower administrative hierarchy in Ethiopia, was selected considering the density of sheep types. The actual survey included a single visit to a sampling site during which quantitative measurements were made on mature sheep. From each district, three kebele (total of nine kebele) were selected.

According to FAO (2012), physical measurements should be taken only from a representative set of adult animals (as judged by dentition): about 100-300 females and 10-30 males. Body weight and linear measurements were taken from a total of 525 mature sheep 405 from indigenous and 120 from local x Awassi crossbred (135 indigenous from Wogide district and 45 per kebele, 150 indigenous from Borena district 50 per kebele and 240 from Legambo district 120 local x Awassi crossbred from Chiro about 37.5-75% Awassi blood level and 120 indigenous sheep from other two kebele). Within each kebele, measurements were made on individual sheep from randomly selected flocks for indigenous sheep. However, for local x Awassi crossbred sheep, Chiro kebele in Legambo district was purposely selected. Then individual animals were randomly selected from purposely selected flocks until the target number of sample animals was reached. Linear body measurements were made using measuring tape while live body weight is taken using suspended spring balance having 50 kg capacity with 0.2 kg precision.

Figure 1. Map of the study areas in South Wollo Zone, Amhara Region, Ethiopia. District of: 1. Wogide (Wegde), 2. Borena (Debresina), 3. Legambo

Table 1. Description of the Districts of South Wollo, Amhara, Ethiopia

Study areas characteristics	Districts Wogide	Borena	Legambo
Distance from Addis Ababa (km)	594	592	501
Distance from Bahir Dar (km)	673	671	580
Distance from Dessie (km)	193	191	100
Altitude (m. asl)	500-2700	500-3200	1500-3700
Latitude and longitude	10°40′N 11°38′E	10°55′N 38°30′E	11°00′N 39°00′E
Rainfall (mm)	600-1100	1500-3660	700-1200
Temperature (°C)	23°c	16.5°c	13°c
Area (km^2)	1,110.69	1,027.61	1,017.35
Human population	151,257	158,209	165,026
Cattle	36,684	78,533	76,464
Sheep	15,442	68,642	146,954
Goat	48,059	35,417	11,815
Horse	63	1,552	6,662
Donkey	16,045	14,171	18,769
Mule	341	1.493	2,842
Poultry	47,389	73,903	74,494

Data collection procedures

Quantitative traits cover the size and dimensions of animals' bodies or body parts, which are more directly correlated to production traits than qualitative traits. For example, body weight and chest girth are directly related to body size and associated production traits. In general, these variables have a continuous expression. This is because of the numerous genes that determine or influence their expression (FAO 2012). Body Measurements: Body weight, chest girth (CG), body length (BL), wither height (WH), tail length (TL), tail circumference (TC), rump height, ear length, horn length, scrotum circumference (SC) and body condition score (BCS) were measured using tailors measuring tape while weight was measured using suspended spring balance having 50 kg capacity. Each animal was identified by its sex, dentition and sampling site. Dentition record was included, as this will be the only reliable means to estimate the approximate age of an animal. Ages of the animals were estimated from dentition class following the procedure described by (ESGPIP 2009).

Adult sheep was classified into five age groups; no pair of permanent incisor (0 PPI), 1 pair of permanent incisor (1 PPI), 2 pairs of permanent incisor (2 PP), 3 pairs of permanent incisor (3 PPI) and 4 pairs of permanent incisor (4 PPI) to represent age of fewer than 12 months, 12 to 18 months, 18 to 24 months, 24 to 36 months and more than three years, respectively based on the (ESGPIP 2009). The data collected from each study area were checked for any error and corrected during the study period, coded and entered into the computer for further analysis.

Data management and statistical analysis

Data collected through body measurement were entered into Microsoft EXCEL 2007, Statistical Package for Social Sciences (SPSS 20 for Windows and SAS 9.1 for Windows). Preliminary data analyses like homogeneity test, normality test and screening of outliers were employed before conducting the main data analysis.

Morphological and body measurement data

Data collected on body weight and other body measurements were analyzed separately for each district. Quantitative characters (body weight and linear body measurements) were analyzed using the Generalized Linear Model (GLM) procedures of the Statistical Analysis System (SAS 9.1 2003). Scrotum circumference was analyzed for each breed by fitting age group as fixed factor. Horn was the specific character of male Wollo sheep only so that analysis of horn length was employed for male Wollo sheep only by fitting age group as fixed factor. When analysis of variance declares significance, least-square means were separated using adjusted Tukey-Kramer test.

Models to analyze adult body weight and other linear body measurements except for scrotum circumference and horn length were**:**

Model 1: $Yijk = \mu + Ai + Dj + Sk + (AxS)ik + eijk$

Where:

Y*ijkl* = the body weight or linear body measurements except scrotum circumference and horn length in the ith age group, jth district and k^{th} sex of

μ = overall mean

Ai = the effect of ith age classes (*i = 0, 1and* ≥2)

Dj = the effect of jth district (j: Wogide, Borena and Legambo district)

Sk = the effects of kth sex (k= male, female)

(AxS) ik = the effect of the interaction of *i* of age group with *k* of sex

Eijk = random residual error

Model 2: $Yijk = \mu + Ai + Bj + Sk + (AxS)ik + eijk$

Where:

Y*ijkl* = the body weight or linear body measurements except scrotum circumference and horn length in the ith age group, jth breed and k^{th} sex of

μ = overall mean

Ai = the effect of ith age classes (*i = 0, 1and* ≥2)

Bj = the effect of jth breed (j = Indigenous and local x Awassi crossbred)

Sk = the effects of kth sex (k= male, female)

(AxS) ik = the effect of the interaction of *i* of age group with *k* of sex

Eijk = random residual error

Model to analyze the scrotum circumference and horn length was:

$$Yijk = \mu + Ai + Dj + eij$$

Where:

Yijk = Scrotum circumference

μ = Overall mean

Ai = the effect of ith age classes (i = 0, 1 and ≥2)

Dj = the effect of j^{th} district (j = Wogide, Borena and Legambo district)

Eij= random residual error

Correlation and regression

Pearson's correlation coefficients for each breed were estimated between body weight and other body measurements within sex (SAS 9.1). Body weight and other body measurements: chest girth (CG), body length (BL), wither height (WH), tail length (TL), body condition score (BCS), tail circumference (TC), ear length (EL) rump height (RH), horn length (HL) and scrotum circumference (SC) were included for males whereas scrotum circumference (SC) was excluded from the analysis of female sheep.

Among the above measurements BL, WH, CG, BCS, TC, TL, RH, EL, HL and SC (for male only) were selected based on their strong correlation with body weight, then body weight was regressed on the body measurements (BL, WH, CG, TL, BCS, TC, RH, EL and HL) for females within each age group using stepwise regression procedure of (SAS 9.1 2003) to determine the best-fitted regression equation for the prediction of body weight from body measurements. Likewise, stepwise regression was also employed for females within each age group by excluding SC from the model. Best-fitted models were selected based on the coefficient of determination (R^2), R^2 change and simplicity of measurement under field conditions. The following models were used for the analysis of multiple linear regressions.

For male:

$$Y_j = \alpha + \beta_1 X_1 + \beta_2 X_2 + \beta_3 X_3 + \beta_4 X_4 + \beta_5 X_5 + \beta_6 X_6 + \beta_7 X_7 + \beta_8 X_8 + \beta_9 X_9 + X_{10}\beta_{10} + ej$$

Where:

Yj = the response variable (body weight)

α = the intercept

X_1, X_2, X_3, X_4, $X5$, X_6, X_7, X_8, X_9 and X_{10} are the explanatory variables body length, wither height, chest girth, tail length, tail circumference, body condition score, scrotal circumference, rump height, horn length and ear length, respectively.

β_1, β_2… β_9 is regression coefficient of the variables X_1, X_2... X_{10}

ej = the residual random error

For female:

$$Yj = \alpha + \beta 1X1 + \beta 2X2 + \beta 3X3 + \beta 4X4 + \beta 5X5 + \beta 6X6 + \beta 7X7 + \beta 8X8 + \beta 9X9 + ej$$

Where:

Yj = the dependent variable (body weight)

α = the intercept

$X1$, $X2$, $X3$, $X4$, $X5$, $X6$., $X7$, $X8$ and X_9 are the independent variables; body length, wither height, chest girth, body condition score, tail length, tail circumference, rump height, horn length and ear length, respectively.

β_1, β_2… β_8 is regression coefficient of the variable X_1, X_2... X_9

ej = the residual random error.

RESULTS AND DISCUSSION

The main source of variation in live body weight and linear body measurement were district, breed, sex, age group and the interaction of both sex and age group. Information on live body weight and linear measurements of the existing breed types has a vital role in the selection program. Information on body and testicle size of specific sheep breeds at constant age has paramount importance in the selection of genetically superior animals for production and reproduction purposes. Therefore, it is not doubted that live body weight and linear body measurement is playing a crucial role in genetic improvement and selection of specific breeds.

Effect of district, breed, sex, age group and interaction of age group and sex

District effect

The least-squares means and standard errors for the effect of district, breed, sex, age group and interaction of age group and sex on body weight and other body measurements are presented in Tables 2 and 3 for indigenous and Awassi crossbred. In this study body weight and most of the linear body measurements (body length, chest girth, wither height and rump height) were significantly affected by the district ($p<0.01$) whereas, scrotum circumference was not influenced by district ($P>0.05$). Alemayehu (2011) and Abera et al. (2016) reported that most of the linear body measurements had significant ($p<0.05$) effect on live body weight and linear body measurements across the districts studied. In contrary to the current study, Shibabaw (2012) indicated that district had no effect ($p>0.05$) except the body weight which was higher in Metta (29.4±0.2) than Deder (28.8±0.2) district in Haragehe highland sheep.

The results of this study revealed that body weight measurement was almost similar for Borena (25.71±0.39 kg) and Wogide sheep (24.90±0.38 kg) but higher than indigenous Legambo sheep (22.19±0.41 kg). It might be due to a serious problem of feed shortage, high population of sheep per household and management difference in Legambo district rather than Wogide and Borena districts. The body weight of indigenous sheep and local x Awassi crossbred in this study was lower than the Doyogena sheep (31.64±0.43 kg) (Taye et al. 2016). This might be due to the small size of Wollo sheep.

Breed effect

Local x Awassi crossbred had significantly higher values for body weight, body length, chest girth, wither height and rump height ($P < 0.01$) and scrotal circumference was no significant ($P>0.05$) between local x Awassi crossbred and indigenous sheep in Legambo. The body weight, body length, chest girth, wither height, rump height and scrotum circumference of local x Awassi crossbred were 28.75±0.49 kg, 60.82±0.49, 72.96±0.53, 65.94±0.43, 69.71±0.46 and 26.16±0.93 cm, respectively. Whereas the body weight, body length, chest girth, wither height, rump height and scrotum circumference of indigenous sheep were 22.19±0.41 kg, 51.03±0.41, 64.16±0.50, 57.60±0.40, 60.67±0.38 and 24.57±0.64 cm, respectively. The result of body weight of local x Awassi crossbred was lower than that of Bonga sheep but higher than Horro sheep (Edea et al. 2009). The result of body weight (28.75 kg) of local x Awassi crossbred was lower than pure Awassi (46.5 kg), and chest girth (72.96) in this study also lower than pure Awassi (86.90) (Macedonian 2011). It might be due to the small size of Wollo sheep.

Sex effect

For indigenous sheep, sex of the sheep had significant ($p<0.01$) effect on body weight, chest girth, wither height and rump height. However, body length of indigenous sheep was not affected ($p>0.05$) by sex of the sheep. This finding is in agreement with Getachew et al. (2009) who reported for Menz and Afar sheep of body weight, chest girth, wither height and rump height had significant ($P<0.01$) but body length was not ($P>0.05$) by sex of the sheep. Taye et al. (2010) reported that a significant effect of sex on body weight, heart girth, body length and height at wither in Washera sheep and Gebreyowhens and Tesfay (2016) who reported that a significant ($p<0.05$) effects on body weight and other linear body measurements for Atsbiwonberta district of Tigray Region. However, this study disagrees with Alemayehu (2011) who found the sex of the sheep had no significant ($p>0.05$) effect on the body weight and other linear measurements. Haylom et al. (2014) also reported that sex has no significant ($p>0.05$) effect on body weight, heart girth, body length and height at wither in highland sheep found in Atsbiwonberta. The body weight of indigenous rams (26.09±0.47 kg) and ewes (22.46±0.31 kg) in the current study was lower than those reported for Gozamen, Sinan and Hulet eju districts in East Gojam rams (30.8 kg) and ewes (28.3 kg) reported by Abera et al. (2016).

However, the body weight of indigenous rams (26.09±0.47) and ewes (22.46±0.31 kg) in the current study was higher than those reported for Menz rams and ewes, with 22 kg and 19.3 kg, respectively (Getachew et al. 2009) and Mohammed et al. (2015) who reported 25.86±0.20 kg for Gubalafto ram but almost similar for Gubalafto ewe (22.55±0.15 kg).

Male sheep were consistently higher in body weight, wither height and rump height ($p<0.01$) than females while body length and chest girth were higher in number but not statistically significant. Differences in live weight and most of the body measurements between sexes observed in indigenous showed that these parameters are sex-dependent. The body weight and other linear measurements of local x Awassi crossbred were higher than the indigenous sheep that are found in the study areas. This finding is higher than reported by Gebreyowhens and Tesfay (2016) for highland sheep population of Tigray region.

Age effect

In the highland sheep found in Atsbiwonberta, age has an effect on the live body and linear body measurements (Gebreyowhens and Tesfay 2016). In this study body weight and all the body measurements of indigenous sheep were significantly ($p<0.01$) affected by age group in the study areas. In these study areas, indigenous breed of body weight and all linear body measurements were increased as the age increased from the youngest (0 PPI) to the oldest (≥ 2 PPI).

The report of the current finding is in line to the previous reports of Edea et al. (2009) for Bonga and Horro sheep and Gebreyowhens and Tesfay (2016) who found that as the age of the individual animal advanced, the average values of live body weight, heart girth, body length, and height at wither are significantly ($P<0.05$) increased in the highland sheep population in Tigray Region. The current study observed that growth rate from 0PPI to 1PPI was higher compared to that from 1PPI to ≥2 PPI in live body weight of the indigenous sheep population.

Age by sex interaction

As the age of the male and female sheep advanced, their body size also increased, resulting in male are heavier than female sheep. It is obvious that both sex and age have a synergetic effect on body size of the animal (Gebreyowhens and Tesfay 2016). The interaction of sex and age group was significant ($p<0.01$) for body weight, chest girth, wither height and rump height but not significant ($p>0.05$) for body length for indigenous sheep. These parameters might not be affected by the sex-age interaction effect.

The current finding in the interaction effect between sex and age is in line to Getachew et al. (2009) for Menz and Afar sheep, Mohammed et al. (2015) for Habru and Gubalafto districts and Gebreyowhens and Tesfay (2016) for Atsbiwonberta district which were significantly ($p<0.05$) affected body weight, heart girth, wither height and rump height while body length was not affected significantly ($P>0.05$). However, previous findings by Abera et al. (2016) reported that age by sex interaction had a significant effect ($p<0.05$) for only body weight. In all age groups of indigenous males sheep were heavier ($p<0.01$) than females.

Body weight obtained at the 0PPI age group of indigenous male and female sheep in this study was lower than that in Mohammed et al. (2015) but at the age of 1PPI and 2PPI of indigenous males was higher, age of 1PPI indigenous females is almost similar, 2PPI higher than that in Mohammed et al. (2015) who reported for Habru and Gubalafto districts. Body weight of in this study of indigenous male and female sheep in all age group is higher than that in Getachew et al. (2009) who reported for male and female Menz sheep.

The overall body weight of indigenous sheep (24.27±0.28 kg) in three districts is similar to Mohammed et al. (2015) (25.51±2.15 kg) for Habru and Gubalafto districts. The overall scrotum circumference of indigenous sheep is in agreement with Mohammed et al. (2015) for Habru and Gubalafto districts. A significant effect of sex and age of sheep on body weight of sheep is reported by many scholars for different breeds of sheep (Getachew et al. 2009; Edea et al. 2009; Bimerew et al. 2011).

Correlation between body weight and linear body measurements

Determining animal live body weight, linear body measurements and their interrelationship and correlation are very important for determining genetic potential, breed standards and improved breeding programs for higher meat production (Younas et al. 2013). Correlation coefficients of live body weight with other quantitative variables for male and female indigenous sheep in three districts and local x Awassi crossbred are presented in Table 4. All linear body measurements of male indigenous sheep showed very high significant (p<0.01) associations with body weight positively. Except for horn length (P>0.05) all linear body measurements of female indigenous sheep showed very high significant (p<0.01) associations with body weight positively. Among measured linear quantitative variables of indigenous male sheep body length (r = 0.85), chest girth (r = 0.91), wither height (r = 0.88) and rump height (r = 0.87) were strongly correlated with the body weight. Among measured linear quantitative variables of indigenous female sheep body length (r = 0.76), chest girth (r = 0.85) was strongly correlated with body weight.

Among measured linear quantitative variables of local x Awassi crossbred, male sheep wither height (r = 0.71) was strongly correlated with the body weight whereas chest girth (r = 0.88) was correlated with body weight for female sheep. This highest correlation of wither height and chest girth with body weight than other body measurements is in line with other results (Gizaw et al. 2008; Getachew et al. 2009; Alemayehu 2011; Abera et al. 2016). The strong correlation of different measurements with body weight would imply these measurements can be used as indirect selection criteria to improve live weight (Gizaw et al. 2008; Getachew et al. 2009; Abera et al. 2014; Mohammed et al. 2015; Abera et al. 2016; Bireda et al. 2016) or could be used to predict body weight. The high correlation coefficients between body weight and body measurements for males and female suggest that either of these variables or their combination could provide a good estimate for predicting the live weight of sheep from body measurements.

Table 2. Least squares mean ± standard errors of body weight (kg) and other linear body measurements (cm) for the effects of districts, sex, age group and sex by age group for indigenous sheep in the study areas

Effects &	BW		BL		CG		WH		RH		SC	
levels	N	LSM±SE	N	LSM±SE	N	LSM±SE	N	LSM±SE	N	LSM±SE	N	LSM±SE
Overall	405	24.27±0.28	405	54.04±0.28	405	68.34±0.34	405	62.16±0.28	405	66.52±0.26	83	24.38±0.35
R^2	405	0.51	405	0.53	405	0.52	405	0.55	405	0.65	83	0.55
CV%	405	16.26	405	7.11	405	6.43	405	6.19	405	5.50	83	12.38
Districts		**		**		**		**		**		NS
Wogide	135	24.90±0.38a	135	55.69±0.38a	135	70.18±0.46a	135	63.42±0.38b	135	69.45±0.36a	30	24.74±0.52
Borena	150	25.71±0.39a	150	55.40±0.39a	150	70.67±0.47a	150	65.45±0.38a	150	69.44±0.36a	30	23.83±0.54
Legambo	120	22.19±0.41b	120	51.03±0.41b	120	64.16±0.50b	120	57.60±0.40c	120	60.67±0.38b	23	24.57±0.64
Sex		**		NS		**		**		**	NA	NA
Male	83	26.09±0.47a	83	54.46±0.47	83	69.50±0.57a	83	63.66±0.46a	83	68.19±0.44a	83	24.38±0.35
Female	322	22.46±0.31b	322	53.62±0.30	322	67.17±0.37b	322	60.64±0.30b	322	64.85±0.29b	-	-
Age group		**		**		**		**		**		**
0PPI	137	19..65±0.35c	137	49.65±0.35c	137	63.68±0.43c	137	58.77±0.35c	137	63.05±0.33c	46	20.50±0.42c
1PPI	46	25.00±0.58b	46	55.10±0.57b	46	69.00±0.70b	46	62.77±0.57b	46	67.23±0.54b	21	24.54±0.63b
≥2PPI	222	28.17±0.51a	222	57.36±0.50a	222	72.33±0.61a	222	64.93±0.50a	222	69.29±0.47a	16	28.05±0.72a
Sex by age		**		NS		**		**		**	NA	NA
Male 0PPI	46	20.24±0.57cd	46	49.73±0.57	46	63.31±0.69c	46	59.16±0.56de	46	63.78±0.54de	-	-
Male 1PPI	21	27.72±0.85ab	21	55.35±0.84	21	71.26±1.03a	21	64.41±0.84ab	21	68.89±0.79ab	-	-
Male ≥2PPI	16	30.31±0.98a	16	58.31±0.97	16	73.94±1.18a	16	67.41±0.96a	16	71.91±0.91a	-	-
Female0PPI	91	19.04±0.41d	91	49.58±0.41	91	64.04±0.49bc	91	58.38±0.40e	91	62.32±0.38e	-	-
Female1PPI	25	22.28±0.78c	25	54.86±0.77	25	66.75±0.94b	25	61.13±0.77cd	25	65.57±0.73cd	-	-
Female≥2PPI	206	26.04±0.27b	206	56.41±0.27	206	70.72±0.33a	206	62.42±0.27bc	206	66.67±0.26bc	-	-

Note: Means with different superscripts within the same column and class are statistically different. Ns: Non-significant; *significant at 0.05; **significant at 0.01, NA: none applicable. N: number of sheep, BW: body weight, BL: body length, CG: chest girth, WH: wither height, rump height and scrotum circumference. 0 PPI: 0 pair of permanent incisors; 1PPI: 1 pair of permanent incisor and ≥ 2 PPI: 2 or more pairs of permanent incisors. LSM: least square mean and SE: standard error.

Table 3. Least squares mean ± standard errors of body weight (kg) and other linear body measurements (cm) for the effects of breed, sex, age group and sex by age group for indigenous and local x Awassi crossbreed sheep for Legambo district

Effects &		**BW**		**BL**		**CG**		**WH**		**RH**		**SC**
levels	**N**	**SM±SE**	**N**	**LSM±SE**	**N**	**LSM±SE**	**N**	**LSM±SE**	**N**	**LSM±SE**	**N**	**LSM±SE**
Overall	240	25.55±0.41	240	56.21±0.41	240	68.57±0.45	240	62.23±0.36	240	65.60±0.38	46	25.95±0.72
R2	240	0.65	240	0.70	240	0.69	240	0.66	240	0.67	46	0.52
CV%	240	15.79	240	7.24	240	6.36	240	5.76	240	5.85	46	16.40
Breed		**		**		**		**		**		NS
Indigenous	120	22.19±0.41b	120	51.03±0.41b	120	64.16±0.50b	120	57.60±0.40b	120	60.67±0.38b	23	24.57±0.64
Awassi crossbred	120	28.75±0.49a	120	60.82±0.49a	120	72.96±0.53a	120	65.94±0.43a	120	69.71±0.46a	23	26.16±0.93
Sex		**		NS		NS		**		**	NA	NA
Male	46	26.99±0.74a	46	56.95±0.75	46	69.29±0.91	46	64.41±0.65a	46	68.01±0.70a	46	25.95±0.72
Female	194	24.11±0.34b	194	55.47±0.35	194	67.85±0.37	194	60.06±0.30b	194	63.19±0.32b	-	-
Age group		**		**		**		**		**		**
0PPI	78	20.42±0.48c	78	51.39±0.48b	78	63.26±0.52c	78	58.01±0.42c	78	61.37±0.45c	28	20.85±0.74b
1PPI	39	26.39±0.68b	39	57.41±0.69a	39	69.64±0.74b	39	62.44±0.60b	39	65.43±0.64b	13	26.65±1.10a
≥2PPI	123	29.84±0.91a	123	59.83±0.92a	123	72.80±0.99a	123	66.24±0.80a	123	70.00±0.85a	5	30.34±1.74a
Sex by age		NS		*		NS		**		**	NA	NA
Male 0PPI	28	21.46±0.75	28	51.16±0.76b	28	63.63±0.82	28	58.49±0.66cd	28	62.41±0.71cd	-	-
Male 1PPI	13	27.08±1.11	13	57.38±1.12a	13	70.16±1.21	13	63.19±0.97b	13	66.19±1.04b	-	-
Male ≥2PPI	5	32.44±1.78	5	62.32±1.80a	5	74.08±1.94	5	71.54±1.56a	5	75.42±1.67a	-	-
Female 0PPI	50	19.39±0.58	50	51.63±0.58b	50	62.89±0.63	50	57.53±0.50d	50	60.33±0.54d	-	-
Female 1PPI	26	25.70±0.78	26	57.45±0.79a	26	69.13±0.85	26	61.70±0.89bc	26	64.68±0.73bc	-	-
Female≥2PPI	118	27.24±0.37	118	57.33±0.37a	118	71.52±0.40	118	60.94±0.32cd	118	64.57±0.35bc	-	-

Note: Means with different superscripts within the same column and class are statistically different. Ns: Non-significant; *significant at 0.05; **significant at 0.01, NA: none applicable. N: number of sheep, BW: body weight, BL: body length, CG: chest girth, WH: wither height, rump height and scrotum circumference. 0 PPI: 0 pair of permanent incisors; 1PPI: 1 pair of permanent incisor and ≥ 2 PPI: 2 or more pairs of permanent incisors. LSM: least square mean and SE: standard error

Prediction of body weight from different linear body measurements

Multiple regression analysis in the study areas is presented in Table 5. Regression analysis is commonly used in animal research to describe quantitative relationships between a response variable and one or more explanatory variables such as body weight and linear body measurements especially when there is no access to weighing equipment (Cankaya 2008). The accuracy of functions used to predict live weight or growth characteristics from live animal measurements is of vast financial contribution to livestock production enterprises. Multiple regression equations were developed for predicting body weight from other linear body measurements. Stepwise regression was carried out for each breed within each sex and age group for each breed by entering all traits at a time for male and by excluding horn length and scrotum circumference for females for selection of independent variables.

In all sex categories of indigenous sheep (for three districts of indigenous) and local x Awassi crossbred chest girth was consistently selected and entered into the model in step one procedure of stepwise regression due to its larger contribution to the model than other variables. At second step of stepwise regression chest girth and body length were selected to be in the model, at third step 3 independent variables, etc.

The coefficient of determination (R^2) represents the proportion of the total variability explained by the model. In most cases chest girth was found to be the most important in accounting large proportion of the changes in body weight, this measurement was reported for Afar and Menz (Getachew et al. 2009), for Bonga and Horro (Edea et al. 2009) and for Gozamen, Sinan and Hulet eju districts (Abera et al. 2016). Chest girth was more reliable in predicting body weight than other linear body measurements at farmer's level when there are no facilities to take the whole measurements.

Scientifically, chest girth, body length, tail circumference, body condition score and wither height were found to have a significant association with body weight for indigenous males which explained the total variability of 92% to the dependent variable body weight. Similarly, chest girth, body length, body condition score, wither height, rump height and tail length were found to have a significant association with body weight for indigenous females which explained the total variability of 80% to the dependent variable body weight.

Wither height and body length were found to have a significant association with body weight for local x Awassi crossbred males which explained total variability of 61% to the dependent variable body weight. Whereas, chest girth and body length had a significant association with body weight for local x Awassi crossbred females which explained the total variability of 80% to the dependent variable body weight.

Table 4. Phenotypic correlation between body weight and linear body measurements for indigenous sheep and local x Awassi crossbred above the diagonal are male and below diagonal are female sheep in selected districts

Breed	Traits	BCS	BW	BL	CG	WH	TL	TC	EL	RH	HL	SC
Indigenous	BCS		0.60**	0.45**	0.55**	0.47**	0.35**	0.30**	0.32**	0.47**	0.31**	0.42**
	BW	0.41**		0.85**	0.91**	0.88**	0.54**	0.40**	0.29**	0.87**	0.67**	0.64**
	BL	0.19**	0.76**		0.75**	0.83**	0.55**	0.32**	0.37**	0.78**	0.52**	0.58**
	CG	0.32**	0.85**	0.73**		0.85**	0.50**	0.30**	0.28**	0.88**	0.64**	0.60**
	WH	0.21**	0.70**	0.67**	0.74**		0.58**	0.28*	0.39**	0.91**	0.61**	0.53**
	TL	0.16**	0.28**	0.29**	0.20**	0.25**		0.25*	0.45**	0.56**	0.38**	0.51**
	TC	0.22**	0.27**	0.17**	0.27**	0.24**	0.10^{NS}		0.03^{NS}	0.29**	0.47**	0.49**
	EL	-0.06^{NS}	0.26**	0.34**	0.34**	0.39**	0.16**	0.13*		0.28**	0.23*	0.27*
	RH	0.11*	0.64**	0.70**	0.75**	0.84**	0.25**	0.21**	0.46**		0.62**	0.53**
	HL	-0.06^{NS}	0.01^{NS}	0.02^{NS}	-0.19^{NS}	0.05^{NS}	0.21^{NS}	0.11^{NS}	0.30^{NS}	0.22^{NS}		0.52**
Local x Awassi crossbred	BCS		0.54**	0.40^{NS}	0.22^{NS}	0.40^{NS}	0.38^{NS}	0.26^{NS}	0.02^{NS}	0.38^{NS}	0.12^{NS}	-0.12^{NS}
	BW	0.29**		0.60**	0.63**	0.71**	0.59**	0.49*	-0.01^{NS}	0.61**	0.51*	0.44*
	BL	0.09^{NS}	0.68**		0.66**	0.48**	0.53**	0.43*	-0.01^{NS}	0.45*	0.67**	0.25^{NS}
	CG	0.27**	0.88**	0.65**		0.65**	0.39	0.31^{NS}	0.25^{NS}	0.58**	0.52*	0.55**
	WH	0.14^{NS}	0.53**	0.47**	0.62**		0.58**	0.52*	0.13^{NS}	0.88**	0.46*	0.45*
	TL	-0.29**	-0.01^{NS}	0.07^{NS}	0.09^{NS}	0.14^{NS}		0.70**	-0.38^{NS}	0.55**	0.47*	0.46*
	TC	0.13^{NS}	0.45**	0.26**	0.43**	0.30**	0.15^{NS}		-0.27^{NS}	0.46*	0.41^{NS}	0.43*
	EL	0.05^{NS}	0.44**	0.31**	0.49**	0.30**	0.15^{NS}	0.43**		-0.03^{NS}	-0.28^{NS}	0.03^{NS}
	RH	0.07^{NS}	0.51**	0.45**	0.52**	0.68**	0.12^{NS}	0.38**	0.22*		0.46*	0.36^{NS}
	HL	-0.42^{NS}	0.31^{NS}	0.38^{NS}	0.25^{NS}	0.56*	-0.23^{NS}	-0.39^{NS}	0.15^{NS}	0.26^{NS}		0.51*

Ns: Non significant; *significant at 0.05; **significant at 0.01. BCS: body condition score, BW: body weight, BL: body length, CG: chest girth, WH: wither height, TL= tail length, TC: tail circumference, EL: ear length, RH: rump height, HL: horn length and SC: scrotum circumference

The R^2 was the criteria are used to select the model. The R^2 always increases as a new variable was added to the model thus we have to consider when new variable is added to the model, which variable will notably increase the R^2 change when added to the model. Generally, in most cases where regression analysis is applied, there can be several potential independent variables that could be included in the model. It is often not easy which variables are really needed in the model. The precision of the model becomes less when we use few variables in the model and inclusion of many variables leads to multicollinearity (Getachew et al. 2009).

The addition of more variable under field condition increase error incurred by the individual taking measurements and some variables are more affected by the animal posture so it is difficult to measure such variables accurately. It was recognized that chest girth is among the variables least affected by the animal posture and easy to measure than other measurements like withering height and body length. Thus under field conditions, live weight estimation using chest girth alone would be preferable to combinations with other measurements because of difficulty of the proper animal restraint during measurement.

Regression models for predicting body weight of males and females of indigenous sheep and Awassi crossbred in south Wollo zone selected districts could be based on regression equation;

For indigenous males:

$$Y=-36.21+0.47CG+0.26BL+0.09TC+0.10BCS+0.19WH$$

where Y= response variable (body weight) and chest girth, body length, tail circumference, body condition score and wither height are explanatory (independent) variables.

For indigenous female:

$$Y=-29.57+0.58CG+0.30BL+0.14BCS+0.19WH+0.19RH+0.05TL$$

where Y = response variable (body weight) and chest girth, body length, body condition score, wither height, rump height and tail length are explanatory (independent) variables.

For Awassi x local crossbred male:

$$Y=-19.04+0.54WH+0.37BL$$

where Y= response variable (body weight) and wither height and body length are explanatory (independent) variables.

For Awassi x local crossbred female

$$Y=-44.32+0.77CG+0.18BL$$

where Y = response variable (body weight) and chest girth and body length are explanatory (independent) variables.

Table 5. Multiple regression analysis of live weight on different linear body measurements for indigenous and local x Awassi crossbred ram and ewe in the study areas

Age	Model	Intercept	β_1	β_2	β_3	β_4	β_5	β_6	R^2	R^2 change
Indigenous										
Male	CG	-26.38±2.54	0.91±0.04						0.83	0.00
	CG+BL	-32.78±2.26	0.64±0.05	0.37±0.06					0.89	0.06
	CG+BL+TC	-34.55±2.27	0.62±0.04	0.35±0.06	0.10±0.06				0.90	0.01
	CG+BL+TC+BCS	-34.49±2.20	0.58±0.05	0.34±0.06	0.09±0.06	0.10±0.50			0.91	0.01
	CG+BL+TC+BCS+WH	-36.21±2.24	0.47±0.06	0.26±0.07	0.09±0.06	0.10±0.49	0.19±0.08		0.92	0.01
Female	CG	-24.30±1.69	0.85±0.02						0.72	0.00
	CG+BL	-27.29±1.62	0.64±0.03	0.29±0.04					0.76	0.04
	CG+BL+BCS	-28.08±1.54	0.57±0.03	0.31±0.04	0.17±0.22				0.79	0.03
	CG+BL+BCS+WH	-29.33±1.63	0.52±0.04	0.28±0.04	0.17±0.22	0.09±0.04			0.79	0.01
	CG+BL+BCS+WH+RH	-28.68±1.62	0.57±0.04	0.31±0.04	0.15±0.22	0.19±0.05	-0.18±0.05		0.80	0.01
	CG+BL+BCS+WH+RH+TL	-29.57±1.67	0.58±0.04	0.30±0.04	0.14±0.22	0.19±0.05	-0.19±0.05	0.05±0.05	0.80	0.01
Local x Awassi crossbred										
Male	WH	-11.83±8.69	0.71±0.13						0.50	0.00
	WH+BL	-19.04±8.52	0.54±0.14	0.37±0.12					0.61	0.10
Female	CG	-38.77±3.69	0.88±0.05						0.78	0.00
	CG+BL	-44.32±4.00	0.77±0.06	0.18±0.08					0.80	0.02

Note: CG: chest girth, BL: body length, BCS: body condition score, TC: tail circumference, WH: wither height, RH: rump height and TL: tail length

To summarize, the study of on-farm phenotypic characterization of indigenous sheep population has been carried out in South Wollo zone mainly in three districts (Wogide, Borena, and Legambo). In addition, local x Awassi crossbred sheep were considered in Legambo district only. The results showed there was a variation in body weight and in all linear body measurements in these districts. This might be due to environmental and genotype differences. The result showed presence of good indigenous sheep genetic potential in the study areas. The breed has the potential to improve livelihood and to increase economy of smallholder farmers in the study areas as well as in the surrounding areas. Notable variation among indigenous population was observed. Indigenous populations in Wogide and Borena were found larger in size. Relatively large size sheep population in Wogide and Borena compared to Legambo might be attributed to better feed resources and natural selection under better environment. Community-based crossbreeding which has been implemented Debre Berhan Agricultural Research Center at Chiro village successfully improved size and the productivity of local sheep in the Legambo district. Good phenotypic correlation was observed among body weight and linear body measurements. This would help for body weight prediction as well as using measurements directly to select best animals for future breeding. In this study, local x Awassi crossbred had significantly higher values for body weight and other linear body measurements than indigenous sheep. In this study, local x Awassi crossbred with indigenous Wollo sheep in Legambo district especially Chiro village was good indicator to improve local sheep through crossbreeding. Thus, strengthening the existing crossbreeding program in Legambo site and scaling out to similar areas in a controlled way is suggested. However, current attempts and interest by the extension to expand crossbreeding in Wogide and Borena areas need to be considered carefully. Such type of approach might not bring attractive benefits rather will dilute such valuable indigenous genetic resources found in the areas. Thus, in-situ conservation and improvement of the indigenous sheep in Wogide and Borena would be crucial before being diluted to the adapted and relatively productive genotype.

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Soil improvement potential of weeds biomass applied as green manure in marginal land

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Abstract. *Hamdani AD, Sulistiyowati E, Khusnuryani A. 2017. Soil improvement potential of weeds biomass applied as green manure in marginal land. Trop Drylands 1: 12-16.* Weeds have been long considered to have negative effect on crops due to competition. Yet, the presence of weeds can be used to build up organic matter and chemical contents of degraded soil. This study aimed to assess the potential of three weed species, i.e. *Chromolaena odorata*, *Mikania micrantha*, and *Synedrella* nodiflora, to be applied as green manure to improve soil quality in marginal land, and to assess the best form of their practical application. Two forms of weed biomass application, fresh and dried chopped, were examined in a pot experiment containing soil from degraded land in Patuk, Gunung Kidul, Yogyakarta, Indonesia, and spinach (*Amaranthus tricolor* L.) as the test crop. The pots were laid out in a randomized complete block design with six replications. Laboratory analysis showed that manurial properties between the weeds were statistically similar, with the average of organic C, N, P and K at 36.9, 3.3, 0.1 and 1.6%, respectively. Of the chemical contents considered, the content of all chemicals measured in experimental soil had increased, particularly the available K_2O from 3.33 to the average of 46.33 ppm. The spinach growth and yield resulting from the treated soils were superior to that from untreated soils. However, there were no significant differences in the plant's response between the types of weeds. Rather, better growth and yield of spinach have resulted from soil treated with dried chopped weed biomass. Thus, weed biomass can be used to improve soil organic matter and therefore should be incorporated with soil fertility maintenance.

Keywords: *Chromolaena odorata*, green manure, *Mikania micrantha*, soil fertility, *Synedrella nodiflora*

INTRODUCTION

The adverse effect of intensive utilization of land for cultivation, particularly related to continued removal of nutrients from soil, in combination with highly use soil-degrading agrochemicals, remains the biggest challenge in developing sustainable agriculture system. Both practices have made it difficult to maintain or enhance soil fertility, which is determined by sufficient availability of nutrients, water, air and microflora in soil to support plant growth. Such problems are more prominent on the soils classified as marginal land which have low potential soil fertility. Therefore, inexpensive systems to continually rejuvenate degraded soils are needed to support sustainable agriculture.

Technically, soil fertility can be maintained or improved by adding organic or inorganic fertilizer, or a combination of both. Nonetheless, many argue that organic amendments are more beneficial than inorganic fertilizer to develop sustainable agriculture system in the long run. Even though the application of organic fertilizer does not always fulfill the nutrient needed since its effectivity depends on many factors, such as the decomposition and nutrients release rates, the application of organic inputs can still improve soil quality in term of physical and biological elements (Hayat et al. 2010; Egbe et al. 2012).

One of the sources of organic fertilizer is plant biomass. Most of the green manures applied for cultivation are generated from biomass of the leguminous plants since they can enrich the soil content of Nitrogen that is required in large supply by most crops (Liu et al. 2008). However, the availability of legume plant biomass is limited so alternative plant sources as base materials for green manure are needed. Another group of plants that can be considered as potential plant materials is weeds. Most weeds are inexpensive, do not require much care, have rapid growth rate and are highly adaptive to various environments. Since they are available in abundance, the large amount of biomass required in their application as green manure can be fulfilled.

Weeds are often considered an enemy of the cultivation process. However, some studies demonstrated that a farmed land that has been allowed to lie idle and occupied by weeds is more productive when cultivated again than it did before because the presence of weeds can build up organic matter and chemical contents of soil. For example, Olabode et al. (2007) reported that yields from okra (*Abelmoschus esculentus*) in pots treated with crushed weeds tithonia (*Tithonia diversifolia*) were 40% higher than those without any amendments. Similarly, Jama et al. (2000) demonstrated that green manure from tithonia improved soil fertility and increase the availability of soil N, P and K. Application of *Erigeron annuus* (L.) improve soil pH and nutrient concentrations in the acid soils of southern China (Liu et al. 2008). Likewise, Patel (2012) also mentioned the potential utilization of weed *Eichhornia crassipes* as materials for green manure. Therefore, when weeds are harvested it can be applied as green manure while on the other hand reducing their competition with the main crops with the expected results of better growth and yield of the cultivated crops.

The highly abundance of weeds should be considered as an advantage to help building up the impoverished soils, especially to solve the problem of soil fertility in marginal land. However, not every weed might be potential as green manure base material. The suitability of plant material for soil amendment depends on the occurrence of high N, P and K concentrations in plant tissues (Gachengo et al. 1999; Tomar et al. 2012). For example, most green manure is plant biomass generated from legume family because they contain high level of N in their tissues. In addition, the quality of green manure is also determined by the relative proportions of lignin, the high rate of decomposition and nutrient release (Olabode et al. 2007). Tithonia, for instance, was considered as a better source of plant materials/biomass for green manure than *Senna spectabilis* (senna) because the release rate of N and P from tithonia is higher than from senna (Gachengo et al. 1999).

In this experiment, we assessed the extent to which organic matters or biomass of three species recognized as weeds, i.e. *Chromolaena odorata*, *Mikania micrantha,* and *Synedrella nodiflora*, can be used to improve the fertility of experimental soil from marginal land in Patuk, Gunung Kidul, Yogyakarta, Indonesia. These weeds species were selected because of their availability in a large supply, had high biomass and were expected to decompose easily due to their low lignin contents. Also, two forms of applications, i.e. fresh and dried chopped weeds, were tested to determine the best mode of their practical application. The application was without fermentation based on the previous study results (Olabode et al. 2007) that soils treated with crushed tithonia showed higher N concentration over the soils treated with fermented tithonia. This could be another strong point since unfermented process would simplify the application of green manures.

MATERIALS AND METHODS

Soil sampling and analysis

The soil samples were collected in September 2015 from the 0-20 cm horizon of all plots in Patuk, Gunung Kidul, Yogyakarta. Physical and chemical characteristics of the soils were determined according to standard methods in BPTP Yogyakarta. Physical analysis included determination of soil texture (sand, dust, clay) using hydrometer method. As for chemical characteristics, the total N was determined by Kjeldahl method, available P_2O_5 was determined by Olsen method, available K_2O was determined by water-soluble extraction, potential P_2O_5 and K_2O were determined by extraction using 25% HCl, and organic carbon was determined by Walkley and Black method.

Plant sampling and analysis

Three species of weeds used in this study were *Chromolaena odorata*, *Mikania micrantha*, and *Synedrella nodiflora*. The weed samples were collected randomly from several farming areas in Sleman, Yogyakarta and then pooled. The plants were harvested at ground level, but only the leaves were used as green manures. Chemical analysis of the weed samples was done according to standard methods in BPTP Yogyakarta. The total N was determined by Kjeldahl method, P was determined by spectrometer, K was determined by AAS, and organic carbon was determined by Walkley and Black method.

Pot experiment

Two treatments, corresponding to two forms of weed preparations (i.e. fresh and dried chopped) for each type of weed, were compared to the control groups of pots without any soil amendments (negative control) and pots supplemented with inorganic fertilizer. Those were fitted into a randomized complete block experiment. There were 24 pots such that three pots represented each group with six replications. Each of the pots was filled with soil taken from the experimental area in Patuk, Gunung Kidul, Yogyakarta, and the roughly chopped leaves (fresh weight) with a ratio of 10:1. The pots were then incubated for two weeks before planting and during the time were watered every morning and afternoon. After two weeks, samples from soils treated with chopped weeds biomass were collected for analysis of available nutrient status in BPTP Yogyakarta.

For the fresh chopped treatment, the chopped leaves were directly mixed thoroughly with the soil, while for the dried chopped treatment; the chopped leaves were mixed with the soil after being dried in the sun for two days. Nothing was added to the soil assigned as the negative control pots while the positive control pots were supplemented with NPK fertilizer (60 kg ha^{-1}) according to the users' guide.

The spinach seeds were sowed in advance for two weeks, before planting in the pots. Each pot was watered as needed as well as the mulching. Data collected included plant height, number of leaves, root length, and fresh weight.

Data analysis

The plant and soil data were analyzed statistically using the analysis of variance (ANOVA) using IBM SPSS Statistics 21. The difference of treatment result was assessed using Duncan test for separating the mean of treatments at 5%.

RESULTS AND DISCUSSION

Physical and chemical characteristics of the soil used in this study were presented in Table 1. The analysis on soil physical properties showed that the soil was dominated by clay content, which reached up to 47%, followed by sand content of as much as 27% and 25.67% of dust, hence the experimental soil was categorized as clay soil. The N, P, K, and organic carbon of soil were generally very low, implying that the level of soil fertility was also very low.

Meanwhile, the tissue nutrient concentrations of *Chromolaena odorata*, *Mikania micrantha*, and *Synedrella nodiflora* used in this study were presented in Table 2. Overall, the N, P, K and organic C tissue contents were statistically similar. The highest organic C was found in *C. odorata*, respectively 8.26 and 7.42% better than *M. micrantha* and *S. nodiflora*. Likewise, *C. odorata* was also 6.58 and 6.92%, respectively better than *M. micrantha* and *S. nodiflora* in N tissue contents. Meanwhile, *M. micrantha* was better than *C. odorata* by 60% P and 77.78% K, also 82.61 and 27.27% better in K tissue contents than in *C. odorata* and *S. nodiflora*, consecutively. With regard to the same ratio of C/N in those weeds, the succulent level of *C. odorata*, *M. micrantha* and *S. nodiflora* were the same, indicating a similar rate of decomposition process. Therefore, *C. odorata*, *M. micrantha* and *S. nodiflora* could have similar manurial potential.

The occurrence of N, P, K and organic C concentrations in all selected weeds were considered high. When compared to the reported values of other sources of green manures, the nutrient contents of *C. odorata*, *M. micrantha* and *S. nodiflora* were comparable to tithonia (*Tithonia diversifolia*), the weed that considered as a high-quality organic source for green manure, which contained 31.76% organic C, 4.46% N, 0.61 P and 3.75% K (Hafifah et al. 2016). However, the nutrient status of plants varied with soil characteristics, geographical conditions and ecological status of the plants. The occurrence of very high nutrient concentration in plant materials can be obtained from fertile soil. For example, N concentration in *C. odorata* samples used in this study was higher than that found in the same species collected from the savanna of Nigeria (3.4 as opposed to 1.76, respectively) (Olabode et al. 2007). Therefore, the effective use of plant materials as green manure should consider the conditions of plant source location, coupled with the availability of plants and the level of nutrient deficiency of treated soil.

The chemical characteristics of soil after incubated with the chopped weeds for two weeks (Table 3) showed the increasing level of N, P, K and organic C, particularly on available K_2O that drastically increase from 3.33 ppm to an average of 46.33 ppm. Overall, the level of soil fertility had increased from very low to low with the addition of chopped weeds.

From the three plant species used in this research, *C. odorata* has the highest potential to increase soil chemical properties. Previous research regarding *C. odorata* showed a similar trend. For example, Nawaz and George (2004) reported that organic compost of *C. odorata* was able to increase crop biomass, and interestingly, this compost had similar effect to the use of cattle manure. When it was used as green manure, *C. odorata* was reported to increase NPK uptake and nutrient efficiency in paddy cultivation (Murthy et al. 2010).

Mikania micrantha showed allelopathic properties when it was used as green manure as it has been reported to have allelopathic effect on some weeds in crop fields (Weng 1964). Ullah et al. (2014) administered extract of *M. micrantha* to *Fymbristylis miliacea* weed and reported that it significantly lower Fymbristylis population. This gives less preferable effect as green manure compared to the other two plant species used in this experiment.

Murthy et al. (2010) asserted that the use of weeds as organic manure could increase the quality of soil chemical properties. The data shown in Table 3 confirmed that statement.

In terms of C and N properties, it seems that dried chopped *C. odorata* gave slightly better C and N improvement. This result is almost in close conformity with the findings of the usage of *C. odorata* for increasing the yield of rice (Paraye 2002). This manurial practice could help in enhancing biochemical activity of microorganisms living in sandy loam and sandy clay soils (Paraye 2002), exactly similar to the type of soil used in this research. In terms of amendment of available P_2O_5, and K_2O, as well as potential P_2O_5 and K_2O, all weeds used, could improve soil chemical properties. Sandy-clay soil is typically low in available phosphorous, hence in traditional farming practice, farmers tend to supply the need of P from inorganic sources. However, in alkaline soil, like clay, P reacts with Fe and Al and creates insoluble P. The use of green manure could reduce this risk, because it increases availability of P and reduces soil capacity to react with P, and thus P is more readily available for plants (Mweta et al. 2007).

Of the plant response considered (Figure 3, Table 4), plant growth and yields from soil supplemented with chopped weeds were superior to that from soil without the addition of fertilizers, for all the measured parameters, and were parallel with those treated with NPK fertilizer. And among the treated soil, better plant growth and yields were observed from soil added with dried chopped weeds than fresh weeds, but not significantly different according to the types of weeds used.

Table 1. The initial physical and chemical properties of soil used for the study

Property	Value
pH	7.00
Particle Density (g cc^{-1})	2.61
Organic Carbon (%)	0.65
Total Nitrogen (%)	0.08
C/N Ratio	8:1
Available P_2O_5-Olsen (ppm)	7.33
Potential P_2O_5-25% HCl (mg $100g^{-1}$)	55.33
Available K_2O-Olsen (ppm)	3.33
Potential K_2O-25% HCl (mg $100g^{-1}$)	5.33
Sand (%)	27.00
Silt (%)	25.67
Clay (%)	47.00

Table 2. Chemical properties of *Chromolaena odorata*, *Mikania micrantha*, and *Synedrella nodiflora.*

Weed species	Manurial properties (%)				
	Org. C	N	P	K	C/N
Mikania micrantha	35.81	3.19	0.16	2.1	11:1
Chromolaena odorata	38.77	3.4	0.09	1.15	11:1
Synedrella nodiflora	36.09	3.18	0.1	1.65	11:1

Table 3. The effect of weeds application on chemicals soil characteristics

Form of weed biomass	Organic C (%)	Total N (%)	Available P_2O_5 (ppm)	Available K_2O (ppm)	Potential P_2O_5 (mg 100 g^{-1})	Potential K_2O (mg 100 g^{-})
Negative control	0.65^{a}	0.07^{a}	7.33^{a}	3.33^{a}	55.33^{a}	5.33^{a}
Mikania micrantha						
Fresh chopped	0.89^{ab}	0.10^{b}	9.33^{a}	48.67^{b}	75.67^{a}	12.00^{bc}
Dried chopped	0.97^{ab}	0.11^{bc}	11.67^{a}	43.33^{b}	63.67^{a}	9.67^{b}
Chromolaena odorata						
Fresh chopped	1.06^{ab}	0.12^{c}	9.00^{a}	49.33^{b}	69.67^{a}	9.00^{b}
Dried chopped	1.27^{b}	0.13^{c}	12.00^{a}	41.00^{b}	61.33^{a}	9.67^{b}
Synedrella nodiflora						
Fresh chopped	1.00^{ab}	0.11^{bc}	8.67^{a}	47.67^{b}	60.67^{a}	10.00^{bc}
Dried chopped	1.09^{ab}	0.11^{bc}	16.00^{a}	48.00^{b}	58.00^{a}	13.00^{c}

Note: Means along the column with the same superscript are not significantly different by Duncan test ($\alpha = 0.05$)

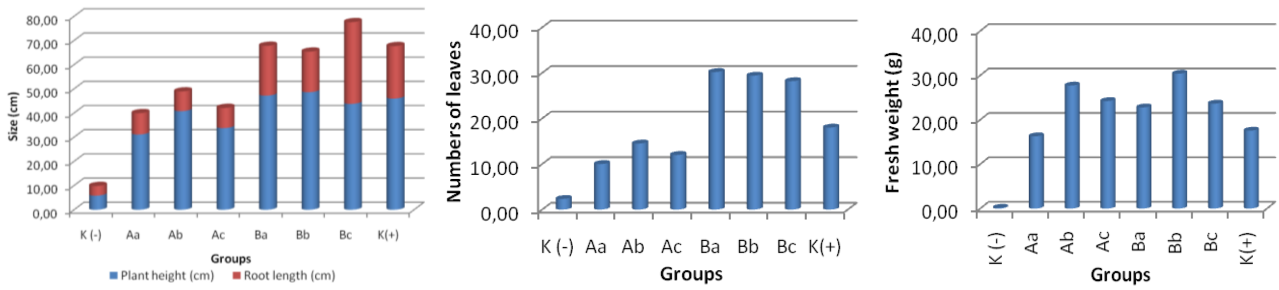
It was not surprising since the manurial properties of *C. odorata*, *M. micrantha* and *S. nodiflora* were similar. However, better results observed from soil treated with dried chopped weeds than those from fresh chopped were inconsistent with the finding of Olabode et al. (2007). This could be caused by different modes of application. Olabode et al. (2007) applied crushed fresh and ground dried to the soil while in this study we used the roughly chopped weeds for practical applications. When incorporated with the experimental soil, the dried chopped weeds were more easily broken into smaller pieces, hence increasing the surface area for decomposition and fastening the process, as opposed to the fresh ones. Therefore, the soil nutrient supplies required for plant growth were more readily available in the soil incorporated with dried chopped weeds.

We concluded from the present study results that the weeds *Chromolaena odorata*, *Mikania micrantha*, and *Synedrella nodiflora* have the potential to be used as green manures to improve soil fertility. According to this study, dried chopped weeds were more preferable to without drying process. This application should, therefore, be incorporated with weeds management for more efficient ecosystems services.

Table 4. The effect of weed biomass application on spinach growth and yields.

Form of weed biomass	Plant height (cm)	Root length (cm)	Numbers of leaves	Fresh weight (g)
Negative control	5.83^{a}	4.00^{a}	2.33^{a}	0.19^{a}
Mikania micrantha				
Fresh chopped	31.12^{b}	8.82^{a}	10.00^{ab}	16.27^{b}
Dried chopped	47.20^{d}	20.60^{b}	30.20^{c}	22.77^{b}
Chromolaena odorata				
Fresh chopped	40.78^{bcd}	8.15^{a}	14.50^{ab}	27.71^{b}
Dried chopped	48.60^{d}	16.80^{b}	29.40^{c}	30.58^{b}
Synedrella nodiflora				
Fresh chopped	33.80^{bc}	8.33^{a}	12.00^{ab}	24.21^{b}
Dried chopped	43.80^{bcd}	33.80^{c}	28.20^{c}	23.63^{b}
Positive control	46.00^{cd}	21.67^{b}	18.00^{bc}	17.54^{b}

Note: Means along the column with the same superscript are not significantly different by Duncan test ($\alpha = 0.05$).



A B C

Figure 3. Plant growth and yields response of spinach planted in soil treated with chopped weeds as opposed to soil without any amendments (negative control) and soil supplemented with NPK fertilizer (positive control). The measured parameters included plant height and root length (A), the number of leaves (B), and fresh weight (C) at the harvest time of 40 days after planting. A= fresh chopped weeds treatment group; B = dried chopped weeds treatment group; a = *Mikania micrantha*; b = *Chromolaena odorata*; c = *Synedrella nodiflora*

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Grain yield and aroma quality of upland rice (var. pare wangi) under various types and periods of drought stress

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Abstract. *Arsa IGBA, Ariffin, Aini N, Lalel HJD. 2017. Grain yield and aroma quality of upland rice (var. pare wangi) under various types and periods of drought stress. Trop Drylands 1: 17-23.* Rice with unique aroma is often preferable and this trait can be achieved through exposing rice plants to drought and salinity stresses. Yet, particular rice varieties might respond differently to such stresses. The objective of this study was to evaluate the effect of type and period of drought stress on grain yield, physiological characters and aroma quality of aromatic Pare Wangi upland rice variety. Two-factor experiment was conducted in a greenhouse employing a Completely Randomized Design with three replications. The first factor was type of drought stress and the second factor was plant growth stage at which drought stress period was started. Observed data included yield components and grain yield pot^{-1}, physiological characters, and aroma quality. Observed data were subjected to analysis of variance, HSD post hoc test, and simple correlation analysis. The research results indicated that the salinity stress effect of 2.0 g NaCl kg soil^{-1} caused a higher percentage of unfilled grain as compared to that of other types of stresses. When the salinity stress was started at the booting stage, the chlorophyll content of the rice plants was lower than that of other stresses. Soil moisture stress of 75% FC that was started at booting stage produced the highest 2AP content and aroma quality. Proline content was not significantly correlated with 2AP content but it showed a positive correlation with grain yield pot^{-1}.

Keywords: Upland rice, drought stress, aroma, proline, chlorophyll

Abbreviations: FC: Field Capacity, DAS: days after seeding, 2AP: 2-Acetil-1-Pyrroline, HSD: Honestly Significant Difference

INTRODUCTION

Crops are generally cultivated under optimum environmental conditions with minimal environmental stresses. Nonetheless, environmental stress is sometimes needed to obtain the desired traits. For example, it was reported that drought stress, to a certain extent, is useful to increase rice aroma quality of aromatic rice (Buttery et al. 1983; Yoshihashi 2005). This is the case for the aromatic Pare Wangi upland rice variety, a superior local upland rice variety that is now widely cultivated by farmers in South West Sumba District, East Nusa Tenggara Province, Indonesia. The local growers prefer Pare Wangi variety due to its taste and scent aroma, in addition to the higher selling price of the aromatic rice as compared to the non-aromatic ones.

The mechanism of the appearance of aroma in rice in relation to drought stress can be explained as follow. Drought stress results in the increase of various osmotic compounds (Bianco et al. 2000), one of which from nitrogen group is proline amino acid (Mohammadkhani and Heidari 2008). In non-aromatic rice, proline will change into glutamate acid whilst in the aromatic rice, biosynthesis process will produce 2-Acetil-1-Pyrrolin or 2AP (Kibria et al. 2008; Fitzgerald et al. 2010). The difference in biosynthesis is associated with a gene mutation in chromosome number 8, exon 7 that is responsible for the missing of aldehyde-betaine-enzyme activity ($BADH_2$) (Bradbury 2009).

The increase of drought stress may be caused by a reduction in water supply from plant roots and excessive transpiration rate during the life cycle of the plant (Farooq et al. 2008, 2009). The reduction in water supply to the plant can be caused by a decrease in soil moisture and sometimes induced by an increase in soil salinity (Levitt 1980). For instance, Yoshihashi et al. (2004) reported the trial of Khao Dawk Mali 105 aromatic rice variety that found the 2AP content of rice planted on the sites with clay soil texture was lower (388 ppb) than that planted on sandy soil texture (518-528 ppb). Roychoudhury (2008) reported a significant increase of rice aroma quality in Gobindobhog variety that was subjected to salinity stresses compared to the control or without salinity treatment, and aroma quality of Gobindobhog variety was also higher than that of the non-aromatic rice variety.

In addition to drought stress, either because of the soil water deficit or the increase in soil salinity, the increase of 2AP content of aromatic rice plants may also be caused by soil nutrient factors such as Zn content (Jin-xia et al. 2009), P content (Rohilla et al. 2000), N content (Yang et al. 2012), and organic matter content (Champagne 2008; Islam and Sikder 2011). In general, it is reported that the balance of macro and micronutrients most significantly affects the increase of aroma quality of aromatic rice (Ram et al. 2013).

The mechanisms of interaction between drought stress and nutrient balance factor in increasing the 2AP content of rice are not well understood until now. The negative correlation between aroma quality and productivity (Arsa et al. 2011; Yang et al. 2012) is possibly associated with the interaction of the above factors, implying that the increase in aroma quality is at the expense of the decrease in productivity. The lower level of drought stress will cause the amount of proline formed to be low enough to inhibit the nutrient uptake. On the other hand, high-level drought stress will induce a high amount of proline that may inhibit nutrient uptake. Thus, the amount of proline and the availability of nutrients in low or high-stress levels are not the optimum conditions for 2AP compound production. Therefore, it is necessary to determine the moderate level of drought stress for a maximum 2AP production. This can be done by arranging drought stress treatments; levels of soil moisture and or soil salinity and drought stress periods. Drought stress occurs during the phase of vegetative to harvest will give higher stress than that during the phase of reproductive or flowering to harvest. Differences in the length of drought stress period will affect the physiological processes of plants, which in turn affect the yield components and grain yield (Akram et al. 2013) and influence the aroma quality of rice.

Based on the above description, the objective of the present study was to evaluate the effect of drought stress by arranging the type and the length of drought stress period on grain yield, physiological characters, and aroma quality of the aromatic Pare Wangi upland rice variety.

MATERIALS AND METHODS

Experimental design

This study employed a Completely Randomized Design with a 3 x 3 factorial treatment design. The first factor was type of drought stress (K), which consisted of 3 levels, i.e. soil moisture of 75% Field Capacity (FC) without NaCl (k1), soil moisture of 100% FC and 2.0 g NaCl kg $soil^{-1}$ (k2), and soil moisture of 75% FC and 1.0 g NaCl kg $soil^{-1}$ (k3). The second factor was drought stress period (F), which consisted of 3 levels, i.e. panicle initiation phase (at 60 DAS) (f1), booting phase (at 80 DAS) (f2), and panicle heading phase (at 100 DAS) (f3). A total of 9 treatment combinations were applied, each was 3 replicates. In total, 27 experimental units were included in the present study. Each experiment unit consisted of 2 planting pots (one pot for destructive and the other for non-destructive observations). Variables observed in this study included (i) yield component characters (number of panicles, number of grains (grains $panicle^{-1}$), unfilled grains (%), weight of 100 grains (g), grain yield pot^{-1} (g)), (ii) leaf tissue analysis, (iii) physiological characters (chlorophyll content and free proline content), and (iv) rice aroma quality (2AP content and aroma score).

Preparation of media, planting, and fertilization

Planting media used in the present study was soil taken from the area where the Pare Wangi upland rice variety has always been cultivated. The soil was first cleaned from plants debris and sieved with a 2.0 mm sieve size. Then, 7 kg of the sun-dried soil was filled into each planting pot. Each pot was then labeled and arranged in a greenhouse following the employed treatment design. Before planting, each pot was watered following the drought stress treatments with the amount of water given into the pot following the method of Wargadiputra and Harran (1983). Planting was done using five seeds per pot, and three healthy seedlings were retained two weeks after germination. The plants were maintained until harvest. Fertilizers were applied at planting time with, respectively, 250 kg Urea ha^{-1} (1.0 g Urea pot^{-1}), 100 kg SP36 ha^{-1} (0.4 g SP36 pot^{-1} and 200 kg KCl ha^{-1} (0.8 g KCl pot^{-1}). Weed was manually controlled throughout the experiment, while the pests were controlled by insecticide (Demolish 18 EC) spray once a week during flowering and grain filling stages.

Treatment application

The initial soil moisture for each pot was maintained at 100% FC (w/w), whereas drought stress treatments were given in a growth stage of the plant according to the assigned treatment. The addition of water to meet the drought stress treatment was given by gravimetric method. The NaCl was given according to the assigned treatment by dissolving the NaCl in water prior to its application to the growing media.

Harvesting and grain sampling

Harvesting was conducted when the panicles turned yellow and hardened. Harvest was done on the whole grain yields in non-destructive pots. The grain yields were then observed for yield components and grain yield per pot. Sufficient amount of grain was then used to test both rice 2AP content and organoleptic assay.

Leaf tissue analysis

Leaf tissue analysis was carried out at flowering stage (110 DAS). Two upper leaves (the second and the third leaf below the flag leaf) of each plant in destructive pot units were taken then were bulked for all replicates. A total of 9 composite leaf samples were produced which were then used to determine the leaf tissue contents of N, P, K, Na, and Zn by using Atomic Absorption Spectrophotometer (Perkin-Elmer 3110; J&W Scientific, Folsom, CA, USA) following the standard method of AOAC (2000).

Measurement of chlorophyll content

Chlorophyll content was measured as follows: 2 g leaf sample was crushed with a mortar, and then added with 10 ml of 80% acetone. After that, the filtrate was poured through a funnel by a filter paper into a glass flask. The filtered filtrate was taken as many as 1 mL, and then diluted to a volume of 10 mL. This filtrate absorbance was measured by using a spectrophotometer. Total chlorophyll content was then calculated with the standard formula according to Arnon (1949), i.e. the sum of chlorophyll a and chlorophyll b. This procedure was applied to all leaf samples used to measure total chlorophyll content. The formula for calculation of chlorophyll-a (mg L^{-1}) was= 12.70 x OD663-2.69 x OD645, while the chlorophyll-b (mg L^{-1}) was= 22.9 x OD645-4.68 x OD663.The unit was then converted into mg g^{-1} fresh weight.

Level of free proline

The level of free proline (KPB) was determined following the procedure of Bates et al. (1973). A total of refined 0.5 g leaf sample was added with 10 mL of 3% (v/v) sulfosalisilic and then stirred. The filtrate was then filtered through a filter paper (Whatman No.40). Two mL of the filtrate were taken and reacted with 2 mL of acidic ninhydrin and 2 mL of glacial acetic acid. This process was carried out in a test tube at 100 °C for 1 hour and was then terminated by immersing the reaction tube in cold water (liquid ice). Proline extracts were obtained by adding 4 mL of toluene to the filtrate mixture for 15-20 seconds, and then stirred with a stirrer (stirrer test tube) and kept at room temperature to allow separation of toluene and water phases. Toluene phase absorbance was measured with a spectrophotometer at a wavelength of 520 nm (toluene was used as a blank). Total KPB was calculated by regression curve generated using the standard solutions.

Measurement of 2AP content and aroma score

The rice 2AP content was measured using composite samples of each treatment following the method of Lalel et al. (2003) and Wongpornchai et al. (2004**)**. The volatile compounds were extracted using headspace solid-phase micro-extraction (HS-SPME) technique with the 100 µm poly dimethyl siloxane SPME manual device (Supelco Co., Bellefonte, PA, USA). Separation and quantification of the 2AP compound were achieved using GC-MS (Hewlett Packard 5890 series, USA) equipped with a DB5MS capillary column (50 m x 0.2 mm id., 0.33 µm film thickness; J & W Scientific, Folsom, CA, USA). Total 2AP was calculated using external standard (calibration curve). The organoleptic assay was conducted by ten trained panelists to assess the rice aroma score following the modified method of Lestari et al. (2011). Rice was cooked in a test tube for 15 minutes and kept warm at 40ºC. Rice aroma quality was determined by scoring in a range of 0-4 (no aroma to very strong aroma).

Statistical analysis

The observed data were subjected to analysis of variance (ANOVA) following a Completely Randomized Design approach. An HSD post hoc test at 5% significance level was then conducted to compare the treatment means. Simple correlation analysis was also performed to examine the correlation between variables.

RESULTS AND DISCUSSION

Yield components and grain yield

Our research results revealed that type of drought stress treatment caused no significant effect on the number of panicles, number of grains, weight of 100 grains and grain yield per pot but the treatment significantly affected the percentage of unfilled grains. The drought stress induced by soil moisture of 100% FC and 2.0 g NaCl kg $soil^{-1}$ (k2 treatment) caused a significantly higher unfilled grain percentage than that of the other treatments. Furthermore, the effect of drought stress period started at booting phase (at 80 DAS) (f2 treatment) caused no significant effect on the observed variables except the 100-grain weight. Drought stress initiated at booting phase (f2) produced a significantly lower 100-grain weight than that started at panicle initiation phase (f1) and panicle heading phase (f3). Grain yield per pot did not differ among the treatments of drought stress period (Table 1).

Chlorophyll and proline content

Drought stress type induced by soil moisture of 75% Field Capacity (FC) without NaCl (k1) and soil moisture of 75% FC and 1.0 g NaCl kg $soil^{-1}$ (k3) caused no significant effect on chlorophyll content, on the other hand, drought stress induced by soil moisture of 100% FC and 2.0 g NaCl kg $soil^{-1}$ (k2) started at booting phase (f2) produced a lower chlorophyll content as compared to that started at panicle initiation phase (f1) or panicle heading phase (f3) (Table 2). Furthermore, k1 treatment produced higher proline content than that of either k2 or k3 treatments, respectively. Drought stress type started at f1 stage also produced higher proline content as compared to that started at f2 and f3 (Table 2).

Table 1. Yield components and yield of Pare Wangi upland rice variety under various types and periods of drought stress

Treatment (s)[1]	**Yield components and yield**[2]				
	NP	**NG**	**UG**	**W100**	**GY**
Drought stress type					
k1	11.11a	144.00a	22.64b	2.29a	29.01a[3]
k2	12.56a	162.56a	32.82a	2.31a	28.01a
k3	12.00a	165.67a	23.46b	2.35a	28.35a
HSD 0.05	2.29	34.33	7.10	0.17	6.06
Drought stress period					
f1	12.11a	155.00a	28.16a	2.42a	29.21a
f2	11.22a	159.67a	24.46a	2.23b	27.84a
f3	12.33a	155.56a	26.30a	2.30ab	28.31a
HSD 0.05	2.29	34.33	7.10	0.17	6.06

Note: [1] k1: soil moisture of 75% FC + 0 g NaCl; k2: soil moisture of 100% FC + 2.0 g NaCl kg $soil^{-1}$; k3: soil moisture of 75% FC + 1.0 g NaCl kg $soil^{-1}$; f1: drought stress initiated at 60 DAS; f2: drought stress initiated at 80 DAS; and f3: drought stress initiated at 100 DAS. [2] NP: number of panicles pot^{-1}, NG: number of grains (grains $panicle^{-1}$), UG: unfilled grains (%), W100: weight of 100 grains (g), GY: grain yield pot^{-1} (g). [3] Numbers followed by the same letter (s) are not significantly different at HSD post hoc test (0.05).

Table 2. Chlorophyll and proline content of Pare Wangi upland rice variety under various types and periods of drought stress

Treatment[1)]	Chlorophyll (mg g BS^{-1})			Average	Proline (µmol g BS^{-1})			Average
	f1	f2	f3		f1	f2	f3	
k1	1.29 a A	1.37 a AB	1.15 a A	1.27	15.60	14.91	14.21	14.91 A[2)]
k2	1.43 a A	1.12 b B	1.34 ab A	1.30	14.88	14.32	14.47	14.56 AB
k3	1.30 a A	1.41 a A	1.25 a A	1.32	14.38	13.85	14.23	14.15 B
Average	1.34	1.30	1.25	1.30	14.95 a	14.36 b	14.30 b	14.54

Note: [1)] k1: soil moisture of 75% FC + 0 g NaCl; k2: soil moisture of 100% FC + 2.0 g NaCl kg soil^{-1}; k3: soil moisture of 75% FC + 1.0 g NaCl kg soil^{-1}; f1: drought stress initiated at 60 DAS; f2: drought stress initiated at 80 DAS; and f3: drought stress initiated at 100 DAS. [2)] Means followed by the same letter (s) are not significantly different at HSD post hoc test (0.05) (small letter shows comparisons in the same row and capital letter shows comparisons in the same column)

Content of 2AP, aroma score, and leaf tissue nutrient

We found in the present study that the highest 2AP content of rice induced by drought stress type due to soil moisture content of 75% FC + 0 g NaCl (k1) was produced when the stress was applied at booting phase (f2) while drought stress type induced by soil moisture of 100% FC + 2.0 g NaCl kg soil^{-1} (k2) and soil moisture of 75% FC + 1.0 g NaCl kg soil^{-1} (k3), respectively, produced the highest 2AP content when these treatments were initiated at panicle heading phase (f3). The k1 treatment applied at f2 stage apparently produced the highest 2AP content (3.07 ppb) (Table 3). Aroma scores of rice determined through organoleptic test fell in the range of 1.7 to 2.2. The drought stress type of k1 treatment applied at f2 stage showed the highest rice aroma score and that of k2 treatment applied at panicle initiation phase (f1) showed the lowest one. The range of aroma scores produced by either k1 treatment or k3 treatment was relatively higher than that of k2 treatment. These findings showed that drought stress caused by water deficit or its combination with salinity application was stronger than the salinity application alone (without water deficit) in increasing the rice aroma (Table 3).

The effect of the single treatment drought stress type (K) and drought stress period (F) on the leaf tissue contents of Na, N, P, K, and Zn are presented in Table 4. Na content of leaf tissue treated with k2 (soil moisture of 100% FC + 2.0 g NaCl kg soil^{-1}) was higher than that of either k1 (soil moisture of 75% FC + 0 g NaCl) or k3 (soil moisture of 75% FC + 1.0 g NaCl kg soil-^{1}) over all periods of drought stress. Effect of k3 treatment tended to increase the leaf tissue Na content as compared to k1 treatment but there was no significant effect of drought stress type and period on leaf tissue contents of N, P, K, and Zn.

Correlation between plant characters

Results of correlation analysis between yield components showed that number of grains (grain panicle^{-1}) was negatively correlated with number of panicles but there was no correlation between 100-grain weight and number of grains. The correlation analysis also revealed that none of the observed yield components was correlated with grain yield pot^{-1} (Table 5). The present study results also showed that N content has no correlation with either each of the yield components or the grain yield pot^{-1}. Similarly, no correlation was observed between Na content of leaf tissue and each of the following characters, respectively: yield components, grain yield pot^{-1}, and Na content. Furthermore, chlorophyll content showed only positive correlation with 100-grain weight while proline content apparently has a positive correlation with grain yield pot^{-1} but it was not correlated with rice 2AP content (Table 5).

Table 3. 2AP and aroma score of Pare Wangi upland rice variety under various types and periods of drought stress

Treatment[1)]	2AP (ppb) +			Aroma Score +		
	f1	f2	f3	f1	f2	f3
k1	2.92	3.07	2.45	2.0	2.2	2.1
k2	2.36	2.01	2.65	1.7	1.8	1.9
k3	2.30	2.33	2.71	2.0	2.0	2.1

Note: [1)] k1: soil moisture of 75% FC + 0 g NaCl; k2: soil moisture of 100% FC + 2.0 g NaCl kg soil^{-1}; k3: soil moisture of 75% FC + 1.0 g NaCl kg soil-^{1}; f1: drought stress initiated at 60 DAS; f2: drought stress initiated at 80 DAS; and f3: drought stress initiated at 100 DAS; +: data was not subjected ANOVA

Table 4. Content of Na, N, P, K and Zn in leaf tissues of Pare Wangi upland rice variety under various types and periods of drought stress

Treatment (s)[1)]	Content in leaf tissues				
	Na (%)	N (%)	P (%)	K (%)	Zn (ppm)
Drought stress type					
k1	0.79	2.19	0.83	1.48	81.16
k2	1.17	2.19	0.87	1.49	80.51
k3	0.91	2.24	0.82	1.55	80.60
Drought stress period					
f1	0.92	2.19	0.82	1.51	80.58
f2	0.95	2.29	0.86	1.48	81.11
f3	1.00	2.14	0.85	1.54	80.57

Note: [1)] k1: soil moisture of 75% FC + 0 g NaCl; k2: soil moisture of 100% FC + 2.0 g NaCl kg soil^{-1}; k3: soil moisture of 75% FC + 1.0 g NaCl kg soil-^{1}; f1: drought stress initiated at 60 DAS; f2: drought stress initiated at 80 DAS; and f3: drought stress initiated at 100 DAS

Table 5. Results of simple correlation analysis between observed variables

Variable	JG	W100	GY	N (%)	Na (%)	Klo	Pro	2AP	Aroma
NP	-0.63 **	0.55 ns	-0.31 ns	-0.54 ns	0.46 ns	0.21 ns	-0.10ns	-0.11ns	-0.29ns
NG		-0.34 ns	0.14 ns	0.46 ns	0.18 ns	-0.18 ns	-0.22ns	-0.36ns	-0.24ns
W100			0.50 ns	-0.02 ns	-0.09 ns	0.70 *	0.36ns	0.30ns	-0.08ns
GY				0.19 ns	-0.56 ns	0.26 ns	0.73*	0.59ns	0.22ns
N (%)					-0.18 ns	0.60 ns	0.03ns	0.23ns	0.12ns
Na (%)						-0.08 ns	-0.35ns	-0.65 *	-0.83 **
Chlo							0.20ns	0.31ns	-0.04 ns
Pro								0.59ns	-0.03 ns
2AP									0.66*

Note: ns: not significant; *,**: significant at p-value≤ 0.05 and p≤ 0.01, respectively. NP: number of panicles pot^{-1} ; NG: number of grains (grains panicle^{-1}); W100: weight of 100 grains (g); GY: grain yield pot^{-1} (g); Chlo: Chlorophyll content (mgg BS^{-1}); Pro: Prolin content (μmol g BS^{-1}); 2AP: 2AP content (ppb), Aroma: Aroma score

Discussion

Number of unfilled grains under the drought stress type of k2 treatment (2.0 g NaCl kg soil^{-1}) was higher than that of either k1 treatment (75% FC and 0 g NaCl) or k3 treatment (75% FC and 1.0 g NaCl kg soil^{-1}) (Table 1). This indicated a higher seed abortion rate under the k2 treatment as compared to either k1 or k3 treatments. The enhancement of Na uptake in k2 treatment (Table 4) showed a tendency of positive correlation with number of panicles but number of panicles has a negative correlation with number of grains, which caused Na content of leaf tissue has a negative correlation with grain yield pot^{-1} (Table 5). These findings indicated that k2 treatment affected the photosynthesis process, which hereinafter affected carbohydrate formation and its translocation to the seeds. In the end, lack of carbohydrates caused more seeds not to be fully formed or to become unfilled grains (Jaleel et al. 2009).

Drought stress that was started at f2 stage (80 DAS) caused the plant to produce a lower 100-grain weight compared to that at f1 stage (60 DAS) but it was not significantly different from that at f3 stage (100 DAS) (Table 1). This implies that the supply of carbohydrates from leaves to the seeds was more significantly reduced when the drought stress was started at f2 than at f1. Plants that revived drought stress treatment at f2 stage exhibited leaf chlorosis symptoms and the leaves were then dried out. The symptom was mainly related to the influence of k2f2 treatment that significantly reduced leaf chlorophyll content as compared to k1f2 and k3f2 treatments (Table 2). The decrease in leaf chlorophyll content affected the effectiveness of the photosynthesis process, which in turn reduced the amount of carbohydrates translocated to the seeds.

The tendency of the decrease in leaf chlorophyll content of the plants treated with drought stress types k1 and k3 at the drought stress period f3 (Table 2) was presumably caused by the plant's adjustment mechanisms. At the drought stress treatment period f3, the plants grew without drought stress in the vegetative stage but were then subjected to drought stress in the generative stage, especially close to the flowering stage when the plants' water consumption increased significantly. One of the plant's adjustment mechanisms in facing such situation was by reducing new chlorophyll formation, and otherwise by increasing chlorophyll molecule degradation. As a result, the decreased leaf chlorophyll content of plants treated with k2 drought stress type at f2 might be related to the increase of leaf tissue Na content (Table 4) triggered by the increase of transpiration process. Visually, this process caused the older thick leaf to be drying out.

Observed rice 2AP content and aroma score presented in Table 3 indicated that the range of 2AP content of plants treated with drought stress type k1 was higher than that of other treatments, either k2 or k3. This might have been caused by the increase of salinity that affected the biosynthesis of proline to become 2AP compound. At the drought stress type k1, it was likely that more proline was used to form 2AP compound as compared to that of k2 and k3. Meanwhile, at the drought stress types k2 and k3, the formation of proline might be more useful as an osmoprotectant than as a precursor of 2AP compound. This was mainly seen in the drought stress types started at f1 and f2 stages, where lower 2AP contents were produced compared to that at f3. On the contrary, the drought stress type k1 started at f3 stage was also likely to reduce proline content (as a precursor of 2AP compound), so that the 2AP content at this treatment was lower than that of k1 started at either f1 or f2 stages (Table 3). Gay et al. (2010) reported that the formation of rice 2AP compound was predicted to occur before heading phase; meanwhile, Itaniet et al. (2004) found that the highest 2AP content was reached at 4-5 weeks after heading phase. Therefore, the different treatments of drought stress levels and starting periods applied in this study presumably determined the observed difference in time (phase) for the highest rice-2AP production we found in the present study.

Grain yield pot^{-1} was positively correlated with 100-seed weight, meanwhile, number of panicles plant^{-1} and number of grains panicle^{-1} showed a negative correlation (Table 4), and consequently, these two yield components have no contribution to grain yield pot^{-1}. Grain yield pot^{-1} was mainly determined by 100-seed weight that was positively correlated with number of panicles. Shahidullah et al. (2009) reported that in aromatic rice, number of panicles was positively correlated with grain yield but its effect has frequently occurred indirectly through other characters such as 100-seed weight and number of grains panicle^{-1}.

There was no correlation between grain yield pot^{-1} and leaf tissue N content, which may likely be related to the observed negative correlation between leaf tissue N content and number of panicles, and on the other side, there was a positive correlation between leaf tissue N content and number of grains panicle^{-1}. It was also revealed in the present study that there was no correlation between Na content and grain yield pot^{-1}. The non-significant positive correlation between leaf tissue Na content and number of panicles, in addition to the negative correlation between number of panicles and number of grains, maybe the causal factor of the non-significant negative correlation between leaf tissue Na content and the grain yield pot^{-1} (Table 5).

The observed positive correlation between rice 2AP content and grain yield pot^{-1} (Table 5) shows that the increase of rice 2AP content will be followed by the increase of grain yield pot^{-1} if the plants were exposed to a moderate level of drought stress after generative phase. This phenomenon was likely to involve endogen proline activity. The significant positive correlation observed between proline and grain yield pot^{-1} was also supported by the tendency of a positive correlation between the leaf tissue proline content and the rice 2AP content. Thus, there is a tendency for a positive correlation between rice 2AP content and grain yield pot^{-1}. The positive correlation between proline content and rice 2AP content observed in the present study demonstrates that Pare Wangi rice variety has experienced a moderate level of drought stress. In contrast to this finding, previous studies (Kibria et al. 2008; Yang et al. 2012) reported that 2AP content of rice was negatively correlated with grain yield.

Based on the results of the present study, the following conclusions are made: (i) The treatment interaction of type and period of drought stress did not affect the grain yield pot^{-1} but affected both leaf tissue chlorophyll content and rice 2AP-compound content. (ii) The main effect of drought stress type separately affected the percentage of unfilled grains, and the drought stress period affected the 100-grain weight. (iii) The percentage of unfilled grains caused by salinity level of 2.0 g NaCl kg soil-^{1} was higher than that caused by soil moisture of 75% FC and the combination of the treatments (soil moisture of 75% FC and salinity level of 1.0 g NaCl kg soil-^{1}). (iv) Stress treatment of salinity level of 2.0 g NaCl kg soil-^{1} started at booting stage (80 DAS) produced lower chlorophyll content as compared to other treatments. (v) Drought stress type of soil moisture of 75% FC started at booting stage produced the highest rice 2AP-compound content and aroma score. (vi) Proline content of leaf tissue has no correlation with rice 2AP- compound content but it has a positive correlation with grain yield pot^{-1}.

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Yield performance of eight mungbean (*Phaseolus radiatus*) genotypes in two locations in Manggarai District, East Nusa Tenggara, Indonesia

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Abstract. *Mau YS, Madu1VFY, Ndiwa ASS, Adar D, Gandut YRY. 2017. Yield performance of eight mungbean (Phaseolus radiatus) genotypes in two locations in Manggarai District, East Nusa Tenggara, Indonesia. Trop Drylands 1: 24-31.* Mungbean [*Phaseolus radiatus* (L.) Wilczek] is among most important crops cultivated in arid and semi-arid regions. Yet, various genotypes of mungbean are available which might differ in yield productivity across locations. The objective of this study was to elucidate yield performance of eight mungbean genotypes at two locations representing different altitudes (i.e. Murai and Watu Baur) in Manggarai District, East Nusa Tenggara Province, Indonesia. A Randomized Block Design was employed consisting of 8 treatments, each with 3 replicates. The assigned treatments were eight mungbean genotypes consisting of one local cultivar and seven Indonesian released superior varieties. Observed variables included yield components and yield of mungbean. Data from each location was subjected to ANOVA, followed by a combined ANOVA involving data from both locations. A post hoc test of DMRT (5%) was performed to compare the treatment means. The study results showed that genotype x location interaction significantly or highly significantly affected most of the observed variables, except plant height at flowering, plant height at harvest, productive branches plant^{-1} and 100-seed weight. Vima-1 and Murai were found to perform better than other genotypes in most yield components and yield traits in both locations. The highest mean seed yield over two locations was produced by Murai (991.87 g plot^{-1}) and Vima-1 (967.90 g plot^{-1}). Mean seed yield of all genotypes at Perak was 365.0 g plot^{-1} while that in Watu Baur was 761.27 g plot^{-1}.

Keywords: Genotype, location, mungbean, performance, yield

INTRODUCTION

Mungbean [*Phaseolus radiatus* (L.**)** Wilczek], also known as *green gram* or *golden gram*, is a dryland arid and semi-arid pulse crop, mostly cultivated as the second crop after maize, rice, sorghum, etc. The crop is well known for its nutritional value and health benefits, and is also used as fodder. In addition to its high protein and carbohydrate contents (Kumar et al. 2014; Dahiya, et al. 2015), mungbean contains high levels of folate and iron (Keatinge et al. 2011). From the perspective of agro-ecology, mungbean is known as a soil nitrogen enhancing crop through its ability to fix atmospheric nitrogen via root rhizobial symbiosis (Graham and Vance 2003). These advantageous characters render the mungbean an important crop in the tropical and sub-tropical regions of the world, including Indonesia.

In the subsistent agriculture system of semi-arid region of East Nusa Tenggara (ENT) Province, Indonesia, mungbean plays an important role as an affordable dietary protein source as well as a cash crop to the farmers. Mungbean is known to be well adapted to the semi-arid growing condition such as in ENT Province. The short duration type of most mungbean cultivars (55-60 days) makes this crop more suitable to be planted in this region as the second crop after paddy rice or maize. The crop is also easy to manage, low chance of harvest failure, high economic value, and can be easily prepared for consumption.

ENT Province is one of the mungbean production centers in Indonesia. During the last few years, however, mungbean production in the province fluctuated significantly due to many factors. It was revealed in the data released by ENT Province Central Bureau of Statistics (2012) that total mungbean production in ENT in 2009 was13,462 tons, which then decreased to only 6,985 tons in 2010 and increased again to 10,408 tons in 2011. In addition, the productivity of mungbean in the region is still low, with only about 0.9 t ha^{-1} seed yield (ENT Central Bureau of Statistics 2012). The fluctuation in mungbean production is mainly due to fluctuation in the planting area and unfavorable climatic conditions such as drought. Meanwhile, poor agricultural practices and the use of low-yielding varieties are the main contributors to low mungbean productivity in this province.

In addition to the above-mentioned problems in mungbean production and productivity, water shortage and foliar diseases such as *Cercospora* leaf spot, rust, powdery mildew, *scab*, and viruses are other factors that also contributed significantly to the low mungbean production (Chand et al. 2015; Gnanaraj et al. 2015; Kumar et al. 2017; Parihar et al. 2017). The use of superior varieties is, therefore, considered the most effective and efficient way of overcoming the above-mentioned mungbean production constraints. The use of superior varieties is cheaper, easily adopted by farmers, and environmentally friendly. The availability of high-yielding varieties coupled with resistance to foliar diseases and drought stress would be a significant contribution in preventing the crop from harvest failure and hence, increasing the farmer's income.

A number of superior mungbean varieties have been produced by Indonesian Legume and Tuber Crops Research Institute (ILETRI); however, these varieties have not been extensively cultivated by the farmers in ENT Province, especially in Manggarai District (one of the mungbean production centers in the province) due to lack of information and access to these varieties by the farmers. Further, irrespective of their release as superior varieties, in fact, these varieties have not been evaluated for yield performance in ENT Province, which has a unique climatic condition (a semi-arid type) compared to that of the rest of Indonesian provinces. Such varieties have only been evaluated mostly in Java and Sumatera Islands which have a more humid climatic condition. It is, therefore, necessary to evaluate the performance of these superior varieties in Manggarai District to ensure their adaptability to the region. The objective of this study was to elucidate the yield and yield components performances of Indonesian released superior mungbean varieties and a local cultivar in two locations in Manggarai District. The most suitable genotypes for each of the two locations were also determined in the present study.

MATERIALS AND METHODS

Research location and design

This research was conducted in the field in two locations during the dry season (June to September) 2013. The two locations were selected to represent mungbean production centers in the district and also to represent two different altitudes, i.e. the low altitude at Watu Baur Village, Reok Sub-District (32 m asl), and the medium altitude at Perak Village, Cibal Sub-District (745 m asl).

The assigned treatment was mungbean genotypes, consisting of one local cultivar of Manggarai District (Local Manggarai) and seven Indonesian released superior varieties (Vima-1, Murai, Perkutut, Kenari, Betet, Walet, and Kutilang) making up a total of 8 treatments. The experiment in each location was laid out in a Randomized Block Design with each treatment consisting of three replicates. In total, 24 experimental units were included in each growing location.

Land preparation and crop planting

In each growing location, the planting site was first cleared from weeds and woods before plowing. The planting site was then sub-divided into three blocks, each was 100 cm apart. Each block was further sub-divided into 8 plots of 3 m x 2 m size with 50 cm distance between plots. In total, 24 planting plots were prepared in each growing location.

Mungbean seeds were planted about 3 cm depth inside the planting holes with a planting space of 40 cm x 20 cm. Two plants were maintained in each planting hole. Fertilizer was used in the form of a compound NPK with a dosage of 100 kg ha^{-1}, equal to 40 g plot^{-1}, applied at planting time. Irrigation was provided daily to meet approximately 70-80% field capacity during the crop growth and development. Weeding was done manually, and pests and diseases control were conducted mechanically and chemically using insecticide and fungicide sprays. Harvesting was carried out when almost all pods in each plant within the plot started to turn black or brown (depending on the genotype) and were easy to explode. Harvesting was done manually.

Observed variables and data analysis

Observed variables included plant height at flowering (cm), harvesting date (DAP), number of productive branches plant^{-1}, number of pod clusters plant^{-1}, number of pods plant^{-1}, pod weight plant^{-1}, 100-seed weight (g), seed weight plant^{-1} (g) and seed weight plot^{-1} (g).

The observed data from each growing location was subjected to analysis of variance using Randomized Block Design approach. A homogeneity variance analysis was then conducted prior to a combined analysis of variance employing data from both locations to determine genotype by location interaction effect on the observed variables. A post hoc test of Duncan Multiple Range Test was also carried out to compare the treatment means. Analysis of variance was carried out using Genstat Version 12 (VSNi 2009).

RESULTS AND DISCUSSION

The treatment effect on the observed variables

We carried out a Bartlet test of homogeneity of variance involving data collected from the two locations prior to combined analysis of variance to determine the genotype by location interaction, and the single factors genotype and location effects on the observed variables. The effect of the treatment on the observed variables is presented in Table 1.

Table 1. Combined analysis of variance of yield component and yield variables of eight mungbean genotypes tested in two locations.

Source of Variation	DF	F-value (calculated)									
		Plant height at flowering	Plant height at harvest	Harvesting date	Productive branches plant^{-1}	Pod clusters plant^{-1}	Pods plant^{-1}	Pod weight plant^{-1}	100-seed weight	Seed weight plant^{-1}	Seed weight plot^{-1}
Location (L)	1	252.28**	243.23**	690.22**	0.90ns	95.81**	73.45**	68.91**	101.67**	73.62**	594.93**
Genotype (G)	7	19.57 **	20.08**	31.26**	9.45**	0.46ns	1.10ns	3.63*	1571.70**	4.79*	5.32*
G x L	7	1.19ns	1.12 ns	36.54**	0.31ns	4.28**	2.87*	4.36**	0.62ns	4.88**	498.67**
CV (%)		6.93	7.02	3.91	9.44	13.14	12.27	10.82	2.68	11.33	10.7

Note: DF: Degree of freedom, **highly significant (P<0.01), *significant (P<0.05), nsnot significant (P>0.05).

Results of pooled ANOVA (Table 1) show a varying treatment effect on the observed variables. Highly significant interaction effect of genotype x location was observed on harvesting date, pod clusters plant^{-1}, pod weight plant^{-1}, seed weight plant^{-1}, and seed weight plat^{-1}. The genotype x location interaction significantly affected pods plant^{-1} but caused no effect on plant height at flowering and harvesting stages, productive branches plant^{-1} and 100-seed weight. Meanwhile, the single factor genotype significantly affected all observed variables except productive branches plant^{-1} while location significantly/highly significantly affected almost all of the observed variables except pod clusters plant^{-1} and pods plant^{-1}.

Significant effect on the interaction between genotype and location implies that the mungbean genotypes were not consistent in their ranks in the observed variables, where the ranks changed following the change of the growing location. The existence of genotype by location may also indicate a specific adaptation of a genotype in a specific location for a certain observed variable. Detailed descriptions of the observed variables are presented below.

Plant height at flowering and harvesting stages

Genotype by location caused no significant effect on the plant height at both flowering and harvesting stages but singly; either mungbean genotype or growing location significantly affected the traits. Data presented in Table 2 show that mean plant height at flowering stage differed significantly among the tested genotypes with Local Manggarai produced the highest (74.5 cm) and Vima-1 produced the shortest (48.8 cm) plant heights (Table 2). The same situation occurred at harvesting stage where Local Manggarai and Vima-1 produced, respectively, the highest and shortest plant heights. Besides, plant heights of other six genotypes ranged, respectively, from 54.6 cm to 68.5 cm at flowering stage and 55.2 cm to 69.1 cm at harvesting stage. The study results also demonstrated that the plant height of each genotype at harvesting was a bit higher but not substantially different from that at flowering, which may indicate the determinate growth type of the tested mungbean genotypes. Local Manggarai produced significantly higher plant height than the released superior varieties at both flowering and harvesting stages, which presumably indicates its unique genetic control in this trait, in addition to its good adaptation to the growing conditions as this local cultivar has been cultivated by farmers from generation to generation.

In addition to the genotypic effect, the growing environment/location also caused a highly significant effect (P<0.01) on mean plant height of the mungbean genotypes. It is revealed in Table 2 that mean plant height was higher when the mungbean genotypes were grown in Watu Batur (low altitude) as compared to Perak (medium altitude). This situation occurred in both flowering and harvesting stages, which may explain the effect of environmental conditions such as temperature, humidity and sunlight intensity on the mungbean plant height performance. Lower altitude such as Watu Baur (32 m asl) tends to have more optimum environmental conditions such as temperature (27-29 °C) and sunlight intensity (10-12 hr day^{-1}) that are likely more favorable for mungbean growth as a C_3 plant type. The altitude influences both the temperature and sunlight intensity. The higher the altitude, the lower is the temperature and the sunlight intensity, and vice versa. Lower temperature and sunlight intensity will ultimately inhibit crop growth and development due to the reduced photosynthesis process, which in turn slows down the plant height increase. The growing conditions at Perak Village (medium altitude, 745 m asl) with a mean temperature of 18.3 °C and sunlight intensity of 8-10 hr day^{-1}, presumably, are not quite favorable for the optimal growth as shown by the significantly lower plant height of the tested mungbean genotypes in this location.

Our research results also revealed a great variation in harvesting date among mungbean genotypes tested in the two locations (Table 2). The Indonesian released variety Vima-1 was harvested earlier at 57 days after planting (DAP) when cultivated at Watu Baur (low altitude) and at 61.3 DAP when cultivated at Perak (medium latitude), while Local Manggarai was harvested the latest, respectively, 81.0 DAP at Watu Baur and 92.7 DAP at Perak. The harvesting dates of Vima-1 (57 DAP at Watu Baur and 61 DAP at Perak, respectively) approximated that of its varietal description, i.e. 58 DAP, which may indicate the genotypic stability of this trait. The longer harvesting dates of Local Manggarai at both locations were in line with the experience of the local farmers who usually take approximately 3 months to get the local mungbean genotype harvested.

As with plant height, the harvesting dates of the tested genotypes at Perak (medium altitude) were also significantly higher than those at Watu Baur (low altitude). Again, this might have been caused by the differences in environmental conditions between the two growing locations, which ultimately affected the plant's age differently. When grown at Perak with an altitude of 745 m

Table 2. Mean plant height (cm) at flowering and harvesting stages of eight mungbean genotypes tested in two locations

Genotype (G)	Plant height at flowering stage (cm)		Plant height at harvesting stage (cm)	
Lokal Manggarai	74.5	e	75.2	e
Murai	59.0	bc	59.6	bc
Betet	60.6	c	61.1	c
Vima-1	48.8	a	49.2	a
Perkutut	55.2	bc	55.7	bc
Kutilang	54.6	b	55.2	b
Walet	68.5	d	69.1	d
Kenari	57.8	bc	58.6	bc
Location (L)	**Perak**	**Watu Baur**	**Perak**	**Watu Baur**
Mean (L)	45.6 A	74.1 B	46.0 A	75.0 B

Note: Means within the same column or row with the same letter (s) are not significantly different at 0.05 DMRT. The capital letter indicates comparison within the same row while small letter indicates comparison within the same column asl., the mungbean crop tended to have low metabolism due to the lower daily temperature (18 °C), which in turn slows down the crop harvesting date. On the other hand, when the mungbean genotypes were tested at low altitude at Watu Baur (32 m asl, 27-29 °C daily temperature), the genotypes' metabolism were, presumably, higher due to the higher temperature of the growing environment. This was in line with Poehlman (1978) in Rao et al. (2016) who suggested that mean temperature of 28-30 °C is the most optimum for mungbean.

Number of productive branches plant^{-1}, pod clusters plant^{-1}, and pods plant^{-1}

In contrast to harvesting date, the present study results demonstrated no significant effect of genotype by location on productive branches plant^{-1}. The same situation was true for the single factor genotypic effect but the location significantly affected the trait. Mean performance of productive branches plant^{-1} is presented in Table 3.

Data in Table 3 show that the evaluated mungbean genotypes produced around 1.2-1.8 productive branches plant^{-1} when grown at Perak, and the range was wider and differed significantly among genotypes (1.3-2.5 branches plant^{-1}) when the genotypes were grown in the low altitude location, Watu Baur. Again, the explanation for these results is related to the different growing conditions of the two locations.

Number of productive branches plant^{-1} in Perak was lower than that in Watu Baur since the higher altitude of Perak provides less favorable conditions to the productive branches production as compared to that in the low altitude Watu Baur. When grown in Watu Baur, Murai performed the best with 2.5 productive branches plant^{-1} but was not significantly different from Vima-1. Kutilang, on the other hand, produced the lowest number of productive branches plant^{-1} (1.3) at each of both locations. The high number of productive branches plant^{-1} exhibited by both Murai and Vima-1 is supported by the more favorable conditions of low altitude growing location at Watu Baur. The crop growth and development is frequently influenced by both genetic and environmental factors so that the growth and yield performances are strongly determined by the genetic makeup of the crop and the growing conditions. The genetic potential of a crop genotype is frequently not fully expressed due to the environmental effect and, therefore, performances of some genotypes may be altered by the changing growing environments. Welsh and Mogea (1991) stated that if the difference in performance of two genotypes within the same environment can be measured, then the difference between the two genotypes results from genotypic variation that exists between the two genotypes. Furthermore, according to Allard (1992), the observed phenotypic variation expressed in the same environment does occur because the crops possess different genetic backgrounds.

Our results revealed highly significant effect ($P<0.01$) of both genotypes by location and location on the number of pod clusters plant^{-1} while the genotype alone caused no significant effect on the trait. The existence of significant genotype by location interaction effect implies changes in the rank of pod clusters plant^{-1} of the mungbean genotypes when grown in different locations. Mean pod clusters plant^{-1} presented in Table 3 shows high variation over the two growing locations. Pod clusters plant^{-1} of most genotypes was higher in Watu Baur as compared to Perak. The highest pod cluster plant^{-1} was produced by Murai (7.8 clusters) but was not substantially different from that of Betet (7.0), Kenari (6.8) and Vima-1 (6.7). By location, mean pod clusters plant^{-1} at Watu Baur was significantly higher than that at Perak.

Table 3. Mean harvesting date (DAP), productive branches plant^{-1}, pod clusters plant^{-1} and pods plant^{-1} of eight mungbean genotypes tested in two locations

Genotype (G)	Harvesting date (DAP)			Productive branches plant^{-1}		Pod clusters plant^{-1}			Pods plant^{-1}		
	Location (L)			Location (L)		Location (L)			Location (L)		
	Perak	Watu Baur	Mean (G)	Perak	Watu Baur	Perak	Watu Baur	Mean (G)	Perak	Watu Baur	Mean (G)
Local Manggarai	92.7 f B	81.0 f A	86.8 e	1.3 a	1.5 ab	5.2 b A	5.5 ab A	5.4 a	17.2 b A	22.0 b B	19.6 a
Murai	73.7 d B	63.0 e A	68.3 d	1.8 a	2.5 e	4.0 ab A	7.8 d B	59 a	14.9 ab A	24.4 bc B	19.6 a
Betet	68.7 c B	60.0 c A	64.3 bcd	1.4 a	1.8 bcd	3.6 a A	7.0 cd B	5.3 a	10.9 a A	22.5 bc B	16.7 a
Vima-1	61.3 a B	57.0 a A	59.2 a	1.6 a	2.1 de	3.7 a A	6.7 bcd B	5.2 a	12.0 a A	26.1 c B	19.1 a
Perkutut	66.7 b B	60.0 c A	63.3 abc	1.5 a	2.0 cd	4.1 ab A	6.4 bc B	5.2 a	13.4 ab A	22.6 bc B	18.0 a
Kutilang	74.3 de B	61.3 d A	67.8 cd	1.2 a	1.3 a	3.8 a A	5.0 a A	4. 5 a	12.4 a A	10.0 a B	15.1 a
Walet	66.3 b B	58.3 b A	62.3 ab	1.4 a	1.7 abc	4.3 ab A	6.2 abc B	5.2 a	13.4 ab A	23.6 bc B	18.5 a
Kenari	75.0 e B	62.7 e A	68.8 d	1.6 a	2.0 cd	4.1 ab A	6.8 bcd B	5.4 a	12.5 a A	20.9 ab B	16.7 a
Mean (L)	72.3 B	62.9 A				4.1 A	6.4 B		13.3 A	22.5 B	

Note: Means within the same coloumn or row with the same letter (s) are not significantly different at 0.05 DMRT. The capital letter indicates comparison within the same row while small letter indicates comparison within the same column.

Combined analysis of variance of pods plant^{-1} showed highly significant differences among locations (Table 3). The present study results are in line with previous studies by Ullah et al. (2011), Raturi et al. (2012) and Alia et al. (2014), who observed highly significant location and genotype by location interaction effects on pods plant^{-1} of mungbean. Data in Table 3 reveal that mean pods plant^{-1} of all genotypes at Watu Baur was about two folds that of Perak. Vima-1 produced the highest number of pods plant^{-1} (26 pods) but was not significantly deviated from that of Murai (24.4), Walet (23.6), Perkutut (22.6), and Betet (22.5). These results might have happened due to fact that these varieties are superior varieties that possess the capacity to make use of the available growth factors resources to increase their photosynthesis process before and during flowering stages to support high rate flower formation, which then resulted in high number pod formation. The fact that these varieties, particularly Murai and Vima-1, also produced the highest productive branches plant^{-1} might have also contributed to the high pods plant^{-1} of these varieties.

Pod weight plant^{-1} and seed weight plant^{-1}

The present study results revealed high variation of pod weight plant^{-1} among the tested mungbean genotypes. Like number of branches plant^{-1}, number of clusters plant^{-1} and number of pods plant^{-1}, the Indonesian released varieties Murai and Vima-1 also produced the highest pod weight plant^{-1}, respectively, 27.19 g and 27.00 g (Table 4), both were produced at Watu Baur. Meanwhile, Local Manggarai and Betet performed the poorest in this trait with only about, respectively, 9.8 g and 9.5 g plant^{-1} when grown at Perak. Similar to other traits, mean pod weight of all genotypes at Watu Baur was higher than that at Perak, which implies more favorable growing environment conditions at the first location to support optimal pod weight production as compared to the latter. Over two locations, Murai and Vima-1 performed the best with, respectively, 21.9 and 21.0 g plant^{-1} while Local Manggarai performed the poorest with only 11.3 g plant^{-1} (Table 3). These findings were supported by Welsh and Mogea (1991) who stated that each crop genotype has a physiologically unique production capacity that is determined by energy, climate, soil nutrients, humidity, and other growth factor variabilities, which then through their interaction with the genetic capacity of the crop will develop the adaptation capability of the crop in increasing its growth and yield performances.

We also found in the present study that seed weight plant^{-1} also varied greatly among mungbean genotypes as presented in Table 4. Seed weight of the mungbean genotypes ranged from 5.95-23.68 g plant^{-1} with Murai and Vima-1 consistently performed the best when grown at Watu Baur. This implies that these two Indonesian released varieties are the best performers in terms of seed production, which is also supported by their best performances in yield supporting component traits. Mean seed yield plant^{-1} of all genotypes at Watu Baur was two folds that of Perak, which implies better environmental conditions at Watu Baur to support the crop growth and seed yield production. Meanwhile, the mean seed yields of the genotypes over two locations also varied considerably with a range of 8.04 to 18.27 g plant^{-1}. Again, Murai and Vima-1 performed the best over two locations while Local Manggarai performed the poorest. It is interesting to note in this study that despite its long adaptation to the environment in the region, Local Manggarai was still left behind by most of the Indonesian released varieties in terms of seed yield production. This presumably caused mainly by the genetic background of the tested genotypes. Most of the superior varieties had been developed in such a way that all necessary genes are combined to support high yield production and the local cultivar, on the other hand, may only be able to undergo natural processes to develop its production capacity, which is very much unlikely to occur maximally in a short time.

100-seed weight and seed weight plot^{-1}

We observed no significant effect of genotype by location interaction on 100-seed weight; however, each of the single factors Genotype and Location highly significantly affected the trait (Table 1). This implies that 100-seed weights of genotypes included in the present study are mostly determined by the genetic factor, and expression of this genetic factor is not influenced by the change in the growing environmental conditions. Previous studies (Ullah et al. 2011; Raturi et al. 2012; Alia et al. 2014) also found a highly significant effect of either genotype or location on the trait but the effect of genotype by location interaction was not significant on the 100-seed weight of mungbean. Meanwhile, Somta et al. (2015) found six quantitative trait loci (QTLs) that were related to 100-seed weight in mungbean genotypes grown in rainy and dry seasons, however, the number of QTLs detected was substantially affected by day length, which implies that seed weight is also determined by the environmental factor.

Data presented in Table 5 shows quite a wide range of 100-seed weight from 3.13 to 7.97 g. The superior variety Kutilang, in contrast to its poor performances in other observed traits, performed the best with a mean 100-seed weight of 7.81 g over two locations, which differed significantly from that of other genotypes. Local Manggarai performed the poorest with only 3.35 g 100-seed^{-1}. The high 100-seed weight of Kutilang implies that this genotype possesses larger seed size than other genotypes. 100-seed weight is positively correlated with seed size, and seed size itself is also determined by pod size. Small pod size produces small seed size due to the narrow pod wall size that limits the seed cell development.

Like other traits, the mean 100-seed weight of all genotypes at Watu Baur was higher than that at Perak. More favorable environmental conditions at Watu Baur is the plausible explanation for this finding. Favorable environmental conditions at Watu Baur enabled the crop to undergo optimal photosynthesis process. Photosynthesis process is strongly influenced by environmental factors such as sunlight intensity, soil nutrient availability, CO_2, humidity, etc. If these environmental elements are optimally utilized by the plant, then the photosynthesis process will be more optimal in accumulating dry matter/assimilate production (Rao et al. 2016).

Table 4. Mean pod weight plant^{-1} (g) and seed weight plant^{-1} (g) of eight mungbean genotypes tested at two locations

Genotype (G)	Pod weight plant^{-1} (g) Location (E)			Seed weigth plant^{-1} (g) Location (E)		
	Perak	**Watu Baur**	**Mean (G)**	**Perak**	**Watu Baur**	**Mean (G)**
Local Manggarai	9.76 a A	12.86 a A	11.31 a	6.38 a A	9.69 a B	8.04 a
Murai	16.69 c A	27.19 e B	21.94 c	12.85 b A	23.68 e B	18.27 c
Betet	9.52 a A	20.47 bc B	15.00 ab	5.95 a A	16.65 b B	11.30 ab
Vima-1	15.02 c A	27.00 e B	21.01 c	12.47 b A	22.99 e B	17.73 c
Perkutut	14.03 bc A	23.59 cd B	18.81 bc	10.04 b A	19.55 cd B	14.80 bc
Kutilang	14.84 c A	19.12 b B	16.98 bc	10.36 b A	15.58 b B	12.97 b
Walet	10.82 ab A	20.95 bcd B	15.89 ab	7.46 a A	17.07 bc B	12.26 ab
Kenari	14.71 c A	24.22 de B	19.47 bc	10.20 b B	19.94 d B	15.07 bc
Mean (L)	**13.17 A**	**21.93 B**		**9.46 A**	**18.14 B**	

Note: Means within the same coloumn or row with the same letter (s) are not significantly different at 0.05 DMRT. The capital letter indicates comparison within the same row while small letter indicates comparison within the same column

Table 5. Mean 100-seed weight and seed weight plot^{-1} of eight mungbean genotypes tested in two locations

Genotype (G)	100-seed weight (g) Location (L)			Seed weight plot^{-1} (g) Location (L)		
	Perak	**Watu Baur**	**Mean (G)**	**Perak**	**Watu Baur**	**Mean (G)**
Local Manggarai	3.13 a A	3.56 a A	3.35 a	242.73 a A	405.03 a B	323.88 a
Murai	6.82 a A	7.16 a A	6.99 e	493.16 f A	991.87 g B	742.51 c
Betet	5.83 a A	6.23 a A	6.03 b	233.34 a A	695.55 c B	464.45 ab
Vima-1	6.29 a A	6.65 a A	6.47 d	481.85 e A	967.90 g B	724.88 c
Perkutut	6.11 a A	6.55 a A	6.33 c	385.81 c A	817.94 e B	601.87 bc
Kutilang	7.66 a A	7.97 a A	7.81 g	400.78 d A	659.57 b B	530.17 b
Walet	7.14 a A	7.66 a A	7.40 f	287.04 b A	719.44 d B	503.24 ab
Kenari	6.71 a B	7.15 a A	6.93 e	395.53 cd A	832.83 f B	614.18 bc
Mean (L)	**6.21 A**	**6.62 B**		**365.03 A**	**761.27 B**	

Note: Means within the same column or row with the same letter (s) are not significantly different at 0.05 DMRT. The capital letter indicates comparison within the same row while small letter indicates comparison within the same column

In contrast to 100-seed weight, our study results revealed that genotype by location interaction highly significantly affected seed weight per plot and so did the effect of single factor Location. Meanwhile, the single factor Genotype affected the trait significantly (Table 1). The existence of genotype by location interaction effect demonstrates inconsistent genotype performances across the changing environments (Ullah et al. 2011). In this regard, seed weight per plot is controlled by genetic factors and the effect of this genetic factor changed according to the location where the mungbean genotypes are grown. This might have occurred since each genotype responded differently to the changing environment that resulted in variable seed production per plot.

Data in Table 5 show that seed weight plot^{-1} of the tested mungbean genotypes ranged from 233.34 to 991.87 g, and the seed weight plot^{-1} of all genotypes grown at Watu Baur was two folds that at Perak. Similar to seed weight plant^{-1} and other seed yield supporting traits, Murai and Vima-1 produced the highest seed yield plot^{-1} when grown at Watu Baur with, respectively, 991.87 g plot^{-1} and 967.90 g plot^{-1}. Mean seed weight plot^{-1} of Murai and

Vima-1 over the two locations were also the highest with, respectively, 742.51 g and 724.88 g plot^{-1}. These findings indicated that the two Indonesian released superior varieties, i.e. Murai and Vima-1 are the best high-yielding mungbean varieties for the region.

The high seed yield performances of Murai and Vima-1 are supported by their performances in yield supporting traits such as number of productive branches plant^{-1}, number of pod clusters plant^{-1}, number of pods plant^{-1}, pod weight plant^{-1} and seed weight plant^{-1}, where these two varieties consistently performed much better than other genotypes. A previous study by Hakim and Suyamto (2012) reported that seed yield of mungbean is significantly and positively correlated with number of pods plant^{-1}, number of seeds pod^{-1}, seed yield plant^{-1} and 100-seed weight. Significant and positive correlation between seed yield of mungbean with each of yield component characters such as number of seed pod^{-1}, number of pods plant^{-1}, number of pods cluster plant^{-1}, number of pods cluater^{-1}, days to 50% flowering, days to maturity was also reported by Om and Singh (2016).

Seed yield production of Murai was 991.87 g pot^{-1} (equals to 2.48 t ha^{-1}) and that of Vima-1 was 967.90 g plot^{-1} (equals to 2.42 t ha^{-1}). Meanwhile, mean seed yields of Murai and Vima-1 as described in their varietal descriptions are, respectively, 1.5 t ha^{-1} and 1.76 t ha^{-1} (Suhartina 2005). These results indicate that the two varieties had performed very well in the two growing locations in Manggarai District despite their different performances over the two locations.

Mean seed yields per-ha of other five superior varieties were 1.16 t ha^{-1} (Betet), 1.51 t ha^{-1} (Perkutut), 1.33 t ha^{-1} (Kutilang), 1.26 t ha^{-1} (Walet), and 1.54 t ha^{-1} (Kenari) while the mean seed yields of these varieties according to their varietal description are, respectively, 1.5 t ha^{-1} (Betet), 1.5 t ha^{-1} (Perkutut), 1.13 t ha^{-1} (Kutilang), 1.7 t ha^{-1} (Walet), and 1.38 t ha^{-1} (Kenari) (Suhartina 2005). These results show that mean seed yield performance of Perkutut in this trial was similar to that of its varietal description while that of Kutilang and Kenari were higher than their respective varietal descriptions. Betet and Walet, on the other hand, performed poorly with much lower seed yield as compared to that in their varietal descriptions. Meanwhile, mean seed yield of Local Manggarai over two locations was only about 0.81 t ha^{-1}, which was much lower than the superior varieties. The above descriptions demonstrate that each genotype behaved differently in their yield performances depending on their genetic backgrounds and the environments where these genetic factors are expressed. Welsh and Mogea (1991) stated that each crop genotype possesses a physiologically unique production capacity that is determined by genetic factors, which through its interaction with environmental factors such as energy (sunlight intensity), climate, soil nutrients, humidity, and other growth factors will develop the crops growth and yield performances. The effect of the environmental elements such as sunlight intensity, especially the UV-B radiation, on mungbean genotypes has also been examined by Choudhary and Agrawal (2014), who observed diverse morphological and physiological changes including the hormonal regulations in the mungbean genotypes.

By location, the present study results revealed that mean seed yield plot^{-1} of all genotypes was 1.9 t ha^{-1} at Watu Baur but only 0.91 t ha^{-1} at Perak. If one has to choose which location to extend the planting area in the region for the purpose of increasing the mungbean production, then the low altitude locations such as Watu Baur, etc. are the more favorable conditions to select for although the medium altitude regions can still be used as mungbean growing centers by utilizing superior varieties such as Murai and Vima-1, which, in the present study, produced a reasonable seed yield (over 1 t ha^{-1}) in this location. Murai and Vima-1 are, therefore, recommended for extensive cultivation in Manggarai District at either Perak or Watu Baur or other locations to increase the mungbean productivity in the region.

A number of conclusions are drawn from the present study results as follows: (i) Most of the variables we observed in the present study were influenced by interaction between genotype and environment, except plant heights at flowering and harvesting stages, productive branches plant^{-1}, and 100-seed weight. (ii) The shortest harvesting date genotype at both locations was Vima-1 (~59 DAP) while the best performers in number of productive branches plant^{-1}, number of pods plant^{-1}, seed yield plant^{-1} and seed yield plot^{-1}in both locations were Murai and Vima-1. (iii) In addition to Betet and Kenari, Murai and Vima-1 also performed the best in number of pod clusters plant^{-1} in both locations. (iv) The highest number of pod plant^{-1} was produced by Vima-1, Murai, Betet, Walet, and Perkutut while the highest pod weight plant^{-1} was obtained from Murai, Vima-1, and Kenari at Watu Baur. Meanwhile, Kutilang produced the highest 100-seed weight in both Perak and Watu Baur. (v) The highest mean seed yield over two locations was produced by Murai (991.87 g plot^{-1}, equals to 2.48 t ha^{-1} and Vima-1 (967.90 g plot^{-1}, equals to 2.42 t ha^{-1}). (vi) Mean seed yield of all genotypes at Perak was 365.0 g plot^{-1} (equals to 0.91 t ha^{-1}) while that in Watu Baur was 761.27 g plot^{-1} (equals to 1.90 t ha^{-1}).

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The effect of plant biomass and interval of water application on water use efficiency and yield of sweet corn (*Zea mays* L. var. *saccharata* Sturt)

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Abstract. *Soetedjo INP. 2017. The effect of plant biomass and interval of water applications on water use efficiency and yield of sweet corn (*Zea mays *L. var.* saccharata *Sturt). Trop Drylands 1: 32-35.* The main problem in dryland farming system is the loss of water during crop growth and development. The use of organic matters such as paddy straw, paddy husk, and biomass of *Gliricidia* and *Chromolaena* has been proved to minimize water loss. Moreover, water use efficiency can be improved by applying water based on the crop growth stage. The main objective of the research was to determine the interaction effect of different buried crop biomass and interval of water application on water use efficiency and yield of sweet corn (*Zea mays* L. var. *saccharata* Sturt). The research was assigned in factorial treatment laid out in Randomized Completely Block Design with 3 replicates. The research results showed no interaction effect of the treatment combinations on soil water content, water use efficiency, and yield of sweet corn. Each of the single factors of the kind of biomass and water application interval, however, significantly affected water use efficiency and yield of sweet corn. Buried paddy straw produced the highest yield and water use efficiency of sweet corn (750.66 g 3m$^{-2)}$, 14.21%). Moreover, three days of water application interval resulted in higher yield and water use efficiency of sweet corn (654.5 g 3m$^{2(-1)}$, 14.68%).

Keywords: Crop biomass, water applications, water content, water use efficiency, sweet corn

INTRODUCTION

Sweet corn (*Zea mays* L. var. *saccharata* Sturt) is one of the major food crops currently widely grown in East Nusa Tenggara Province, Indonesia. This crop has a high carbohydrate content, low protein and fat, and sweet taste. However, the productivity in sweet corn cultivation varies with the yield depending strongly on soil nutrients and water availability. Meanwhile, the availability of soil nutrients and water becomes the main constraint in East Nusa Tenggara. These constraints could be overcome by practicing farming system that might be able to minimize water loss and improve the availability of nutrients.

The application of organic matters, such as paddy straw, paddy husk, *Gliricidia*, and *Chromolaena* biomass is a promising way to improve the availability of soil nutrients and water. This is mainly because these sources of organic matter contain lignin, silica, cellulose, and organic-C, which might be able to improve soil bulk density, porosity, and water holding capacity (Arsyad 2010). Some studies showed that the use of organic matters, such as stems of corn, sorghum, and paddy husk has the capability to improve soil aeration, and soil water holding capacity. Some organic matters, such as waste of *Gliricidia* (*Gliricidia zepium*) and kirinyu *(Chromolaena odorata*), could increase the availability of Phosphorus, and organic-C content. Moreover, it was shown that organic-C content could improve soil water holding capacity (Tisdale et al. 1985; Praktino et al. 2002; Ajidirman, 2010).

All organic matters can be applied by either spreading them on the soil surface or burying them under the soil. However, burying organic matter under the soil might be more efficient as it is able to reduce evapotranspiration and increase decomposition processes (Puspareny et al. 2008). A previous study by Makarim et al. (2007) showed that burying paddy straw was able to improve the efficiency of soil water holding capacity, resulting in an increase of yield of maize by 22%. Moreover, the efficiency of water holding capacity and water use by crops might be improved by arranging the time of water application. Some studies showed that the use of water by crop depends strongly on the crop growth stages and physical characteristics of the soil. Therefore, optimal water application might significantly affect the nutrient absorption, which then affects the growth and yield of the crop, if water is applied based on the crop water requirement.

Based on all those mentioned study results, this research was conducted to know the interaction effect of different kinds of buried plant biomass (i.e., paddy straw, paddy husk, *Gliricidia*, and *Chromolaena*) and interval of water application (daily, every 3 days, every 5 days) on soil water content, yield of sweet corn and water use efficiency.

MATERIALS AND METHODS

The present research was conducted in East Penfui village, Central Kupang Sub-District, Kupang District from August to November 2014. The research used Factorial research design which was laid out in Randomized Completely Block Design with three replications. Two treatment factors were assigned, namely kinds of buried biomass (i.e. paddy straw, paddy husk, *Gliricidia*, and *Chromolaena*) and interval of water application (daily, every 3 days, every 5 days).

Variables observed in the present study included the nutrient content of the biomass, water use, water use efficiency, and yield of sweet corn. Nutrient contents of the biomass were analyzed 2 weeks before their application into the soil. The observed nutrient content of buried biomass included organic-C content, total N content, and the available P content. Organic-C content was measured by using ashing method (Walkley 1947), total N (%) content was measured by using semi-automatic Kjedhal digestion (AOAC 1975), and the availability of P (ppm) was measured by using Olsen method (Olsen et al. 1954). Water use was measured based on soil water content (i.e. early growth stage, maximum vegetative growth stage, and harvesting stage) by using gravimetric method (SSIR 1984). All collected data were subjected to analyses of variance, followed by a post hoc test of Duncan Multiple Range test (DRMT) at 5% level (Steel and Torie 1989) to compare the treatment means.

RESULTS AND DISCUSSION

Organic-C content of buried biomass

The results of present study showed that *Gliricidia* biomass contained a higher organic-C (41.0%) compared to others buried biomass. Meanwhile, paddy straw had the highest content of C/N ratio (61.0%) (Table 1). The ratio of C/N significantly determined the time of decomposition process in which high C/N ratio will cause a longer decomposition process. This is mainly due to increasing content of lignin and cellulose as result of high C/N ratio. Moreover, it might improve water holding capacity.

Water use

Results of the present study revealed that there was no significant interaction effect between the kinds of buried biomass and the interval of water application on total water use during crop growth stage. However, water use was significantly affected by the single treatment factors of kinds of buried biomass and interval of water application (Table 2).

Results of DRMT at 5% level of data of the single factor on the kinds of buried biomass showed that paddy straw was capable of decreasing the water used by sweet corn during growth stage as compared to other kinds of biomass. This is mainly due to paddy straw having a higher content of C/N ratio, resulting in a better capability to hold the available soil water content. In addition, this biomass is also able to minimize water loss through soil evaporation. Our finding is in line with some studies showing that paddy straw was able to improve soil water holding capacity (Makarim et al. 2007; Murdolelona et al. 2008). Meanwhile, buried *Gliricidia* biomass increased water use significantly during the crop growth stage. This is mainly because buried *Gliricidia* biomass has a lower C/N ratio content compared to that of other kinds of biomass. This low C/N ratio caused a lower capability in improving soil water holding capacity.

Table 1. Nutrient content of paddy straw, paddy husk, *Chromolaena*, and *Gliricidia* biomass

Kinds of biomass	N	P	C-Organic	C/N
Paddy straw	0.64	0.15	39.3	61.00
Paddy husk	0.94	2.40	33.6	35.74
Chromolaena	2.65	0.53	38.6	14.56
Gliricidia	3.13	0.22	41.0	13.00

Table 2. The effect of various buried biomass and interval of water application on water use of sweet corn during the growth stage

Treatment	Water use (mm^3)	
Kinds of biomass		
Paddy straw	630.53	a
Paddy husk	630.58	b
Chromolaena	630.60	b
Gliricidia	630.62	c
Water application interval		
Daily	1092.62	c
3 days	445.76	b
5 days	353.37	a

Note: Values in the same treatment followed by different letter at the same column means significantly different at Duncan Multiple Range Test 5% level

Analysis of variance of data on the interval of water application showed that daily water application resulted in significantly higher water use during crop growth stage as compared to other water application intervals. This is mainly due to daily water application being able to increase soil water content and crop water availability. Meanwhile, soil water content tended to be low when the watering was applied in every 3 and 5 days intervals. As a result, the crop might have used water less than it should be to support the crop growth and yield.

Yield of sweet corn

Results of the present research showed no significant interaction effect between kinds of buried biomass and water application interval on yield of sweet corn. However, the single treatment factor of either kind of biomass or water application interval significantly affected sweet corn yield (Table 3).

Comparison of the treatment means using DMRT post hoc test at 5% level revealed that the highest yield of sweet corn was achieved by the application of buried paddy straw, which was then followed by paddy husk, *Chromolaena odorata*, and *Gliricidia* (Table 3). The high sweet corn yield produced by buried paddy straw was mainly due to the fact that this kind of biomass is able to improve physical characteristics of the soil, resulting in the availability of water content. This condition might have improved the number of soluble nutrients and nutrients exchange in supporting the growth and yield of sweet corn. In contrast, the biomass of *Gliricidia* that was buried into the soil was more easily decomposed, which might have resulted in a lower soil water holding capacity, a lower number of soluble nutrients and nutrients exchange. Previous studies (e.g. Agustina 2007; Adirman 2010; Arsyad 2010) demonstrated that buried biomass with a high content of C organic was able to improve soil physical characteristics, increase water holding capacity 2-3 times their weight, and increase soil water content up to 43%.

Our data on the single factor treatment of water application interval also showed that daily water application significantly resulted in a higher yield of sweet corn compared to other treatments (Table 3). This result is related to better soil water availability, better soluble nutrient content, and better nutrient absorption of this treatment to support growth and yield of sweet corn.

Water use efficiency

The present study results revealed no interaction effect of kinds of buried biomass and interval of water application on water use efficiency. However, each of the two single treatment factors caused a significant effect on water use efficiency of sweet corn.

Analysis of data of the single treatment factor on the kinds of buried biomass showed that paddy straw and paddy husk provided a higher water use efficiency as compared to buried *Gliricidia* biomass (Table 4). This is mainly due to the high C/N ratio of paddy straw and paddy husk biomass, resulting in longer decomposing processes of the biomass by the microorganism. The biomass of paddy straw and paddy husk could increase water holding capacity of the biomass and the soil. Some studies showed that yield of the crop could be improved by 1.5-2 higher than their yield potential as a result of the increase of soil water holding capacity (Adirman 2010; Arsyad 2010).

Furthermore, the 3 days and 5 days intervals of water application were more efficient than daily water application (Table 4). This is mainly because soil water availability of either 3 days or 5 days water application intervals was lower than that of daily water application, however, crop yield of sweet corn irrigated every day was higher than that of 3 and 5 days irrigation intervals. This implies that better water use efficiency is not always correlated with a better yield of sweet corn. Previous studies showed that improvement of crop water use efficiency depends strongly on the capability of the crop in using water and soluble nutrients in supporting their growth and yield (Islami and Utomo, 1995 and Supriadi 2008).

Based on the present study results, several conclusions were made as follows: (i) There was no interaction effect between the kinds of buried plant biomass and the interval of water application on water use, the yield of sweet corn, and water use efficiency, but each of the single treatment factors significantly affected the observed variables. (ii) Within the single treatment factor of kinds of biomass, buried paddy straw provided the lowest water use, the highest sweet corn yield, and the most efficient water use. (iii) Within the single factor treatment of water application interval, 3 days water application interval provided the lowest water use, the highest yield of sweet corn, and the most efficient water use.

Table 3. The effect of various buried biomass and interval of water application on sweet corn yield (g $3m^{-2}$).

Treatment	Sweet corn yield (g $3m^{-2}$)	
Kinds of biomass		
Paddy straw	750.77	b
Paddy husk	683.33	b
Chromolaena	630.00	ab
Gliricidia	561.00	a
Water application interval		
Daily	752.25	c
3 days	654.50	b
5 days	562.00	a

Note: Values in the same treatment followed by different letters at the same column means significantly different at Duncan Multiple Range Test 5% level.

Table 4. The effect of various buried biomass and interval of water application on water use efficiency.

Treatment	Water use efficiency	
Kinds of biomass		
Paddy straw	14.22	b
Paddy husk	13.01	b
Chromolaena	11.89	ab
Gliricidia	10.81	a
Water Application Interval		
Daily	6.88	a
3 days	14.68	b
5 days	15.90	b

Note: Values in the same treatment followed by different letter at the same column means significantly different at Duncan Multiple Range Test 5% level.

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Species distribution model of invasive alien species *Acacia nilotica* for Central-Eastern Indonesia using Biodiversity Climate Change Virtual Laboratory (BCCVL)

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Abstract. *Sutomo, Van Etten E. 2017. Species distribution model of invasive alien species Acacia nilotica for Central-Eastern Indonesia using Biodiversity Climate Change Virtual Laboratory (BCCVL). Trop Drylands 1: 36-42.* Climate change may facilitate alien species invasion into new areas. This study uses Biodiversity and Climate Change Virtual Laboratory to develop a species distribution model (SDM) of *Acacia nilotica* (L.) Willd. ex Delile. based upon its naturalized distribution to project the potential distribution of *A. nilotica* throughout tropical environment of Indonesia under current and future climate conditions. Global Biodiversity Information Facility (GBIF) database was utilized to obtain the species occurrences data. The climate factors were precipitation and temperature layers, available in WorldClim current conditions (1950-2000) at 2.5 arcmin. We used Generalized Linear Model. The result was then projected to the year 2045 using RCP 8.5 greenhouse gas emissions scenarios to influence the climate model (CSIRO Mark 3.0. with 30'' resolution). Final results show that global climate change is likely to increase markedly the potential distribution of *A. nilotica* in Indonesia. By the year 2045, *A. nilotica* is most likely to spread to eastern parts of Indonesia. In general, our model performance is good (AUC = 0.82), however, like many other SDMs, it does not take into account biotic interactions as well as other environmental factors. Nonetheless, climatic suitability is an essential requirement for successful establishment of an invasive species and species distribution models that can disclose general patterns and convey useful estimates.

Keywords: *Acacia nilotica*, invasive alien species, species distribution model, climate change

Abbreviations: GCM: Global Climate Change Model, SDM: Species Distribution Model, BCCVL: Bidoversity and Climate Change Virtual Laboratory, GBIF: Global Biodiversity Information Facility, GLM: Generalized Linear Model, AUC: Area Under the Curve, ROC: Receiver-Operating Characteristics, TPR: True Positive Rate, TNR: True Negative Rate, HSI: Habitat Suitability Index, IAS: Invasive Alien Species, NTT: Nusa Tenggara Timur

INTRODUCTION

Acacia nilotica (L.) Willd. ex Delile is a small tree species widespread in the northern savanna regions of Africa, and its range extends from Mali to Sudan and Egypt. *A. nilotica* is known to be abundant in its native habitat in Africa (Brenan 1983), but no study has been specifically focused on this species. Outside of its original distribution, *A. nilotica* is considered an invasive alien species or noxious weed. In Australia, this species is found to spread along the western part of Queensland while in other states such as Western Australia, New South Wales, Adelaide and Northern Territory, this species was found in only a few numbers (Reynolds and Carter 1990). *A. nilotica* is considered to have a negative impact on savannas since adult trees of this species are tolerant to fire (Radford et al. 2001). A previous study by Burrows et al. (1991) observed that on Mitchell grasslands in Australia, *A. nilotica* was invading the grassland and forming thorny thicket formations.

A. nilotica was first introduced in Indonesia in the late 1960s in Baluran National Park in East Java. The original purpose of introducing this species was to create fire breaks to prevent the fire from spreading from Baluran Savanna into the surrounding forests. Along the time, the rapid spread of *A. nilotica* has threatened the existence of Baluran Savanna which has caused changes in some areas of the national park from open savannas to the more closed canopy areas (Djufri, 2004). If unabated, the continuing spread and over dominance of *A. nilotica* could shift the savanna into another ecosystem state such as the secondary/dry forest.

It was predicted by Global Climate change Models (GCMs) (IPCC 2007) that by the end of 21st century, global warming would cause an increase of mean temperature of about 3-4 °C, a decrease of rainfall of about 30-40%, and significant changes in seasonal as well as severe weather events. Molloy et al. (2013) stated that, at a regional scale, most species and ecological communities exist within a definable bioclimatic niche, where precipitation and temperature are among a set of variable climatic parameters that control the habitat values. If these variables change, then the habitat value for that area will also change. The task of understanding how species and communities respond to changes is crucial. Climate change may facilitate alien species invasion into new areas, particularly for species from warm native ranges introduced into areas currently marginal for temperature (Sheppard et al. 2014).

Species distribution model or SDM has the capability to assess current distribution and simulate climate-induced range shifts under different global change scenarios at the single-species and community levels (Crego et al. 2014). Therefore, the model could identify areas at risk of further invasion by invasive alien species so that pre-emptive actions could be undertaken in a plausible approach. Franklin et al. (2014) described steps in species distribution modelling as follows. Species occurrence data (such as presence-only, presence-absence or abundance) are the response variable, and environmental variables are the predictors used in a multiple regression-like modelling framework. The model can be fitted in data space using a wide variety of statistical learning methods. Estimated parameters are then applied back to environmental data layers (mapped grids) to predict the probability of species occurrences in geographical space.

SDMs has enabled conservationists to predict future landscape and has been applied in many areas of research such as for invasive species management (Webber et al. 2011), conservation and reintroduction of endangered species (Adhikari et al. 2012; Molloy et al. 2016), adaptive management of protected areas (Mairota et al. 2014), restoring landscapes connectivity (Gurrutxaga and Saura 2014) and many others. Molloy et al. (2013) did SDMs using bioclimatic variables to determine the impacts of a changing climate on the western ringtail possum (*Pseudocheirus occidentals*; Pseudocheiridae) in South-west Australia. Kritikos et al. (2003) conducted SDM using CLIMEX and predicted the potential distribution under future climate change scenarios for invasive alien species *A. nilotica* subspecies *indica* in Australia. Their results showed that the potential distribution of the species in Australia under current climatic conditions is vast and far greater than the current distribution. The authors also highlighted the impact of global climate change that may likely increase the species potential distribution in Australia and significantly increase the area at risk of invasion.

This study used Biodiversity and Climate Change Virtual Laboratory, BCCVL (http://www.bccvl.org.au/) (Hallgren et al. 2016) to develop a species distribution model of *A. nilotica* based upon its naturalized distribution in and outside of Indonesia to project the potential distribution of *A. nilotica* throughout Indonesia under current climate conditions and to assess the sensitivity of this distribution to climate change. BCCVL is a unique cloud-based virtual laboratory that provides access to numerous species distribution modelling tools; a large and growing collection of biological, climate, and other environmental datasets; and a variety of experiment types to conduct research into the impact of climate change on biodiversity (Hallgren et al. 2016). Understanding the likely potential distribution of this obnoxious plant under current and future climate scenarios will help policy makers and land managers to set up proper approaches to handling the invasion.

MATERIALS AND METHODS

Procedures

We made use of the Biodiversity and Climate Change Virtual Laboratory, BCCVL (http://www.bccvl.org.au/) to conduct the modelling analysis (Hallgren et al. 2016). Global Biodiversity Information Facility, GBIF (http://www.gbif.org/) was utilized to obtain the species occurrence data for *A. nilotica* (GBIF 2016). Global Biodiversity Information Facility (GBIF) is an international open data infrastructure funded by governments. Currently, there are various subspecies of *A. nilotica* listed in the GBIF. We chose to use the common *A. nilotica* (L.) Willd. ex Delile. as it is recorded to be present at Baluran National Park in East Java, Indonesia (Tjitrosoedirdjo 2008). The GBIF database has about 879 occurrence records of *A. nilotica* with 232 occurrences were geo-referenced. This dataset was then imported into Biodiversity and Climate Change Virtual Laboratory (BCCVL). This data of species occurrence acted as the response variable, and the predictors were the environmental variables.

In this simulation, we used WorldClim current conditions (1950-2000) at 2.5 arcmins as environmental predictors. The WorldClim collections consist of an array of global climate layers (climate grids) covering all global land areas except Antarctica. They are in the latitude /longitude coordinate reference system (not projected) and the datum is WGS84. The average monthly climate data from weather stations were used to produce the data layers. The WorldClim collection is composed of two main components, i.e. the current climate and the future climate (Hijmans et al. 2005). We selected precipitation and temperature variables because they represent important factors determining vegetation range and abundance (Krebs 1985; Van Steenis 1972), and that *A. nilotica* SDM in Australia correspond well with these climate variables (Kriticos et al. 2003). We selected seven climate variables namely maximum temperature of warmest month (B05), minimum temperature of coldest month (B06), mean temperature of wettest quarter (B08), mean temperature of driest quarter (B09), precipitation of wettest month (B13) and driest month (B14), precipitation seasonality (B15) and temperature seasonality (B04). Extreme variables are better represented by the ranges of conditions where species can occur (Crego et al. 2014).

Data analysis

The Species Distribution Model Experiment (SDM) allows us to investigate the potential distribution of a species under current climatic conditions. The BCCVL currently provides 17 different algorithms across four different categories to run species distribution model (e.g. profile model, statistical regression model, machine learning model and geographic model). In this study, we used statistical regression model namely Generalized Linear Model (GLM) to process our SDM. The GLM was used for the reason that the model accommodates regression model for data with a non-normal distribution, fitted with maximum likelihood estimation. This model produces estimates of the effect of different environmental variables on the distribution of a species. The model uses all the data available to estimate the parameters of the environmental variables and construct a function that best describes the effect of these predictors on species occurrence. The suitability of a particular model is often defined by specific model assumptions. The prediction is visualized as the suitability of a grid cell on a scale from 0 to 1, where 0 refers to very low suitability and 1 refers to very high suitability. Results from BCCVL are .tiff file which then processed further using ARC MAP 10.1. Raster value was created and classified, and then different color was given to easily enable us to different areas with different habitat suitability index.

The primary output of an SDM was a map that showed the predicted distribution of *A. nilotica* under the baseline conditions. The prediction is not really refer to where the species occurs, but rather the distribution of suitable habitat as defined by the environmental variables (in this case current climate condition) included in the model. The second output was response curve. The response curve that shows the relationship between the probability of occurrence for a species and each of the environmental variables was modelled according to the method of Richmond and Huijbers (2016). Model robustness was evaluated using the AUC (Area Under the Curve) of the ROC curve (Receiver-Operating Characteristics), which is a nonparametric threshold-independent measure of accuracy commonly used to evaluate species distribution model (Bertelsmeier and Courchamp 2014). The ROC plot is a graph of the False Positive Rate (1-Specificity) on the x-axis and the True Positive Rate (Sensitivity) on the y-axis plotted across the range of threshold probability values. The value for ROC is the area under the curve (AUC). A value of 0.5 represents a random prediction, and thus values above 0.5 indicate predictions better than random. We interpreted the AUC score as follow: a value above 0.9 is excellent, good 0.9 > AUC > 0.8, fair 0.8 > AUC > 0.7, poor 0.7 > AUC > 0.6 and fail 0.6 > AUC > 0.5 (Crego et al. 2014; Sweets 1988).

Further analysis was conducted to investigate the distribution of *A. nilotica* under potential future climatic conditions. In BCCVL this is named the Climate Change Experiment. Climate change experiment obtains a prediction of where *A. nilotica* could occur in the future under a particular climate change scenario. This analysis uses the results from the SDM experiment and projects that distribution for a certain year in the future with the climate information from one of several climate models. In this study, we selected RCP 8.5 (business as usual) greenhouse gas emissions scenarios to influence the climate model; in this case, we used CSIRO Mark 3.0 with 30" (~1km) resolution. We projected our SDM to the year 2045.

RESULTS AND DISCUSSION

Results

The potential distribution of *A. nilotica* in Indonesia under current climatic conditions is greater than the current distribution. Under the current climate, it is predicted that *A. nilotica* is most likely to spread to eastern parts of Indonesia although some parts of Java especially in the south coast of East and Central Java, also are predicted to be suitable for *A. nilotica* (Figure 1). To the eastern Indonesia, *A. nilotica* is predicted to be suitable to inhabit several sites in south-eastern Bali, southwest of Lombok, south-eastern Sumba, most of Kupang District on West side of Timor Island, and also some part of along south coast of West Papua. In West Papua, the species is predicted to be potentially suitable to inhabit (±0.64) southern coast along Merauke. Also interesting to note that small island just off the coast between Merauke and Tual is predicted by the model to be potentially suitable (±0.61) habitat for this invasive species (Figure 1). There is a trend of increasing value of mean suitability index (0.47 to 0.6) as we moved from central to eastern part of the archipelago (Figure 2).

Global climate change is likely to increase the potential distribution of *A. nilotica* in Indonesia, increasing the area at risk of invasion. Bali Island, Lombok Island (West Nusa Tenggara NTB), Sumba Island and Kupang (East Nusa Tenggara), as well as West Papua, have increased the area at risk of invasion. On Sumba Island (Figure 3), the current prediction has one area (*Baing* District in the south-eastern Sumba) where the HSI (Habitat Suitability Index) is in the range of 0.7 - 0.9, and by 2045 it is predicted that *Melolo* Sub-district in north-eastern Sumba will also have similar HSI to *Baing* District. For Kupang on the west side of Timor Island, it is detected that there will be an increase in the HSI around the Tenau and Oesau Sub-districts by 2045 (Figure 3). In general, our model performance is of good results as AUC values (0.82) are still in the range of 0.8 - 0.9 (Figure 5).

All of the chosen climate variables are responsive to *A. nilotica* distribution (Figure 4). The response curves in this plot show that the probability of occurrence of *A. nilotica* follows an optimum curve for the specific variable. *A. nilotica* occurs in areas that are very seasonal. It can grow in places with low rainfall and high rainfall (10 - 150 mm month^{-1}). It does not like extreme cold or frost, it can grow in areas where the minimum temperature of the coldest month is around 12 to 13 °C and it also grows in areas where the maximum temperature of the warmest month is around ~35 °C.

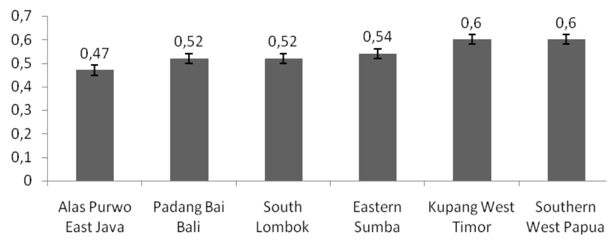


Figure 2. Mean of suitability index value for *Acacia nilotica* model using GLM in BCCVL.

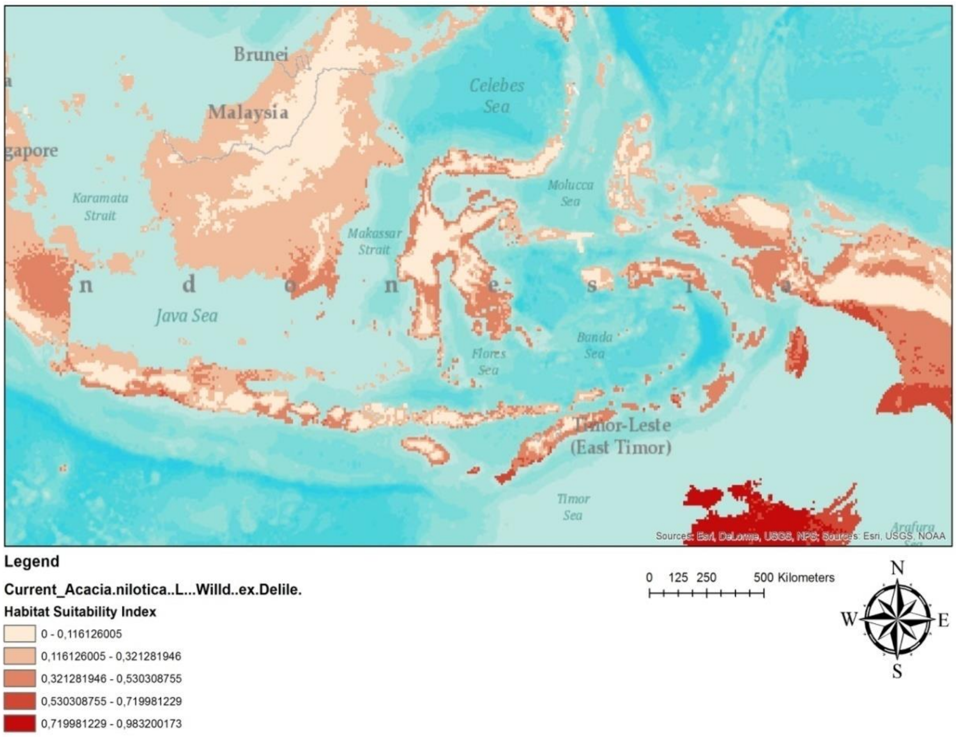
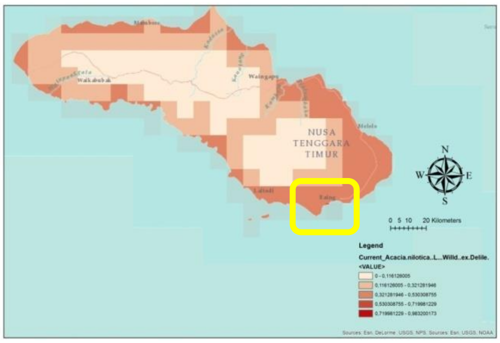
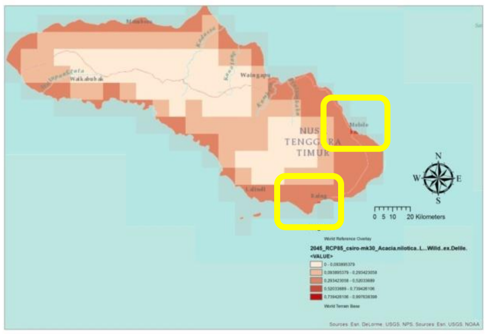


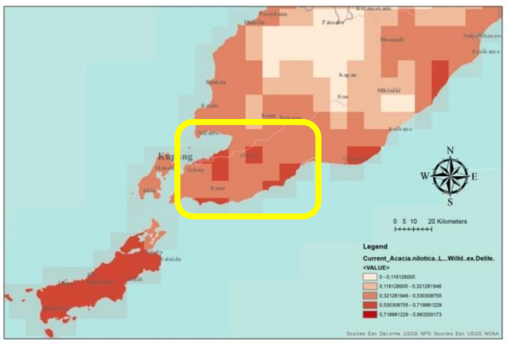
Figure 1. Predicted current distribution (habitat suitability map) of Invasive Alien Species (IAS) *Acacia nilotica* in Indonesia under current climate conditions using Generalized Linear Model (GLM) algorithm in BCCVL. Darker areas represent a higher likelihood that the species can occur.



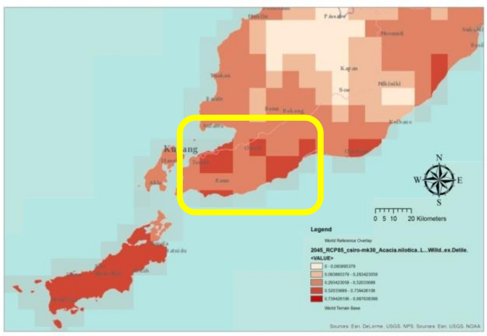
Sumba, Current



Sumba, 2045



Kupang, Current



Kupang, 2045

Figure 3. Projection of *Acacia nilotica* species distribution/Habitat Suitability Index (HSI) on Sumba Island and Kupang, East Nusa Tenggara Province, Indonesia by 2045. Left: Predicted current distribution/current HSI. Right: Projected distribution/projected HSI in 2045. Yellow square to show to emphasize where the HSI changed.

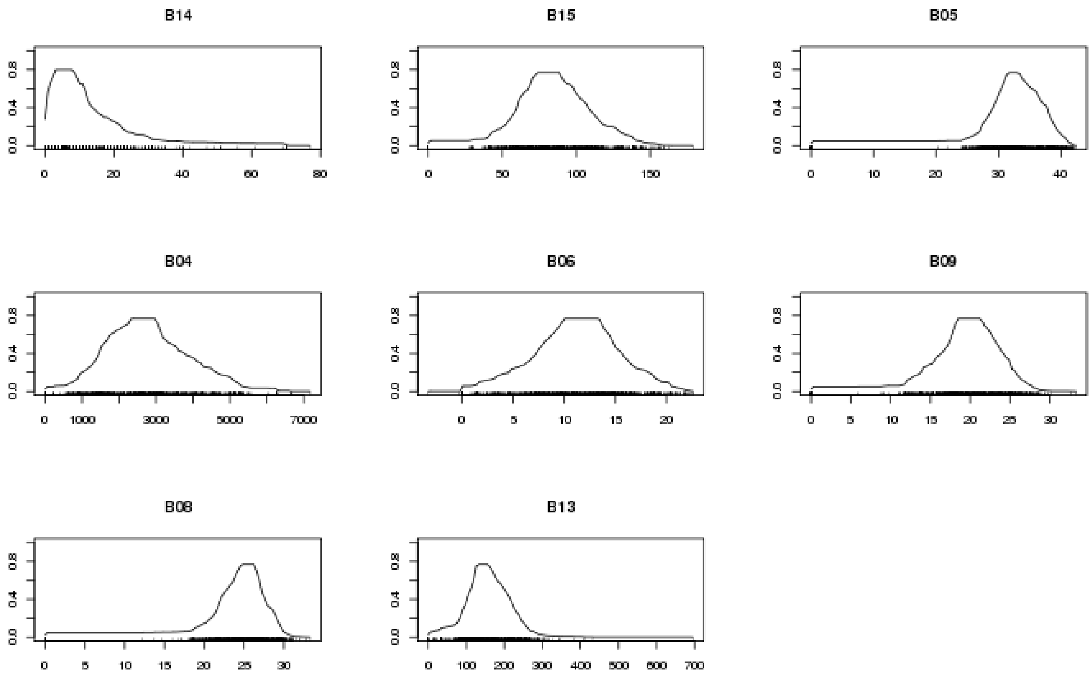


Figure 4. The response curve for *Acacia nilotica* distribution model. B05: maximum temperature of warmest month, B06: minimum temperature of coldest month, B08: mean temperature of wettest quarter, B09: mean temperature of driest quarter, B13: precipitation of wettest month, B14: precipitation of driest month, B15: precipitation seasonality and B04: temperature seasonality

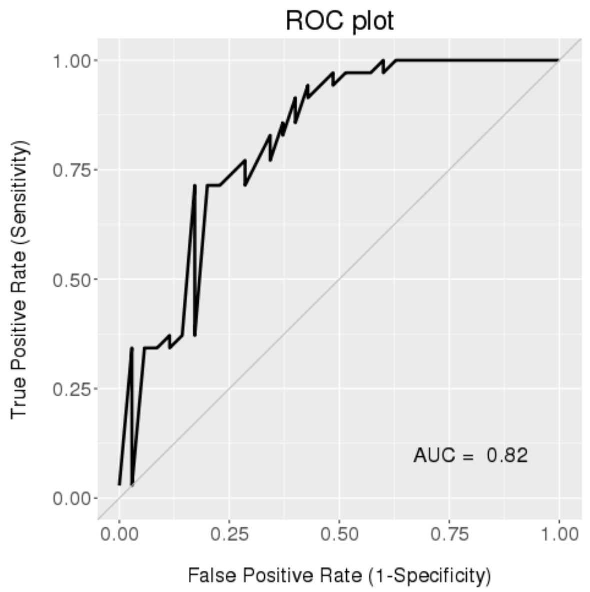


Figure 5. ROC plot which shows AUC value for *Acacia nilotica* model using GLM in BCCVL.

Discussion

The study shows that areas with the highest concentration of potential invasion of *A. nilotica* in Indonesia are mainly located along the coastline, especially in the southeast of Indonesia archipelagoes such as East Java, Bali, Lombok, Sumba, Kupang and West Papua. This result agrees with the current location of *A. nilotica* ever recorded in Indonesia in the literature and is also based on our encounter in the field. Fisher (2010) mentioned the presence of *A. nilotica* in savanna in Kupang East Nusa Tenggara Indonesia in 2010. Additionally, on our field trip to Kupang in 2015, we also noted our encounter with this thorny Acacia in savanna near a roadside to Tablolong Beach. In his expedition in 2010, Arinasa (personal communication) noted the presence of *A. nilotica* in Bali Barat National Park with the herbarium, seed and cone samples were processed and registered at the Hortus Botanicus Baliensis in Bali Botanical Garden. Previous work by Tjitrosoedirdjo (2008) also reported the spread of the species in other islands outside Java such as Timor and Papua. *A. nilotica* was reported as dominant colonizer at Baluran National Park in East Java Province and Wasur National Park in Papua (Tjitrosoedirdjo 2008). However, at a local scale, the model did not recognize Baluran National Park, at the north tip of East Java Province with high HSI and thus underestimate the occurrence of *A. nilotica* in this locality where it has been a problem for more than 20 years.

The current SDM model, when overlaid with other environmental and landscape layers, could have provided more insight. *A. nilotica* is found in savannas and grasslands at various soil types and various temperatures but is reported to be very sensitive to frost but will grow in an area where the mean monthly temperature of the coldest month is 16°C (FAO 2014). This is in line with our findings. Our model shows that *A. nilotica* habitats are very seasonal; it follows an optimum curve for the selected temperature and precipitation with a high probability in areas that have a temperature between 13 and 35 °C. This factor has some connection with land use where this species occurs which is mostly in savanna or grassland as places that have high seasonality as can be seen in Baluran National Park East Java and on Sumba Island (Bond and Keeley 2005; Sutomo and van Etten 2016). *A. nilotica* was not only found in a savanna ecosystem with the volcanic-origin type of soil as in Baluran National Park (Sutomo 2015), but also in other savannas where the soils are not of volcanic origin such as savannas in Sumba and Kupang in NTT Province. Savanna is the dominant type of land cover on Sumba Island and it is also the most burnt (Sutomo and van Etten 2016, unpublished data). Sumba Island receives an average annual precipitation of 900 mm and experience more than five months of the dry period and is located in region A (southern monsoonal region) of Indonesian climate classification region (Aldrian and Susanto 2003; Hanifah 2014). The average high temperature and low rainfall with prolonged dry season as well as local practices caused fires is more prominent in this landscape (Fisher et al. 2006; Monk et al. 2000). East Sumba District was where most of the dominant fire hotspots were detected (Sutomo and van Etten 2016, unpublished data). Our SDM also found that high HSI areas were found in East Sumba and that by 2045 the HSI is more likely to increase.

Our model projection shows a fairly small increase in potential distribution of *A. nilotica* in south-eastern parts of Indonesia by 2045 which is reasonable for a tropical climate. Our model had moderate to good performance (AUC) but yet they share a certain number of fundamental assumptions common to all species distribution models. These models do not take into account biotic interactions or microclimate conditions which may decide at a very local scale whether or not a particularly invasive species eventually becomes established in a given area (Bertelsmeier and Courchamp 2014). Nonetheless, climatic suitability is an essential requirement for successful establishment of an invasive species and species distribution models can disclose general patterns and convey useful estimates (Bertelsmeier and Courchamp 2014).

The SDM model, however, does not take into account biotic interactions and at a local scale; this interaction has been proved to have significantly affected the species of interest. For example, the interaction between *A. nilotica* with herbivores has benefited *A. nilotica*. The lowland savanna area is an important habitat for big mammal grazers such as wild buffalo, deer and banteng. Invasion of *A. nilotica* in these areas may have caused changes in the feeding behavior of these grazers and may have been causing the spread of this invasive species further in the Baluran National Park. *A. nilotica* is also unpalatable by the herbivores as this plant possesses thorny spikes on its branches which make it difficult for the herbivores to consume the leaf. However, the pods that drop to the ground are usually consumed by herbivores during the prolonged dry period when fresh shoots and grasses are scarce. At the end of the wet season toward the dry season, mature *A. nilotica* pods drop from the trees and are consumed by herbivores such as water buffalo (Tjitrosoedirdjo et al. 2013) that spread *A. nilotica* further in the national park. Sutomo et al. (2015) found that there was a large number of seeds found in buffalo dung. In approximately 100 grams of buffalo dung/dropping, there were approximately 166 seeds of *A. nilotica*. In Baluran National Park, Landsat TM image analysis shows that in fourteen years (2000 - 2014), savanna size has decreased by 1,361 ha. Meanwhile, *A. nilotica* stands to increase by 1,886 ha (Sutomo and van Etten 2016, unpublished data).

In addition, other environmental factors such as soil nutrients, land-use changes and disturbance agents such as fire also need to be used in consideration with the model. Moreover, our understandings of the capability of *A. nilotica* to adapt to the changes in these factors is still limited (but see Kriticos et al., 2003). Based on their species distribution modelling and climate change projection for *A. nilotica (*subsp. *indicia)* in Australia, it is expected that there may be increases in water-use efficiency of the species due to increased atmospheric CO^2 concentrations, which allows it to invade more xeric sites further inland and increased temperatures. *A. nilotica* is frost intolerant, grows in areas where the mean monthly temperature of the coldest month is 16° C and it can endure temperatures up to 5°C (Kriticos et al. 2003). Conservationists should not be deterred from using the predictive power of SDMs although modelling limitations apply. A dynamic SDM, based on well-surveyed populations responding to changes in known critical parameters, is one of the most excellent tools existing for conservationists to visually suggest future conditions. Users should be aware of limitations, but the model insights are a vital preliminary point for decision making (Carvalho et al. 2011).

Finally, this study is the first to model and highlight regions with a high risk of invasion by invasive alien species *A. nilotica* in the tropical environment of Southeast Asia, especially central and eastern Indonesia. Future research should endeavor at developing extrapolative modelling based on traits related to invasiveness that can be combined with the current model of estimation of geographic likelihood establishment. More research is needed on how to use SDMs in the service of informing public policy, stakeholders' scenario analysis and applied conservation (Driscol et al. 2012). When SDMs can bring together scientists, public stakeholders, and policy makers, and are used as an adaptive management tool to understand complex landscapes that are undergoing changes, only then it has achieve their full potential (Polunin 2014).

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Acclimatization of plant collection from Moyo Island, West Nusa Tenggara, Indonesia in Purwodadi Botanic Gardens

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Abstract. *Trimanto, Rahadiantoro A. 2017. Acclimatization of plant collection from Moyo Island, West Nusa Tenggara, Indonesia in Purwodadi Botanic Gardens. Trop Drylands 1: 43-49.* There is limited knowledge on the handling of plant specimens for botanical garden collection following field exploration. One key step is called acclimatization with the aim to adapt the newly collected specimens from their natural habitat into a new environment of Botanic Gardens. This research aimed to observe acclimatization process of living plant specimens collected from Moyo Island, West Nusa Tenggara in Purwodadi Botanic Gardens, East Java, Indonesia. We observed the acclimatization of 123 accession numbers of general collection and 17 accession numbers of orchid collection collected from Moyo Island. We investigated successfully adapting plants from the exploration site in Purwodadi Botanic Gardens as well as the non-survived specimens and analyze the mechanism of their adaptation. The study was conducted at a nursery unit in 2013, with an observational method. The results show that 75% of plant living specimens (i.e. 96 accessions of the general plant and nine accessions of orchid) collected from Moyo Island could adapt in Purwodadi Botanic Gardens, while the rest were unable to survive in the garden. Similarity in environmental variables between Moyo Island and Purwodadi Botanic Gardens seemed to influence the comparatively high survivorship of the plant specimens. Nonetheless, the orchid collection was harder to adapt in the garden compared to the general plant collection.

Keywords: Acclimatization, Moyo Island, Purwodadi Botanic Gardens

INTRODUCTION

Botanical exploration is a key activity to support conservation programs particularly in the context of *ex-situ* conservation such as Botanic Gardens, arboretum, etc. This activity is aimed to collect plants from their natural habitat (e.g. forest) with special attention is focused on rare and endemic species. Most of the botanical collections conserved in botanical gardens in Indonesia were originated from exploration activities including plant collection in Purwodadi Botanic Gardens in East Java, Indonesia.

During the last few years, botanists of Indonesian botanical gardens have chosen Nusa Tenggara region as the location for exploration because of its uniqueness due to its location in the Wallacea region. This biogeographical context has led to very diverse flora and fauna (Monk et al. 2007) in Nusa Tenggara. Nonetheless, in Purwodadi Botanic Gardens, there were 44 accessions of general plant (Lestarini et al. 2013) and eight accessions of orchid (Lestarini et al. 2009) from Nusa Tenggara that was conserved until 2013. This number is comparatively low than the high diversity and endemism of flora of the region. Therefore, in 2013, exploration was conducted in Moyo Island, West Nusa Tenggara Province, aiming to add botanical collection in the Purwodadi Botanic Gardens.

Moyo Island is located in Sumbawa Besar, Labuan Padas District, West Nusa Tenggara (8º9'36"-8º23'19" S latitude and 117º27'45"-117º35'42" E longitude). In 1972, Moyo Island was designated as a conservation area with a total extent of 18,765 ha and lowland altitude up to 600 m a.s.l. This area consists of a Mediterranean soil type with sandy soil texture, poor soil nutrient content due to rapid soil permeability and soil leaching. The Moyo Island climate type is similar to that of Botanic Purwodadi Garden which is categorized as dry lowland areas and share similar environmental factors such as rainfall, temperature, soil pH, humidity and light intensity (Trimanto et al. 2013).

After botanical exploration and before plant collection in the Botanic Gardens, there is one key step to ease the adaptation of plant specimens in the garden which is so-called acclimatization. Acclimatization process will determine to what extent the plants can adapt since the native species need a suitable climate to be able to grow in a new environment. A proper method of acclimatization is required to increase the survivorship of the plant when planted in the garden for *ex-situ* collection. Nonetheless, there is limited knowledge on the techniques of acclimatization of plant for collection in botanical gardens. This research aimed to observe acclimatization of living plant specimens collected from Moyo Island at the Purwodadi Botanic Gardens. The results of this research might enrich the understanding of acclimatization process of plant collection in Botanic Gardens, especially in context of the dry tropical lowland ecoregion.

MATERIALS AND METHODS

This research was conducted at the greenhouse in nursery complex of Purwodadi Botanic Gardens from April to December 2013. The plant specimens included 140 accession numbers from Moyo Island, West Nusa Tenggara, Indonesia. An observation method was used to determine the acclimatization process and determine the number of non-adapting plants in Purwodadi Botanic Gardens. Observed temperature, humidity, medium pH, and light intensity were used to determine and compare the environmental factors in the nursery unit.

Acclimatization process was divided into two groups of plants, the general collection (non-orchids) and the orchid collection (Trimanto 2013). General treatment steps of plant materials from exploration (seedlings, plant cuttings and seeds) in the nursery of Purwodadi Botanic Gardens included: (i) Trimming the leaves of plant materials to minimize evaporation and wrapping the roots with moss to maintain moisture content, (ii) Dipping the plant materials in a plant growth regulator (i.e. Rootone-F) with a concentration of 100 mg L^{-1}, (iii) Putting the plant materials in a plastic bag, blowing and covering tightly to keep the plant fresh, (iv) Growing the plant materials on pure sand, (v) Covering the plant materials with plastic until the plants grow well (indicated with the appearance of leaf buds), (vi) Transplanting the plant on polybag media with a mixture of soil, compost, sand and rice husk of 1:1:1:1/2 ratio, (vii) Maintaining the plant in paranet house including watering, adding media and pest control. For orchid collection, the treatment step of plant materials included planting of the orchid collection in the orchid greenhouse. Planting medium for epiphytic orchids was tree fern fiber (or sphagnum moss) and that for the terrestrial orchid was a mixture of compost, soil, and sand medium in a ratio of 1:1:1 (Figure 1).

RESULTS AND DISCUSSION

The present study results showed that the majority of plant specimens collected from Moyo Island were able to adapt well at Purwodadi Botanic Gardens. There were 78 accession numbers of general collection (non-orchid) and 12 accession numbers of orchid that can survive in Purwodadi Botanic Gardens (Table 1), while only 22% of the general collection of plants and 29% of orchid collection were unable to adapt at the garden until December 2013. During April 2013 to February 2014, the vegetative growth was very good in the nursery unit which also applied to the terrestrial orchid collection.

Figure 1. Steps of procedure in handing plant specimens collected from botanical exploration in Purwodadi Botanic Gardens, Pasuruan, East Java, Indonesia. A. Reducing leaves and wrapping the seedlings roots with moss, B. Dipping the seedling materials into a root one solution, C. Covering the plant materials with plastic immediately after they arrived in the nursery, and D. Growing the plant materials in pure sand, E-F. Covering the plant materials with plastic, G. The growth performance of plant materials, H. Transplanting the growing plants into polybags and maintaining them in paranet house, I. Treatment of epiphytic orchid in field

There are three most important environmental factors that influence plant growth, namely temperature, water (precipitation), and light (Liwellin 2001). Documenting the environmental factors in the nursery unit of Purwodadi Botanic Gardens is needed to know the condition of acclimatization. Environmental conditions of acclimatization in the nursery such as temperature, humidity, and pH were favorable for the plants to survive (Table 2). The survival rate of the plant collection from Moyo Island was equal to that of Waru-waru coast, Sempu Island, East Java but was higher than those of Egon forest and Mutis Mountain, East Nusa Tenggara. This similar survival rate is likely influenced by environmental factors of Purwodadi Botanic Gardens that are similar to those of Moyo Island and Sempu Island. The acclimatization of plants originating from East Nusa Tenggara (Egon Forest and Mutis Mountain) with different environmental conditions (altitudes above 800 m asl) indicated lower adaptation rates in which only half the collected plants survived in Purwodadi Botanic Gardens. The result of our study is in line with the previous findings (Trimanto 2013; Permatasari and Rahadiantoro 2015) that the elevation of exploration site affects the adaptability of living collections.

In their native habitat in Moyo Island, the collected plants can grow well in low rainfall and dryland conditions. The altitude of the native habitat is not higher than 600 m a.s.l., which is similar to that of Purwodadi Botanic Gardens. Overall, the climatic condition in Purwodadi Botanic Gardens is similar to that of Moyo Island so that many plants collected in Moyo Island could survive in Purwodadi Botanic Gardens, although several of the collected plants could not survive at Purwodadi Botanic Gardens (Table 3).

Since 2006, the nursery in Purwodadi Botanic Gardens has developed several methods of acclimatization to adapt plant material from various locations in Indonesia. Every plant material needs a different way to survive in Purwodadi Botanic Gardens because of their different original climate type. The observation of plant specimens collected from East Nusa Tenggara (Egon Forest, Mutis, and Camplong Park) during 2011-2012 showed that only 50% of plants survive in Purwodadi Botanic Gardens. In contrast, there were 78% of the general plant collection (non-orchid) and 71% of the orchid material collection from Moyo Island, West Nusa Tenggara could survive, implying that the acclimatization of plants from Moyo Island is better than that of Egon Forest, Mutis, and Camplong Park (Trimanto 2013).

Acclimatization of general collection (non-orchid)

The acclimatization provided a favorable condition for the successful adaptation of the tested plants. Our research results revealed that most of the acclimated plant materials grew well in the sand growing media; especially those received the first treatment. The majority of plant materials included in this study were seedling materials, most of which were able to survive in the sand growing media. The successfully adapting seedling materials included *Alstonia spectabilis* R.Br. *Adenanthera microsperma* Teijsm, *Aglaia*

Table 1. Survival percentage of plant collection from Moyo Island

Category	Plant collection	
	General	Orchid
Total accession number	123	17
Survival accession number	96	12
Survival percentage	78%	71%

Table 2. Environmental factors of sites in Purwodadi Botanic Gardens and Moyo Island

Location	Environmental factors			
	Temp. (^{0}C)	Hum. (%)	pH	Light intensity (Lux)
Greenhouse (sand media)-Purwodadi Botanic Gardens	27-28	72-88	6.2	500-8.820
Paranet house (polybag media)-Purwodadi Botanic Gardens	26-29	76-82	6.0	180-3.800
Orchid house-Purwodadi Botanic Gardens	28-29	76-82	6.4	820-17.500
Moyo Island Forest	29-31	70-90	4.8-6.2	4.300-131.000

Table 3. List of plant collections that were not able to adapt in Purwodadi Botanic Gardens

Category	List of accession plant (species)	Family
General collection	*Abrus precatorius*	Fabaceae
	Aglaia lawii	Meliaceae
	Alstonia macrophylla	Apocynaceae
	Bambusa sp.	Poaceae
	Barringtonia racemosa, *Barringtonia* sp.	Lecythidaceae
	Bauhinia purpurea	Fabaceae
	Buchanania arborescens	Anacardiaceae
	Casearia grewiifolia	Samydaceae
	Clausena excavata	Rutaceae
	Clerodendrum paniculatum, *Clerodendrum* sp.	Verbenaceae
	Dioscorea bulbifera, *Dioscorea* sp.	Dioscoreaceae
	Diospyros malabarica	Ebenaceae
	Elatostema rostratum	Urticaceae
	Ficus sp.	Moraceae
	Gigantochloa atter	Poaceae
	Memecylon edule	Melastomataceae
	Palaquium rostratum	Sapotaceae
	Piper retrofractum	Piperaceae
	Pittosporum moluccanum	Pittosporaceae
	Sandoricum koetjape	Meliaceae
	Schoutenia ovata	Tiliaceae
	Syzygium gracilis, *Syzygium* sp.	Myrtaceae
	Uvaria javana	Annonaceae
Orchid collection	*Corymborchis* sp.	Orchidaceae
	Eria sp.	Orchidaceae
	Pteroceras javanica	Orchidaceae
	Dendrobium sp.	Orchidaceae
	Habenaria sp.	Orchidaceae
	Phreatia sp.	Orchidaceae


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Q
R

Figure 2. Several plants from Moyo Island survived in Purwodadi Botanic Gardens nursery. A. *Tacca palmata,* B. *Amorphophallus paeoniifolius,* C. *Polyalthia* sp*.,* D. *Dioscorea hispida,* E. *Adenanthera* sp*.,* F. *Aglaia argentea,* G. *Clerodendrum chinense,* H. *Tacca leontopetaloides,* I. *Cissus javana*, J. *Begonia* sp., K. *Platycerium bifurcatum*, L. *Gymnema* sp., M. *Barringtonia racemosa,* N. *Tetrameles nudifora,* O. *Hymenodictyon excelsum,* P. *Abelmoschus moschatus*, Q. *Schefflera elliptica,* R. *Clausena excavata*

argentea Blume, *Antiaris toxicaria* Lesch., *Hymenodictyon excelsum* (Roxb) *Schefflera elliptica* Blume Harms, *Buchanania arborescens* (Blume), *Amphineuron marginatum* Roxb D.J. Midleton, *Uvaria littoralis* Blume, etc. The plastic used to cover the sand media protected plants from high evapotranspiration rate so that the humidity was preserved. The plant materials prepared in the form of cuttings were not successfully adapted, i.e. none of 10 specimens/cuttings of *Piper retrofractum* could grow while only one of 10 *Pleomele angustifolia* cutting materials were able to grow in sand media. Two species of Bamboo (genus *Gigantochloa*, *Bambusa*) survived only for one month in sand media. Araliaceae family such as *Schefflera elliptica* had high survivorship (up to 80%) as occurred in Bogor Botanic Gardens (Wawangningrum and Puspitaningtyas 2008). Benlate fungicide solution was successfully used to reduce the decay of the cutting materials (Danthu et al. 2002). When the stem cuttings were set in a convenient situation for rooting, the first adventitious roots that appeared from callus became the primary roots for cuttings. Callus contains a high amount of auxin. IBA (indole butyric acid) was found to increase the percentage of rooting in stem cuttings (Saffari 2012).

Several seed materials could grow in sand media. *Abelmoschus moschatus* and species from genus *Polyalthia*, *Ervatamia, Mucuna* also germinated and grew well in sand media. *Abelmoschus moschatus*, *Polyalthia* and *Mucuna* seeds sprouted one month after planting. More than 50 specimens of *Polyalthia* were able to germinate. Species from genus *Ervatamia* could grow only after three months. As with other species, seeds of *Amorphophallus paeoniifolius* also well germinated. The seeds from herbs such as species from genus *Passiflora* and *Ficus* were decaying and could not grow in the nursery. It is important to prevent seeds from rotting in the field. Seeds from the field must be clean and dry to make them prevented from decaying during the field collection. Seeds that are not capable of germinating can be overcome by addition of PGR (Plant Growth Regulators). The PGR was found to speed up the seed germination but gave no effect on the height, diameter and total number of leaves of the plant (Suparwoto et al. 2006). The effect of growth regulators on seed germination has been previously observed by Kumaran (1994), and that on herb seeds was found by Dhoran and Gudadhe (2012).

We found in the present study that epiphytic plants could survive and adapt in the nursery. The native habitat in Moyo Island is dry with low rainfall. *Epipremnum pinnatum, Hoya verticillate* and *Hoya elliptica* grew well in the nursery. The same was true for *Drynaria quercifolia*, *Adiantum caudatum, Platycerium bifurcatum, Lygodium flexuosum,* and species from genus *Asplenium*. The use of moss and potting soil made the epiphytic fern fresh. Tuber plant materials gave successful acclimatization. Five species of tuber plant could grow in the nursery, i.e. *Dioscorea hispida*, *Dioscorea pentaphylla*, *Tacca palmata*, *Tacca leontopetaloides*, and *Amorphophallus paeoniifolius*. Two tuber plant species planted in the garden, i.e., *Dioscorea pentaphylla* and *Tacca leontopetaloides* needed a long time (4 months) to grow. Bulbil planting materials of *Dioscorea pentaphylla* were also collected, and they took about six months to germinate. In Moyo forest habitat, tuber plants can grow well in dryland.

We found in the present study that many plants died after being transferred to polybags. Transferring plants into polybags caused the plants' roots to adapt again with the new growing media; therefore, there is an urgent need for appropriate techniques to grow the plants without changing the growing media/polybag very often. The planting materials should be first planted in polybag media to reduce the stress condition. Growing media consisted of soil: rice husk with a ratio of 1:1/2 could make successful acclimatization. Growing media can be added to compost to supply nutrition for the plant. A good growing media is the one with good water holding capacity, good aeration, good drainage, a pH corresponding to the type of plant, and contains nutrients to support the plant growth. A suitable composition of growing media is needed for a plant to thrive. Dewi (2005) reported that the planting medium with a mixture of sand, cattle dung, and soil in a ratio of 1:1:2 was the best for total plant height and length of shoots in mango nurseries. Susilawati (2007) reported that the media composition of husk charcoal media, soil, and compost in 1:2:1 ratio best influenced the vegetative and generative growths of *Helichrysum bracteatum*. Experiments on the mixture of appropriate growing media are required to determine a suitable growing media in the process of acclimatization in the Purwodadi Botanic Gardens. The addition of fertilizer or compost in the growing media is required to support plant growth. Several researchers found that addition of fertilizer provided significant effect on the growth of a plant. Applying organic-N fertilizer gave the vigorous plants expressed as plant height, leaves size as well as dry weight (Karanatsidis and Berova 2009), and growth and flowering (Bi et al. 2010).

Endemic and rare species are important to be collected in Purwodadi Botanic Gardens. Several threatened species based on IUCN (International Union for Conservation of Nature) Red List can survive in Purwodadi Botanic Gardens. *Canarium littorale* Blume, *Aglaia argentea* Blume, *Tacca leontopetaloides* (L.) Kunze, *Calophyllum soulattri* Burm f, *Alstonia macrophylla* Wall ex G.Don plants with the least concern status in IUCN that were able to survive in Purwodadi Botanic Gardens. *Cycas rumphii* is only one of the Gymnospermae collected from Moyo Island which is listed as near threatened in IUCN. The study about acclimatization is needed to make plant material from their natural habitat able to survive in

Purwodadi Botanic Gardens so that many plant species can be conserved. Several plants from Moyo Island were identified as new collection in Purwodadi Botanic Gardens.

Acclimatization of orchid collection

There were 17 accession numbers of the orchid collection. The diversity of orchids in the exploration site was considered low and more terrestrial orchid species were found than epiphytic orchids. The low diversity of orchids might have been caused by the dry and low rainfall conditions in the forest. The orchid from Moyo Island adapted well to Purwodadi Botanic Gardens climate, although several orchids could not survive in the greenhouse. The acclimatization of orchids resulted in 12 orchids collection could survive while five accession numbers were not able to adapt in Purwodadi Botanic Gardens. The terrestrial orchid bulbs took about four months to germinate as seen in their morphological growth in Figure 3.

Some orchids were found to have dried up, especially the epiphytic type. Epiphytic orchid was acclimatized with fern media watered using sprinkling water without any addition of nutrients. Epiphytic orchids need additional nutrients to grow properly because in their habitat these orchids obtain nutrients from humus in their medium. Tirta (2006) reported that a mixture of ferns and kadaka media (1:1) plus 2.5 mL of fertilizer inabio L^{-1} provided a good growth on *Dendrobium*. Micronutrients are given through the leaf by spraying or watering the plants, so it can be absorbed to cover the nutrition requirements for growth and development. Watering is a major factor for epiphytic orchids' acclimatization. *Vanda limbata* Blume and *Vanda* sp. survived well in the nursery, but the leaves grew poorly.

Several species of orchid such as *Dendrobium* and *Eria* adapted well in tree-fern media although the sprout of this plant grew slowly. The condition of several specimens of *Eria* and *Dendrobium* were dry and decaying. We found no pests and diseases in this plant species, presumably due to its malnourished state. Endemic orchid species collected from Moyo Island was *Pteroceras javanica*. This orchid presumably requires a particular host plant as it was only found in *Schleichera oleosa*. This species survived only for two months in Purwodadi Botanic Gardens. We collected this species several times, but none of them was able to survive, probably due to its need for particular growing media. Tree-fern media has weaknesses such as low water holding capacity, etc. The combination of tree fern with moss may provide water and keep humidity for a longer time, which makes it suitable for an epiphytic orchid.

The accessions of terrestrial orchids looked so fresh, probably due to the availability of nutrients in the soil. Orchids need nutrients to support their growth. The addition of fertilizer is required to satisfy the nutrient needs within the soil. Inorganic and organic fertilizers can support orchid plant development (Jenny et al. 2009). The addition of fertilizers can be done by spraying on the leaves or directly applied on the medium. The combination of growing media in a mixture of compost, soil, and sand in a ratio of 1:1:1 provides a suitable growing condition. The sand makes the media aeration well; the medium pH was normal (6-7) and is free from diseases. The good medium made the bulb of *Nervilia aragoana* Gaodich grow well. *Malaxis latifolia* Blume and *Calanthe triplicata* (Willemet) Ames also grew well in this media. This species could grow and flower after nine months in the greenhouse. There were species of terrestrial orchid that were hard to adapt such as *Corymborchis*. Although being acclimatized the *Corymborchis* could not survive. Similar case occurred to *Corybas ensiformis*, an endemic and terrestrial orchid collected from Egon, East Nusa Tenggara (Fiqa et al. 2011) which was found hard to adapt in the greenhouse of Purwodadi Botanic Gardens.

Figure 3. Orchid from Moyo Island in the nursery at Purwodadi Botanic Gardens. A. *Pteroceras javanica* only survive for two months. B. *Malaxis latifolia*, C. *Nervilia aragoana*, D. *Calanthe triplicata*, E. *Vanda limbata*

Several terrestrial orchids need to interact with mycorrhizae fungi to survive. The mycorrhizas are fungi associations required for the survival of orchids that live naturally in ecosystems since this group of plants depends on the fungi to germinate, as well as for their establishment (Victor 2013). Terrestrial orchids were taken along with soil from their native habitat to anticipate mycorrhizae presence in their possible interaction.

To conclude, the present study results showed that 75% of plants collected from Moyo Island were able to survive in Purwodadi Botanic Gardens. As many 96 accession numbers of the general collection and nine accession numbers of the orchid collection were found to be able to survive. Many of the general plant collections were dead after transplantation. The orchid collection was harder to adapt.

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Determinant factors of the Javan green peafowl (*Pavo muticus muticus* Linnaeus, 1758) habitat in Baluran and Alas Purwo National Parks, East Java, Indonesia

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Abstract. *Hernowo JB. 2017. Determinant factors of the Javan green peafowl* (*Pavo muticus* muticus) *Linnaeus 1758 habitat in Baluran and Alas Purwo National Parks, East Java, Indonesia. Trop Drylands 1: 50-56.* There is limited understanding of the factors determining the habitat variables preferred by the Javan green peafowl (*Pavo muticus muticus* Linnaeus, 1758). Baluran National Park (BNP) and Alas Purwo National Park (APNP) in East Java, Indonesia are habitats of the natural distribution of the Javan green peafowl population. While they occur on various types of habitat (e.g. savanna, beach forest, and monsoon forest in BNP; and grazing area and intercropping teak plantations in APNP), it is not clear what environmental factors affecting their abundance. This research aimed to investigate the determinant factors of Javan green peafowl habitat in correlation with food, water, roosting, shelter, and nesting site. The Principal Component Analysis (PCA) was used to analyze determinant factors of habitat components. The results showed that the determinant factors of the Javan green peafowl habitat in BNP were the density of shelter, the density of food, the number of roost trees and the height of roost trees. Meanwhile, in APNP, the determinant habitat factors were the number of roost trees, the height of roost trees, and the number of continuously available water. The findings of this study highlight the importance of preserving and maintaining the discriminant components of the habitat of the Javan green peafowl in BNP and APNP.

Keywords: Alas Puwo, Baluran, determinant, food, green peafowl, nesting, roosting

INTRODUCTION

The Javan green peafowl (*Pavo muticus muticus* Linnaeus, 1758) is an avifauna native to Southeast Asia. It is listed as Endangered in IUCN Red List due to habitat loss and fragmentation. In general, the peafowl selects particular habitat for feeding sites, sheltering, roosting and nesting site (Hernowo 1995). This Javan green peafowl birds prefer open areas with grasses and shrubs as feeding sites, particular trees as the roosting site, and they also shelter at shaded places close to the feeding site (Hernowo 1995, Hernowo 1999, Hernowo and Hernawan 2003, Hernowo and Wasono 2006, Hernowo 2011).

Determinant factors of the habitat types on the presence of the Javan green peafowl are not clearly understood. Meanwhile, knowledge about the suitable habitat of Javan green peafowl's habitat is vital for the conservation of the bird. Anecdotal evidence indicates that the habitat types are chosen by Javan green peafowl in Baluran National Park (BNP) in East Java, Indonesia include savanna, beach forest, and monsoon forest, but those in the nearby Alas Purwo National Park (APNP) are lowland tropical rain forest, grassland and teak plantation in an intercropping system. The suitability of the habitat is related to the population density of the birds in each habitat type. Hernowo (1997) provided an example in BNP that the green peafowl were more abundant at habitat type of savanna-monsoon forest as compared to other habitats. Meanwhile in APNP, habitat types of Sadengan grassland and intercropping of teak plantations were more preferred than other habitat types (Hernowo and Wasono 2006, Yuniar 2007, Hernowo 2011).

Very few studies on Javan green peafowl habitat have been done particularly regarding the influencing factors of habitat component that might affect peafowl population health. The present research aimed to investigate the determinant factors of Javan green peafowl habitat which have correlation with food, water, roosting, shelter, and nesting site. Those factors are important to be analyzed to determine their impact on habitat type selection.

MATERIALS AND METHODS

Study site

Baluran National Park

Baluran National Park (BNP) is located at the tip of northeastern part of Java Island ($7^0$29`10” - $7^0$55`55” latitude South and $114^0$29`10” - $114^0$39`10” longitude East), covers an area of about 25,000 ha. The national park is bordered by Madura Strait to the north and Bali Strait to the east. The southern west of the park was bordered with Bajulmati and Klokoran Rivers (Hernowo 1995).

The geological situation of BNP is described as part of a small volcano with Plio-Pleistocene deposits. Baluran Mountain is 1247 meters above sea level (m asl) high, close to the center of the national park. Most of the area in the national park is flat (0 - 10 m), except those close to

Baluran Mountain, Priok Mountain, Montor Mountain, and Glengseran Mountain, which are mostly hilly areas. The two major soil types in BNP are volcanic and marine origins. The most important component is volcanic soils that are rich in minerals but poor in organic materials. The soils are high in soil chemical fertility but low in soil physical fertility because of high soil porosity and low soil water holding capacity. Black soil covers about half of the lowland including most of the monsoon forest and savanna grassland (Hernowo 1995).

Baluran has a typical monsoon climate type with a long dry season. This climate type is heavily influenced by the southeast wind during April to October, with less precipitation. The dry period lasts about 7-8 months of the year. The annual precipitation ranges from 900 to 1600 mm. Due to the long dry period, water is the most limiting factor in BNP. The local distribution of wild animals is influenced by water availability. During the dry season, animals can easily be observed near the water hole, but in the rainy season, they spread everywhere (Hernowo 1995).

The vegetation types that have developed in BNP include savanna grassland, beach forest, mangrove, deciduous forest or monsoon forest, evergreen forest, swampy area and sub-mountain forest. Mangrove occurs at Bilik, Lempuyangan, Mesigit, Tanjung Sedano and Kelor. Typical vegetation at mangroves is *Avicennia alba*, *Sonneratia caseolaris, Ceriops tagal, Rhizophora apiculata, Bruguiera gymnorrhiza*, and *Luminitzera racemosa*. Beach forest presents between Pandean and Tanjung Candibang and in some places, such as Labuan Merak and also east of Gatal. This type of forest is dominated by *Barringtonia racemosa*, *Terminalia catappa, Pandanus tectorius*, and *Hibiscus tiliaceus*. The savanna grassland with fire-climax vegetation is strongly influenced by human intervention. The dominant tree species in that area are *Acacia nilotica* (an introduced African exotic species), a few *Acacia leucophloea, Schleichera oleosa, Zizyphus rotundifolia* and *Corypha utan*. Dominant grass species are *Dichantium coricosum, Brachiaria mutica*, and *Sorgum nitidus*. Monsoon forest is characterized by dominant tree species of *Tamarindus indica, Schoutenia ovata, Grewia eriocarpa, Flacortia indica, Cordia obliqua, Azadirachta indica*, and *Sterculia foetida*. Mountain forests and evergreen forests consist of trees species including *Mallotus philippensis*, *Homalium foetidum*, *Emblica officinalis*, and *Aleurites moluccana* (Hernowo 1995).

Wild animals existing in BNP that have relation with green peafowl are leopard (*Panthera pardus*), civet (*Viverra malacensis*, *Paradoxurus hermaphroditus*), mongoose (*Herpestes javanica*), red dog (*Cuon alpinus*), piton (*Phyton reticulatus*), monitor (*Varanus salvator)* and serpent eagle (*Spilornis cheela*) (Hernowo 1995).

Alas Purwo National Park

Alas Purwo National Park (APNP) covers an area of about 43,420 ha. The national park is located at the tip of southeastern Java Island ($8^0$26`45” - $8^0$47`00” latitude South and $114^0$20`16” - $114^0$36`00” longitude East). The eastern part of the national park is bordered by Bali Strait and the south and west parts are bordered by the Indian Ocean. An intensive study was focused on Sadengan grazing area, lowland tropical forest and teak forest plantation of Rowobendo. Topography at the national park consists of flat area (0 – 8% slope) of about 10,554 ha, undulating area with the slope of 8 – 15% around 19,474 ha, meanwhile rolling part (15 – 25% slope) around 11,901 ha and a small portion of mountainous areas about 2301 ha. There are four types of soil in study area, i.e. Mediterranean red litosol complex about 2106 ha, gray regosol 6238 ha, gray grumusol about 379 ha and alluvial hydromorph at around 34,697 ha. Numerous small streams flow at AP National Park with a radial flowing pattern. All of the rivers flow into the Indian Ocean. Several underground rivers occur at karst complex such as Pancur River (Hernowo et al. 2011).

According to Smith and Ferguson climate classification, the study area was classified as B climate type with annual precipitation ranging from 1079 to 1554 mm per year and 79 - 112 rainfall days. The mean annual temperature is around 27.1 °C, and relative humidity is about 85% (Hernowo et al. 2011).

Five types of vegetation have developed in Alas Purwo national park, i.e. beach forest, mangrove, lowland tropical forest, bamboo forest, and teak plantation. Beside those vegetation types, human-made grazing area occurs at Sadengan. Beach forest occurs at the southern park from Grajagan to Plengkung about 30 km and Plengkung to Tanjung Slakah around 50 km. The beach forest is also found in about 40 km at the northern park. The dominant species at the beach forest are ketapang (*Terminalia catappa*), waru (*Hibiscus tiliaceus*), keben (*Barringtonia asiatica*) and nyamplung (*Calophyllum inophyllum*). Mangrove is existing at Grajagan with species of vegetation such as bakau (*Rhizophora* spp), tanjang (*Bruguiera* spp), api-api (*Avicennia* sp), pedada (*Sonneratia caseolaris*) and nyirih (*Xylocarpus granatum*). Tropical lowland forest has a big portion at the park. The vegetations that exist in those forests are *Ficus* spp, bendo (*Artocarpus elastica*), rao (*Dracontomelon mangiferum*), pule (*Alstonia* spp), santen (*Lannea grandis*), gintungan (*Bischovia javanica*), and pohpohan (*Buchanania arborescens)*. Vegetation types found in a drier condition in the forest are kepuh (*Sterculia foetida*), asam (*Tamarindus indica*), and randu alas (*Bombax valetoni*). Besides, bamboo formation and consociation of sawo kecik (*Manilkara kauki*) also occur in the park (Hernowo et al. 2011).

Several wild animals existing at APNP that might have relation to green peafowl are leopard (*Panthera pardus*), wild boar (*Sus scrofa*) civet (*Paradoxurus hermaphroditus*), mongoose (*Herpestes javanica*), red dog (*Cuon alpinus*), piton (*Phyton reticulatus*), monitor (*Varanus salvator)*, serpent eagle (*Spilornis cheela*) and white-bellied sea eagle (*Haliaeetus leucogaster*) (Hernowo et al. 2011).

Figure 1. Map of study site in A) Baluran National Park (BNP); and B) Alas Purwo National Park (APNP), East Java, Indonesia.

Methods

This research was conducted at Baluran and Alas Purwo National Parks from June to October 2006 and August to December 2007. The study was focused on the local distribution of Javan green peafowl at Bekol resort (savanna, beach forest, and monsoon forest) in Baluran National Park (BNP) and at Rowobendo resort (Sadengan grazing area, intercropping area teak plantation) in Alas Purwo National Park (APNP).

The determinant habitat components that have an influence on the presence of the javan green peafowl at certain habitat types were analyzed using PCA (Principal Component Analysis) method (Rencher 2002) as follow:

Y_j = Principle component of-j (j = 1, 2,....n)

$X_{1,2,3,...,n}$ = Variable of habitat component 1,2,3,...,n

$a_{1j}, a_{2j}, a_{3j}, a_{nj}$ = Eigen vector variable of, 1,2,3,…,n with priciple component of j

Where,

Y = Individual number of the green peafowl at each habitat type

X_1 = Number of food species

X_2 = Density of food

X_3 = Number of roost trees

X_4 = Height of roost trees

X_5 = Number of continuously water

X_6 = Size of dancing area

X_7 = Density of shelter

X_8 = Density of cover

The PCA model was tested by Kaiser-Meyer-Olkim (KMO) test and Bartlett test. The Kaiser-Meyer-Olkim (KMO) test was used to measure the adequacy and Bartlett test was used to know the sphericity of the variables with a significant correlation. If the value analysis of KMO is > 0.5, then the model is feasible to be continued.

Anti-Image Matric used with the criteria are as mentioned below: (i) MSA (Measure Sampling Adequacy) = 1, the variable can be predicted with zero mistakes, (ii) MSA (Measure Sampling Adequacy) > 05, the variable can be predicted and the analysis is continued, (iii) MSA (Measure Sampling Adequacy) < 0.5, the variable cannot be predicted and the analysis is not continued.

Commonalities are processes to show how much variance can be explained by formed factors with multiple squares of the correlation value.

Total Variance Explained is demonstrated by variance value which is also known as the eigenvalue. Eigenvalue is composed of the biggest value to the smallest, and the value used if it is > 1.

Component Matric is a table composed of loading factors (Correlation Value) between analysis variable with factors dominant that has been formed.

RESULTS AND DISCUSSION

Influence of habitat type on the abundance of Javan green peafowl

Results of the analysis of variance revealed that the effect of habitat types on the abundance of the bird was significant (F = 68.74, P<0.01) in BNP. Duncan`s Multiple Range Test showed that the bird's abundance differed among habitat types as shown in Table 1. The difference was caused by the availability of feeding sites, nesting sites, roosting sites, and water resources (Hernowo 1999). In Baluran National Park, the javan green peafowl was more abundant in Savanna Bekol (Pattaratuma 1977; Mulyana 1988; Winarto 1993; Hernowo 1995; Hernowo 1999; Yuniar 2007; Risnawati 2008; Hernowo et al. 2011).

Table 1. Variance analysis of Javan green peafowl abundance at several habitat types in Baluran National Park, East Java, Indonesia

Habitat types	Mean abundance
Savanna Bekol	47.20[a]
Beach Forest Bama-Manting	7.65[b]
Monsoon Forest Bekol	7.80[b]
Evergreen-Monsoon Forest Bekol	7.25[b]

Note: Means within the same column with the same letter (s) are not significantly different at 0.05 DMRT.

Table 2. Variance analysis of Javan green peafowl abundance at several habitat types in Alas Purwo National Park, East Java, Indonesia

Habitat types	Mean abundance
Grazing area with Lowland TRF, Sadengan	27.85[b]
Teak Plantation & Intercropping, Gunting	36.90[a]
Mix Plantation & Intercropping, Rowobendo	8.55[c]
Teak Plantation, Sumber Gedang	2.50[d]
Teak Plantation, Ngagelan	2.35[d]

Note: Means within the same column with the same letter (s) are not significantly different at 0.05 DMRT

As with BNP, variance analysis results also demonstrated a significant effect of habitat type (F= 163.55, P< 0.01) on the abundance of green peafowl in APNP. Similarly, Duncan's Multiple Range Test showed a significant difference in the birds' mean abundance among the habitat types as shown in Table 2.

The Duncans' post hoc test showed that the abundance of the Javan green peafowl differed by habitat type in Alas Purwo National Park. The Javan green peafowl was distributed in a higher number in Sadengan grazing area with a lowland tropical forest (TRF) as compared to other habitat types. A plausible explanation for this is that the Sadengan grazing area with lowland tropical forest provided more availability of food resources, sheltering site, roosting site, and cover site (Supratman 1998; Hernowo and Wasono 2006; Yuniar 2007; Risnawati 2008; Hernowo et al. 2011).

Analysis of determinant factors of habitat component in Baluran National Park (BNP)

Analysis of variance of habitat component variables was done for several habitat types in Baluran National Park (BNP). The analysis result showed that eight variables of habitat component could be grouped into two principal components with represent value of 61.50% (Table 3).

Based on values of loading factors, the first principal component consisted of the density of shelter (x_7), and the second principal component consisted of the density of food (x_2), the number of roost trees (x_3), and the height of roost trees (x_4). The determinant factors of Javan green peafowl habitat in Baluran National Park were the density of shelter (x_7), density of food (x_2), the number of roost trees (x_3), and the height of roost trees (x_4) (Table 4 and Figure 3).

The result of PCA analysis above suggests that there were two principal components of the determinant factor that influenced the individual abundance of Javan green peafowl. The first principal component of habitat determinant factor was the density of shelter site and the second principal components were the density of food, number of roost trees and high of roost trees. During hot days, the Javan green peafowl is sheltering under a lush tree at Savanna Bekol (Pattaratuma 1977; Mulyana 1988; Winarto 1993; Hernowo 1995; Hernowo 1999; Yuniar 2007; Risnawati 2008; Hernowo et al. 2011). The density of shelter sites in BNP is critical during the dry season. In the dry season, most of the area in BNP becomes harsh, and the Javan green peafowl is usually sheltering from 9.30 a.m – 14.00 p.m as they need the availability and density of the housing site (Hernowo 1995; Hernowo 1999).

The density of food in savanna Bekol is high in forms of grasses and shrubs species (Pattaratuma 1977; Mulyana 1988; Winarto 1993; Hernowo 1995; Hernowo 1999; Yuniar 2007; Risnawati 2008; Septania 2009; Hernowo et al. 2011). Leaves and fruits or seeds of grasses and shrubs are the main diet for green peafowl. There were 19 species of grasses and 16 shrubs recorded to be eaten by green peafowl in BNP. The green peafowl feeds on leaves, flower, and seeds of the grasses and shrubs. The Javan green peafowl feeds on quite broad range of species of grasses and shrubs (Rini 2005). The Javan green peafowl also belongs to polyphagous species, meaning that this bird feeds on a quite wide range of kinds of food (Septania 2009). The green peafowl sleep on the tree (Pattaratuma 1977; Mulyana 1988; Ponsena 1988; Hernowo 1995). According to Hernowo (1999), the trees selected by the Javan green peafowl as roost site at BNP were 12 – 20 m high, and the preferred roost trees were Pilang (*Acacia leucophloea*) and dead Gebang (*Corypha utan*) (Figure 2).

Analysis of determinant factors of habitat component in Alas Purwo National Park (APNP)

Analysis of component variables at several habitat types in Alas Purwo National Park (APNP) showed that eight variables habitat components could be grouped into three principal components, representing a value of 62.30% proportion (Table 5).

Based on the eight habitat component variables we analyzed, three principal components became the determinant factors. The first principal component was the number of roost trees (x_3) and the second principal component was the height of roost trees (x_4). The third principal component was the number of continuous water (x_5). The determinant factors of Javan green peafowl habitat in Alas Puwo National Park were the number of roost trees (x_3), the height of roost tree (x_4), and the number of continuous water (x_5) (Table 6).

Table 3. Eigen factors analysis of several habitat types of the Javan green peafowl in Baluran National Park, East Java, Indonesia year 2006 and 2007

Habitat Component Variables*	Eigen Vector		
	Total	% Proportion	% Cumulative
X_1	**2.0062**	0.251	0.251
X_2	**1.7171**	0.215	0.465
X_3	**1.1999**	0.150	0.615
X_4	0.9732	0.122	0.737
X_5	0.7996	0.1000	0.8370
X_6	0.6044	0.0760	0.9130
X_7	0.4396	0.0550	0.9670
X_8	0.2601	0.0330	1.0000

Note: Number of food species (x_1), Density of food (x_2), Number of roost tree (x_3), Height of roost tree (x_4), Number of continuous water (x_5), Size of dancing area (x_6), Density of shelter site (x_7), Density of cover site (x_8).

Table 4. Loadings factors of principles component at several habitat types of the Javan green peafowl in Baluran National Park, East Java, Indonesia year 2006 and 2007

Habitat Component Variables	Value of Principles Component	
	1	2
Number of food species (x_1)	0.391	-0.086
Density of food (x_2)	0.472	**0.540**
Number of roost trees (x_3)	0.094	**0.506**
Height of roost trees (x_4)	0.140	**-0.578**
Number of continuous water (x_5)	0.396	-0.126
Size of dancing area (x_6)	0.141	0.177
Density of shelter sites (x_7)	**0.632**	-0.252
Density of cover sites (x_8)	-0.138	0.026

Table 5. Eigen factors analysis at several habitat types of the green peafowl in Alas Purwo National Park, East Java, Indonesia year 2006 and 2007

Habitat Component Variable*	Eigen Vector		
	Total	% Proportion	% Cumulative
X_1	**2.1724**	0.272	0.272
X_2	**1.6374**	0.205	0.476
X_3	**1.1766**	0.147	0.623
X_4	0.8920	0.111	0.735
X_5	0.7357	0.0920	0.8270
X_6	0.6307	0.0790	0.9060
X_7	0.4248	0.0530	0.9590
X_8	0.3304	0.0410	1.0000

Note: Number of food species (x_1), Density of food (x_2), Number of roost tree (x_3), Height of roost tree (x_4), Number of continuous water (x_5), Large of dancing area (x_6), Density of shelter site (x_7), Density of cover site (x_8).

A B

Figure 2. A) Female Javan green peafowls are roosting at dead gebang; B) Female Javan green peafowls are feeding at Savanna Bekol in Baluran National Park, East Java, Indonesia

A B

Figure 3. A) Male of java green peafowl is roosting at teak; B) Male bird is drinking on puddle filled up with water

Table 6. Determinant factors of principles component at several habitat types of the Javan green peafowl in Alas Purwo National Park, East Java, Indonesia year 2006 and 2007

Habitat Principles Component	**Value of Principles Component**		
	1	**2**	**3**
Number of food species (x_1)	-0.398	0.487	-0.086
Density of food (x_2)	-0.401	0.347	-0.352
Number of roost trees (x_3)	**-0.547**	-0.185	-0.149
High of roost trees (x_4)	-0.056	**-0.572**	-0.024
Number of continuous water (x_5)	0.075	-0.255	**-0.710**
Large dancing area (x_6)	0.342	0.336	-0.389
Density of shelter site (x_7)	-0.333	-0.321	-0.217
Density of cover site (x_8)	-0.381	0.008	0.379

In APNP, water was continuously available at Sadengan grazing area because the water was flowing from water resources of Basori cave. This finding is similar to other studies that found the number of green peafowl in Dak Lak Province, Vietnam was more abundant when closed to the river bank (Brickle 2002). Ponsena (1988) also stated that the individual number of green peafowl at Huai Kang Khaeng Wildlife Sanctuary was more abundant in the riparian area.

In Sadengan grazing area of APNP, Apak was mostly preferred by the Javan green peafowl as a roosting site (Supratman 1998; Hernowo and Wasono 2006; Yuniar 2006; Risnawati 2008). Subramanian and John (2001) reported that the Indian blue peafowl (*Pavo cristatus*) at

Reserve forest of Deer Park, Tirunevelvi Tamil Nadu preferred roosting on tamarind (*Tamarindus indicus*), vagai (*Albizia lebbeck*), neem (*Azadirachta indica*), usilai (*Albizia amara*), and palmyra (*Borassus flabellifer*) and also less frequently on manjanathi (*Morinda tenctoria*) and velvelam (*Acacia leucophloea*). At Vivekananda Kendra, the bird primarily preferred roosting on coconut palm (*Cocos nucifera*), while tamarind, neem, manggo (*Mangifera indica*), and umbrella thorn (*Acacia planifrons*) were the second choice. Even telecommunication pylons are used as a roosting site. Pilang (*Acacia leucophloea*) and dead Gebang (*Corypha utan*) as the preferred roost trees were sporadically distributed and low in density, especially those with height above 12 m.

Based on the results of this study, we concluded that the determinant factors of the Javan green peafowl habitat in Baluran National Park were the density of shelter, the density of food, the number of roost trees and height of roost trees. Meanwhile, in Alas Purwo national park, the determinant habitat factors were the number of roost trees, the height of roost trees, and the number of continuous water.

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Evaluation of five pearl millet ecotypes susceptibility to the nymphal instars of migratory locust

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Abstract. *Gomaa KFS, Bashir MO. 2017. Evaluation of five pearl millet ecotypes susceptibility to the nymphal instars of migratory locust. Trop Drylands 1: 57-63.* African migratory locust (*Locusta migratoria migratorioides*) is among the most devastating pests in African dry land, threatening the sustainability of agricultural crops including the pearl millet (*Pennisetum glaucum*). This study aimed to investigate the susceptibility of five ecotypes of pearl millet to the nymphal instars of the solitary African migratory locust and to detect the most preferred millet variety. The experiments were undertaken under semi-field (cage-experiment) conditions. Five different experiments with five ecotype varieties of pearl millet *Pennisetum glaucum* var. were setup. Each experiment with three replications was applied topically on the 3rd and 4th nymphal instars. The food preference and ecotype susceptibility evaluation were based on the amount of food intake in grams, weight of food ingested and digested in gram, percentages, and amount of weight of feces in gram. The results showed a significant difference in susceptibility of five ecotypes of pearl millet. The susceptibility of feeding by 3rd and 4th nymphal instars was observed in all ecotypes. The study results showed that the preference of ecotypes by nymphs was increased according to increasing application time. The highest one was Bayouda-late maturing compared to four other ecotypes. During the experiments, we noticed that the preferences of five ecotypes were elevated in the first and third readings of experiments. However, we discovered in the study that the susceptibility of five pearl millet ecotypes was according to the remaining amount of feces in any ecotype variety. The least amount of reminder feces was in Bayouda-late maturing ecotype, which means that most of the food intake was ingested, digested and assimilated as with the four other ecotypes. The greater percentages of ingested amount of food consumption for five ecotypes of pearl millet, with 3rd and 4th instars of African migratory locust, ranked as mentioned above. The research concludes that further works on evaluation of susceptibility of pearl millet varieties with different pests, particularly in migratory locust is recommended to detect the palatable target variety as a trap in pest management control.

Keywords: *Pennisetum glaucum*, ecotypes, nymphal instars, migratory locust

INTRODUCTION

Locusts belong to a large group of insects commonly called short-horned grasshoppers within the superfamily Acridoidea, and the most important locusts are all in the family Acrididae (Anonymous 1966). They differ from grasshoppers in their behavior and phase of transformation. An important feature of locusts is their ability to transform reversibly between the two extreme phases of solitaria and gregaria which differ in morphology, physiology, and behavior (Uvarov 1966). Gregarious locusts have the tendency to stay together in dense groups, march in bands as wingless hoppers or adult swarms over long distances (Steedman 1988).

Locusts are considered among the major pests in the Sahel zone of Africa. The migratory locust *Locusta migratoria* (Reiche and Farmaire) belongs to a monospecific genus of *Locusta* Linnaeus. Migratory locust is one of the most important destructive agricultural pests in the world, and its outbreaks were recorded early in the 13th century BC (Fan 1983; Vijay et al. 2013). Migratory locust is a highly migratory species with a greater distribution in temperate and tropical regions of the eastern hemisphere (Asia, Europe, Africa and Australia) and was also found to present up to 4600 m above sea level in the Tibet Plateau (Guo et al. 1991; Meinzingen 1993).

Previous studies (Uvarov 1921, 1977) reported that *L. migratoria* also has significant phase polymorphism ability including changes in their morphology, physiology, and behavior with transitory morphs between solitarious to gregarious phases or vice versa. Both nymphs and adults of the locust exhibit density-dependent phase polymorphism and they also have cryptic body coloration at low-density conditions. This body coloration includes green, orange, brown, or black and is sedentary in nature. At low density, the locust also shows little or no tendency to aggregate. Besides having morphological changes, however, when their population density increases then both nymphs and adults show strong tendency for aggregation and dispersion. Nymphs march in bands on ground while adults form swarms and migrate over long distances, sometimes several hundred kilometers (Uvarov 1977).

Traits change in morphology, coloration, physiology, and behavior in the migratory locust and its wide distribution lead to identification of different species of this species. Thus, this species was differently named as *L. australis, L. danica, L. gallica, L. rossica,* and *L. solitaria*. Seven subspecies within *Locusta migratori* that are recognized at present are *L. m burmana, L. m capito, L. m cinerascens, L. m migratoria*, L. *m manilensis*, *L. m migratorioides, and L. m tibetensis* (Zhang et al. 2009). Some studies have identified differentiations among various populations of *L. migratoria* and three genetically different groups viz *L. m migratoria*, *L. m manilensis*, and *L. m tibetensis*. Dispersal routes of the migratory locust show that global populations can be divided into two different lineages, the northern lineage, and southern lineage.

The outbreak of *L. migratoria* has caused severe damage to the pasture land and agriculture besides terrifying people and causing traffic accidents by the swarming locusts (Kumar and Ramamurthy 2009). Summer breeding takes place in eastern Sudan especially in Elgedaref state, Gezira State, Sinar State, Blue Nile State, Northern State and River Nile State (Hamid 2003).

Millet (*Pennisetum glaucum* L.) is one of the preferred cereals besides the wheat, rice, and maize attacked by the pest. Millet is a major food source for millions of people, especially those who live in hot, dry areas of the world. It is grown mostly in marginal areas under agricultural conditions in which major cereals fail to give substantial yields (Adekunle 2012). Millet is classified with maize, sorghum, and Coix (Job's tears) in the grass subfamily Panicoideae (Yang et al. 2012). Millet is an important food in many underdeveloped countries because of its ability to grow under adverse weather conditions such as limited rainfall. Millet is the primary source of energy and protein for millions of people in Africa. Previous studies reported that millet has many nutritious and medical attributes (Obilana and Manyasa 2002; Yang et al. 2012). Millet is a drought-resistant crop and can be stored for a long time without insect damage (Adekunle 2012); hence, it can be important during famine.

Millets are unique among the cereals because of their richness in calcium, dietary fiber, polyphenols and protein (Devi et al. 2011). Millets contain significant amounts of essential amino acids particularly the sulfur-containing amino acids (methionine and cysteine); they are also high in fat content than maize, rice, and sorghum (Obilana and Manyasa 2002). They provide fatty acids, minerals, vitamins and typical millet protein contains high quantity of essential amino acids (FAO 2009).

However, millets are subjected to many insect pests, and the most dangerous ones are locusts. The locust concentrations are often found to be associated with particular species of food plants. Among the cultivated crop, we discovered a close association between locust and millet crop which might be due to the type of variety and cropped area, ecotype, or other factors. There are more than five varieties of *Pennisetum glaucum* in the Darfur region, Sudan but these five varieties are frequently found in the locust habitat. An advantage of testing this ecotype is that it is possible to know the preferred variety, which will make the survey of locusts more limited. Knowing the preferred variety will be very cost-effective as well as time and effort saving. It will also help to suggest and detect any gregarisation that may happen. It is better to control the locust before they swarm and migrate as it may be too late to stop the damage they cause.

This study investigates and evaluates the susceptibility of five ecotypes of Pearl millet (*Pennisetum glaucum*) to the nymphal instars of the solitary African Migratory Locust *Locusta migratoria migratorioides* and detects the most preferred millet variety.

MATERIALS AND METHODS

We carried out this study under laboratory and semi-field conditions at the Biological Control Unit of Insectary Laboratory, Department of Crop Protection, Faculty of Agriculture, University of Khartoum, Shambat, Sudan during the period from May 2014 to December 2014 (Temp. 22 ˚C-39˚C, R.H 17-19%, and normal daylight).

Rearing of migratory locust

The culture (Figure 1) was started from the initial material (nymphs and mature adult individuals of the migratory locust obtained from eastern part of Sudan, Elgedarif State (Elmaganez District) which lies on Latitude N 14 31 14.4 and Longitude E 035 12 45.0. Two hundred individual nymphs and mature adult locusts of male and female were used to start the mass rearing. The new hatches were reared up to the second generation to ensure the homogeneity of the population. The third and fourth instars were then used as the experimental insects.

Rearing and egg-laying cages

The rearing cage was made of mosquito wire mesh sides; the bottom was made of plywood. It measured 70 cm × 60 cm ×50 cm. One side of the cage was covered with a light cloth in the form of a sleeve to facilitate the easy handling of insects to perform various activities inside the cage such as feeding and cleaning without the insects being able to escape from the cage. On the bottom surface of the cage, six holes were made for fixing plastic cups, filled with a wet mixture of sandy clay soil (3:1). These cups were used to provide sites for egg laying. The insects were fed on millet and fresh sorghum fresh leaves, and wheat bran. The rearing-egg laying cage was cleaned daily from insect fecal pellets, with a brush. The insects were monitored, and various activities of mature adults including soil probing, copulation, and egg-laying were observed. After the eggs had been laid, the cups which contained sufficient egg pods were removed and replaced by new ones, using gloves to protect hands and mask to avoid odors. The cups containing the egg pods were covered with cheesecloth and the soil periodically moistened until egg hatching. The hatched nymphs were reared up to the 3^{rd} and 4^{th} instars, which were used in the experiments.

Hoppers rearing cage

The hoppers rearing cage measured 40 cm x 30 cm x 30 cm (Figure 2) was made of wood and mosquito wire net on four sides. The fifth is side made of light cloth fitted with a zip fastener, in the form of a strip. This zipper was made to facilitate carrying out the activities of feeding and cleaning by hand inside the cage, without the insects being able to escape away from the rearing cage.

Food materials

Five varieties of millet plants were brought from the Darfur State. These five ecotype varieties included: (i) Early Bayouda is dark gray, medium and strong in seeds, it is grown in North Darfur, Saraf Omra area in cracked clay soil. (ii) Late Bayouda is slightly yellow, long stack, large and strong and has conveyed seeds, grown in cracked clay soil in Jebal Mara. (iii) Wad Elahow late-maturing is yellowish and spherical seeds, long plant; it is grown in south of Nyala semi cracking clay soil. (iv) Kano is slight gray small seeds, long plant; it is grown in cracking loamy clay soil southwest of Nyala. and (v) Dembi is relatively a dwarf short day to maturity, a red seeded variety grown in North Darfur Shangil Toobaya area. The taller, longer-season and white-seeded type have different names in different places (Abuelgasim 1989; Sabil 1991; Abuelgasim 2011).

These five ecotypes were grown in ground basins of 200 cm ×100 cm. Millet seedlings of the five varieties were grown in plastic cups to provide daily feed to the nymphs and adults (Figure 3). Also, wheat bran was provided for additional supplementary feeding. The cages were checked daily for cleaning and provision of food. Adult locusts that emerged were transferred to egg-laying cages.

Experimental cages

The cages used in the tests were made of wood and wire mesh. Each cage measured 25 cm × 25 cm × 30 cm (Figure 4). Millet was provided as food for the treated migratory locust. The insects were treated topically and then released into the cages.

Food preference experiments (feeding tests)

Five varieties or ecotypes of Millet plant were sown in five different ground basins. Then after emerging and during shooting period, before application experiments, 20 grams of fresh plant shoot were weighed and dried for later parameters calculation. The plant shoots were weighed using a sensitive balance and then placed inside the cages which contained twenty nymphs which were considered as an experimental replication unit. The experiment was arranged in a completely randomized design (CRD) and factorial experiment (FE) with three replications. The feeding rate of the five millet plant ecotypes and survival of the tested insects were used as parameters to evaluate preference.

The weight of food consumption (food intake) of each ecotype fed by hoppers (3rd and 4th instars) was recorded after 24 hrs, 48 hrs and 72 hrs for each experiment. The weights of fecal pellets in all cages for three days were also recorded. The parameters (ingested food, assimilated food, fecal pellets and their percentages) were calculated using the following equations:

Ingested food (D) = A-B.
Assimilated food (E) = A- (B+C).
Ingested food % = D×100/A.
Assimilated food % = (B+C) ×100/A.
Where:
A: Wet weight of shoot amount. B: Spill, C: Feces

Statistical analysis

This study was assigned in a factorial experiment laid out in a Completely Randomized Design (CRD). The first factor was pearl millet ecotypes (five ecotypes/varieties) and the second was time of observation (24 hrs, 48 hrs, and 72 hrs). The obtained data were analyzed according to SAS program version 3, SAS 1997. The accepted level of significance was ≤ 0.05 and differences between the treatment means were detected using the least significant difference (LSD) according to Gomez and Gomez (1984).

Figure 1. Oviposition unit setup for rearing of migratory locust

Figure 2. Migratory locust hopper's rearing cage

Figure 3. Millet seedlings grown in plastic cups and used to feed adults and nymphs.

Figure 4. Experimental cages

RESULTS AND DISCUSSION

Food preference (Feeding tests)

The results of all treatments indicated significant difference in the susceptibility of ecotypes. The result presented in Table 1 and Figure 5 showed a significant difference in the susceptibility of five pearl millet ecotypes. The preference of ecotypes by nymphs increased as the time of observation increased.

The highly susceptible variety was Bayouda-cold resistant (V5) compared to four other ecotypes of treatments. The others were Kano-late maturing (V3), Wad elahow-late maturing (V4), Bayouda-early maturing (V2) and Dembi-short maturing (V1), respectively.

Table 1: Amount of food-intake (in gram) by the migratory locust nymphs

Ecotype	24hrs	48hrs	72hrs	Mean	SE±
V1	16.90±0.44	17.93±0.79	18.15±1.52	17.66	0.92
V2	16.69±0.24	17.49±0.49	18.42±0.88	17.54	0.64
V3	17.29±1.18	18.20±0.36	18.27±0.28	17.92	0.61
V4	16.48±0.67	18.29±0.84	18.54±0.65	17.77	0.72
V5	17.82±0.78	18.19±1.26	18.46±0.40	18.15	0.81
Mean	17.04	18.02	18.37	17.81	0.74

Note: V1: Dembi-short maturing, V2: Bayouda-early maturing, V3: Kano-late maturing, V4: Wad elahow-late maturing and V5: Bayouda-cold resistant

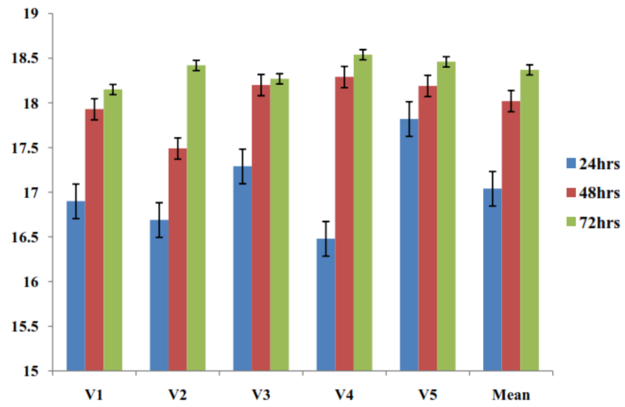

Figure 5. Food-intake in gram of the 5 test varieties by the 3rd and 4th migratory locust nymphs. Note: V1: Dembi, V2: Bayouda-early, V3: Kano-late, V4: Wad elahowV5: Bayouda-late.

Percentage of food intake by migratory locust hoppers on the five millet ecotypes.

The percentages of five pearl millet ecotypes intake by hoppers of migratory locust are presented in Table 2 and Figure 6. There was clear significant difference among the five ecotypes. The highest percentage was recorded in Bayouda-cold resistant V5 ecotype (90.4%), next is Wad elahow-late maturing V4 ecotype (89.4%), followed by Kano-late maturing V3 ecotype (89.3%), then Dembi-short maturing V1 ecotype (89.3%) and Dembi-short maturing V2 ecotype (87.8%). During the experiments, it was noticed that the preference of ecotypes V1 and V5 after the second day, the percentage of food intake was similar. However, the percentage of food-intake of ecotype V5 was highest on the first and third day of experiments.

Weight of ingested food

Table 2 and Figure 7 show clear variation in amount of food consumption of five ecotypes with 3rd and 4th instars of migratory locust. The variations were 17.5 g in Kano ecotype, 17.48 g in Wad elahow ecotype, 17.24 g in Bayouda-early ecotype, 17.12 g in Bayouda-late ecotype and 17.04 g in Dembi ecotype. The lowest amount of food ingested in the experiment was 17.04 g in Dembi ecotype variety, and the highest amount ingested of food taken by hoppers, compared with other four ecotype varieties of pearl millet was 17.5 g in Kano ecotype.

Table 2: Food intake by Migratory locust hoppers of the five millet ecotypes

Ecotype	24hrs	48hrs	72hrs	Mean
V1	88.5%	90.5%	89.1%	89.3%
V2	88.1%	86.6%	88.7%	87.8%
V3	89.2%	90.2%	88.6%	89.3%
V4	90.4%	88.2%	89.5%	89.4%
V5	90.8%	90.4%	90 %	90.4%
Mean	89.4%	89.1%	89.2%	89.2%

Note: V1: Dembi-short maturing, V2: Bayouda-early maturing, V3: Kano-late maturing, V4: Wad elahow-late maturing and V5: Bayouda-cold resistant

Table 3: Weight of ingested food in grams

Ecotype	24hrs	48hrs	72hrs	Mean	SE±
V1	16.89±2.60	17.41±1.21	16.83±1.05	17.04 g	1.62
V2	17.40±1.69	17.1±1.05	17.23±1.29	17.24 g	1.34
V3	16.72±0.95	17.65±2.66	18.12±0.34	17.5 g	1.32
V4	17.07±1.89	17.61±3.47	17.75±0.46	17.48 g	0.86
V5	16.71±0.68	16.9±1.14	17.76±0.79	17.12 g	0.87
Mean	16.96	17.33	17.54	17.28 g	1.2

Note: V1: Dembi-short maturing, V2: Bayouda-early maturing, V3: Kano-late maturing, V4: Wad elahow-late maturing and V5: Bayouda-cold resistant.

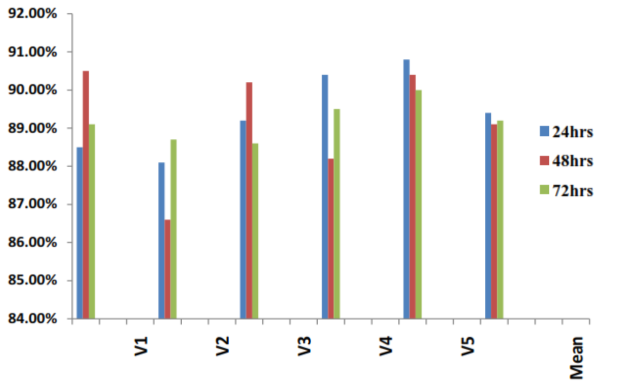

Figure 6. Percentage of Ecotypes preference or food intake by Migratory locust hoppers. Note: V1: Dembi, V2: Bayouda-early, V3: Kano-late, V4: Wad elahow, V5: Bayouda-late.

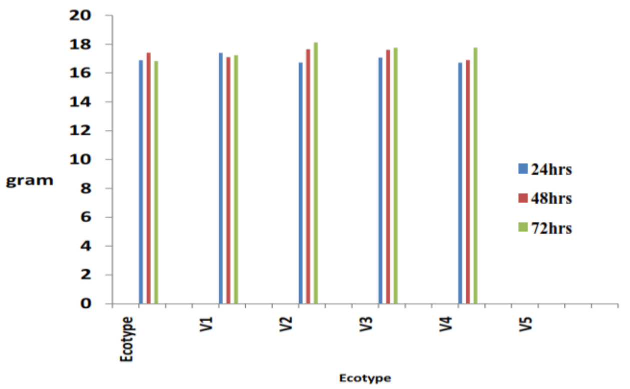

Figure 7. The weight of food ingested. Note: V1: Dembi-short maturing, V2: Bayouda-early maturing, V3: Kano-late maturing, V4: Wad elahow-late maturing and V5: Bayouda-cold resistant.

Percentage of ingested food

Table 4 and Figure 8 show the different percentages of amount of ingested food of five ecotypes of pearl millet *Pennisetum glaucum* variety, by 3rd and 4th instars of migratory locust. The difference percentage was 98.2% in Wad elahow ecotype (V4), 97.3% in Bayouda-late ecotype (V5), 96.7% in Kano ecotype (V3), 96.5% in Dembi ecotype (V1) and 96.1% in Bayouda-early ecotype (V2) respectively.

Weight of feces in grams

Table 5 and Figure 9 show the susceptibility of five ecotypes of pearl millet, according to remains of feces in any ecotype variety. The least amount of feces was 0.28 g in ecotype (V5) Bayouda late. This means that a large part of the food taken was ingested, digested and assimilated in comparison with the other four ecotypes. The different percentages of ingested food consumption of the five ecotypes, by 3rd and 4th instars of migratory locust, execrated feces ranked 0.47 g in Bayouda-early ecotype (V2), 0.66 g in Dembi ecotype (V1), 0.67 g in Wad elahow ecotype (V4) and 0.68 g in Kano ecotype (V3) respectively.

Discussion

Sudan is one of the countries whose national income relies heavily on agricultural commodities and natural resources. Pearl millet, *Pennisetum glaucum* is an agricultural commodity that represents the staple cereal of many millions of the world's poorest people in the semi-arid regions of tropical and subtropical developing countries in Asia and Africa.

Table 4: Percentage of ingested food.

Ecotype	24hrs	48hrs	72hrs	Mean
V1	95.4%	97.2%	97.1%	96.5%
V2	97.2%	95.1%	96.1%	96.1%
V3	96.3%	95.3%	98.5%	96.7%
V4	96.9%	99.5%	98.2%	98.2%
V5	98.1%	94.9%	98.8%	97.3%
Mean	96.8%	96.4%	97.8%	97%

Note: V1: Dembi-short maturing, V2: Bayouda-early maturing, V3: Kano-late maturing, V4: Wad elahow-late maturing and V5: Bayouda-cold resistant

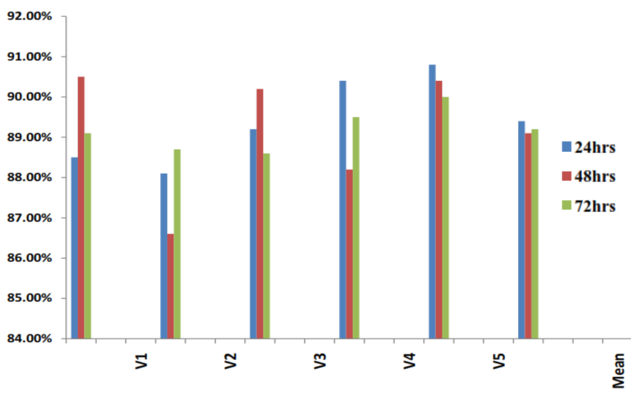

Figure 8: Percentage of ingested food. Note: V1: Dembi-short maturing, V2: Bayouda-early maturing, V3: Kano-late maturing, V4: Wad elahow-late maturing and V5: Bayouda-cold resistant.

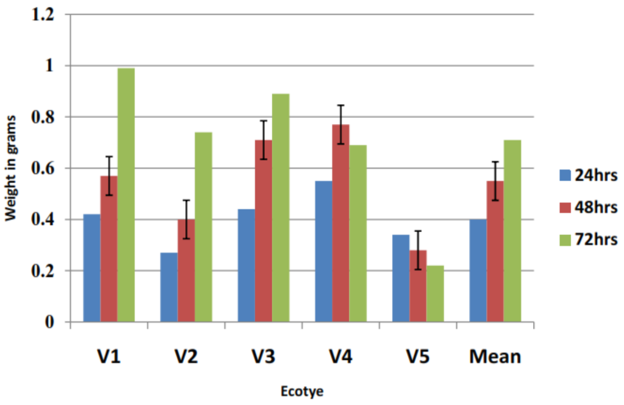

Figure 9. The weight of feces in gram. Note: V1: Dembi-short maturing, V2: Bayouda-early maturing, V3: Kano-late maturing, V4: Wad elahow-late maturing and V5: Bayouda-cold resistant

In Sudan, the pearl millet (Dukhun) is mainly grown in the western parts of Sudan (Darfur and Kordofan states) and also in eastern the region, Red Sea, Kassala, and the Gedaref States. Pearl millet in these areas is frequently attacked by locusts. Among these, it was found that the African migratory locust *Locusta migratoria migratorioides* is considered a devastating pest on pearl millet through its ability to form dense aggregations of nymphs (hoppers) and highly mobile aggregations of adults (swarms) that feed on various graminaceous crops (Sorghum and Millet). Since the migratory locust is a primary threat to these crops and resources, its outbreaks are severely followed and a prompt of control measures. So, the possibility of managing the African migratory locust on Pearl millet is by planting resistant cultivars prevailed.

The present work was conducted to investigate the susceptibility of five pearl millet varieties (ecotypes) to infestation by the 3rd and 4th nymphal instars of the species. According to the results of the study, it was evident that all five ecotype varieties of pearl millet were palatable to the migratory locust 3rd and 4th nymphal instars. These results, in general, were similar to the results of pearl millet pests inventoried by Kamal et al. (2013) who found that pearl millet was infested by many pests, such as the larvae of *Spodoptera* sp, locusts (*Schistocerca gregaria* and *Locusta migratoria migratorioides*). The results showed that there was variation in infestation rate in the five ecotypes. This finding was in agreement with Siddig et al. (2013) who stated that different varieties of pearl millet reflect varying levels of susceptibility and resistance to pests.

From the results of this study, it was noticed that the nymphs and adults of African migratory locust prefer the vegetation shoots of pearl millet to bran and grains. This result is in agreement with Sharma et al. (1996) who mentioned that nymphs and adults of the African migratory locust occasionally attack all stages of the pearl millet causing heavy damage during outbreaks but, prefer to feed on leaves, flowers, and developing grain.

According to the result in Table 1 and Figure 5, the susceptibility and food preference on the five varieties by the 3rd and 4th nymphal instars of the African migratory locust was high on Bayouda-Late maturing, Kano-late maturing, Wad elahow-late maturing, Bayouda-early maturing and Dembi-early maturing successively and in descending order. Also, these results are confirmed the amount of food intake (in grams) as in Table 2 and Figure 6.

The results on the amount of feces excreted by the hoppers in Table 5 and histogram in Figure 9 indicated that there was significant difference at 1% level, among remain of feces of any ecotype variety and application period. It was found that the percentage of taken and ingested food was 86.4%. Of the assimilated food from total ingested was 97% and only 3% was excreted in feces. These results were in line with Sharma and Davies (1988) who showed that the locust hoppers and bands are particularly devastating and invade millet in Sudan. It is also in agreement with result stated by Niassy et al. (2011) who mentioned that the late instars and adults cause economic damage directly to pasture and crops such as millet. Moreover, Latchininsky (2013) confirmed that the African migratory locust has the largest distribution among all grasshoppers and locusts but, the ecological requirements of the migratory locust are quite narrow, and the devastating damage caused by hopper bands and swarms are primarily restricted to grasses, millet, and others.

From the results in Tables 1, 2, 3 and Figures 5, 6, 7, it is noticed that the most susceptible ecotype was Bayouda-early, Bayouda-late, Wad elahow, Kano and the least susceptible was Dembi-early. In theory, Abuelgasim (1989), Sabil (1991) and Abuelgasim (2011) mentioned that early Bayouda is grown in cracked clay soil in Jebal Mara, late Bayouda is grown in cracked clay soil in Kabkabia, Wad elahow is grown in south of Nyala semi cracking clay soil, Kano is grown in cracking loamy clay soil south-west of Nyala and Dembi is grown in mixed sandy clay soil. This was confirmed by Guichard (1955), SEA (1990), who mentioned that the locust prefers to live in black cracking clay land and boundary lands of savannah, which means that its feed behavioral preferring plants grow in similar environmental areas, although there are differences in the condition. Where the varieties are cultivated of the location where the study is made, there was no difference regarding susceptibility of the five varieties to the insect pest.

In conclusion, the results showed significant difference in the susceptibility of five varieties of ecotype of pearl millet. The preference and susceptibility of ecotypes by nymphs were increasing with the increase of application time. The highest percentages of ingested food in the five ecotypes of pearl millet *Pennisetum glaucum* variety, with 3rd and ^{4th} instars of African migratory locust, was in Bayouda-late maturing. Further testing of different graminaceous species on different nymphs of African migratory locust is needed. Further research works on evaluation of food preference and susceptibility of infestations of pearl millet varieties with other pests, is recommended.

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