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Sorghum snapseed photo by Mark's Mindful Munchies, Inc.

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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]

Information from internet:

Balagadde FK, Song H, Ozaki J, Collins CH, Barnet M, Arnold FH, Quake SR, You L. 2008. A synthetic *Escherichia coli* predator-prey ecosystem. Mol Syst Biol 4: 187. www.molecularsystemsbiology.com. DOI:10.1038/msb.2008.24

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Short Communication: Profitability of beekeeping using locally made transitional top bar beehive in Wolmera Woreda, Oromia Region, Ethiopia

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Abstract. *Gobena WE. 2020. Short Communication: Profitability of beekeeping using locally made transitional top bar beehive in Wolmera Woreda, Oromia Region, Ethiopia. Asian J Agric 4: 1-4.* Beekeeping is an important source of livelihood and can be integrated with other agricultural activities. The objective of this study is to assess the profitability of beekeeping using locally built transitional top bar hives. A two-stage sampling procedure and stratified sampling technique were used in the study. Primary and secondary data were collected and analyzed using descriptive statistics and partial budgeting. The partial budgeting result reveals that beekeeping is profitable by using this hive with an incremental net benefit of 462.12 ETB. The beekeepers increased their benefits from the hive by more than 2.9-fold by using this beehive as compared to traditional hive. The study concludes beekeeping with this hive can be profitable business for the marginal farmers who have little business capital and land resource. Moreover, income from a single bee colony at beekeeper's backyard can be improved with minimum cost if this hive is used. The overall finding of this study underlined the importance of extension support and technical backing for beekeepers to use this hive.

Keywords: Beehive, beekeeping, locally made, profitability, transitional top bar

INTRODUCTION

Beekeeping plays a valuable part in improving rural livelihoods of many countries like Tanzania and Nigeria (Ajao and Oladimeji 2013; Ntalwila et al. 2017). Its success can be noted in countries like Ethiopia (Mazorodze 2015). Many studies show the importance of beekeeping from different aspects. It can be viewed as a means of combating poverty (Goldenberg 2004; Mickels-Kokwe 2006; Ogaba 2002; Lalika 2009; Ayansola 2012). Other studies have shown that beekeeping practices are an important income-generating activity, promoting employment and tourism revenue (Joni 2004; Wodajo 2011; Ajao and Oladimeji 2013; Chazovachii et. al. 2013; Qaiser et al. 2013; Wongelu 2014). It also can play major role in natural resource management and ecosystem service via pollination (Chazovachii et al. 2013; Ndegwa 2014). Several studies have shown that investment costs are relatively low, being less than 50% of income generated, making beekeeping a thriving business that can contribute invaluably to a household income (Saha 2002; Bradbear 2009; Ndegwa 2014; Wongelu 2017). Evidence from the Central Statistical Agency (CSA) shows there is an increase in the number of hives from 6.2 million in 2017 to 6.5 million hives in 2018.

Ethiopia has large apicultural resources and the potential of producing over 500,000 tones of honey per year (Ethiopian Apiculture Board [EAB] 2016]. The annual production of honey and beeswax is low compared to its potential (EAB 2016). This is due to more than 95% of our beekeepers using traditional hive management practices which affect yield. This results in traditional production system which results in low production and productivity, poor pre- and post-harvest processing and handling techniques and practices combined with poor marketing efforts have kept it part of the subsistent sector (Meaza 2010). In most cases, Ethiopian beekeepers are observed to use traditional hives, which is very difficult to manage honeybees, and to produce honey and honey products in the required quality and quantity. The maximum yield obtained from a traditional beehive so far is estimated on average to be below 7 kg /hive (Nuru 2004). However, it has been observed that more than 15kg /hive crude honey can be produced if top-bar hive is used (Nuru 2004). Locally made transitional top bar beehive is important for our farmers as it is extremely inexpensive and equally important as that of machine-made top bar hives (Melaku 2005; Wodajo 2011; Wongelu 2014; 2017). A study conducted by Wongelu (2017) showed honey yield which ranges from 10.25kg/hive/season to 37kg/hives/season harvested using this hive. Profitability of the beekeeping business is influenced by type used, ecological condition, colony strength and management practices (Tucak et al. 2004; Al-Ghamdi et al. 2017). Therefore, the main objective of this study is to assess the profitability of beekeeping using locally made transitional top bar hives within the study area.

MATERIALS AND METHODS

Study area

The study was conducted in Wolmera District, Oromia Special Zone Surrounding Finfinne, Oromia Region, Ethiopia from 2012-2013. A detailed description of study area is presented below.

Wolmera district

The Wolmera district is one of the districts in Oromia Special Zone Surrounding Finfinne, Oromia region. It is about 30 km away in West of Addis Ababa along the Ambo road at $9^0$02N and 38^0 34E. The district is split into two agro-climatic zones namely highland 61%, mid-highland 39 % (Bureau of Agriculture [BoA] 2013). Crop-livestock mixed farming system characterizes agriculture in the district. The major crops in the farming system that provide foraging resources for honeybees are fabacean, chickpea and lentils (BoA 2013). In the district, about 3,566 hives exist out of which about 1853 are traditional, 870 transitional and 843 box hives (BoA 2013).

Method of data collection

The study used both primary and secondary data sources. Primary data was collected from sample households using structured interview schedule, personal observation of sites and group discussion. Secondary data that supported primary data was collected from different sources like journals, research articles, internet, and concerned offices.

Data analysis

To perform profitability analysis, production costs for both traditional and locally made transitional beehive types were considered. The analysis was carried to estimate per hive net return from both types of hive. Based on the survey data, the costs of production and returns at the prevailing prices were used to estimate the benefits. This section aims at identifying and quantifying the different costs, which are incurred by the beekeepers in production process. Beehive, bee colony, supplementary feed, labor, transport cost, depreciation cost on beehives and interest on input costs, were the cost items that are needed to run locally made transitional top bar and traditional beehive honey production. Honey yield was the benefit of both types of beehives.

Profitability analysis of each beehive type was determined using the following formula shown below. Simple descriptive statistics farm budget techniques and Gross Return analysis frequency, percentages and tables were utilized. The farm income model is as shown:

$$NI = GR-TC$$

Where:
NI : Net Income for honey production.
GR : Gross Revenue to honey production (the revenue from honey sale)
TC : Total production cost (direct expenses and purchases for beekeeping activities).

RESULTS AND DISCUSSION

Profitability of the Hive

Honey yield is an important determinant factor in adopting the technology. To compare the performances of the locally made transitional top bar and traditional beehive yield, the cost and net returns obtained from sampled respondents were recorded and compared. The analysis was done to arrive at per hive net return from both types of beehives. As shown in Table 1, hive, bee colony, supplementary feed, labor, and transport costs were the cost items that needed to run locally made transitional and traditional beehive honey production and categorized under column one, category of cost.

Table 1. Partial budget for locally made transitional and traditional beehive

Added cost (Birr)	Costs		Revenue		
	Locally made transitional beehive	Traditional beehive	Additional return (Birr)	Locally made transitional beehive	Traditional beehive
Hive cost	45.0	4	Honey yield in pound	20.75	10.6
Colony cost	90.0	90.0	Total added return	799.85	312.65
Supplementary feed	2.76	1.91			
Labor cost	27.75	46.63			
Transport cost to market	3.87	2.74			
Interest	8.47	7.49			
Total costs of production	177.85	152.77	Total return from sell of honey	799.85	312.65

Note: Net income from locally made transitional top bar beehive (799.85-177.85=622.00 ETB); Net income from traditional beehive (312.65-152.77=159.88 ETB); Incremental net benefit of locally made transitional top bar beehive is (622.00-155.99=462.12 ETB)

Figure 1. A. Locally made transitional top bar beehive; B. laminating the hive; C. Honeybee colony established using the give; D. honey harvested from the hive

Based on the data collected from sampled beekeepers, hive cost and service life for the hives were on average 435.00 Ethiopian Birr (ETB) and 10 years for locally made transitional and 20 ETB and 5 years for traditional hive (1USD=18.2226 ETB in January 2013). The average price paid to purchase a bee colony was 450.00ETB at current market price (2013), the commercial life for the honeybee colony was assumed to be 5 years provided that this colony is not used for reproduction and renting the colony for pollination service (this is not practiced in the country). This cost was common for both types of hives. Labor cost was calculated based on hours spent in beekeeping for both types of beehives per month, summed for a year and converted to Birr which was 35.00 ETB for daily laborer. Similarly, feed cost and transport cost was calculated based on cost spent on the items divided by number of beehives and interest 5% was added on total costs.

On the other hand, honey yield was the benefit for both types of beehives and categorized under column two, return. To get the total revenue from each type of hive, honey yield obtained over the year was multiplied by selling price. In the study area, the average honey yield per annum for traditional and locally made transitional top bar beehive was 9.41 kg and 4.81 kg, respectively. It is below the national average which is 10-15 kg and 7kg respectively (Nuru 2004). The price of one-kilogram honey from locally made transitional top bar and traditional beehive was 65 and 85 birr, respectively. The price difference was due to the quality of honey harvested from the beehives.

The partial budgeting result reveals that beekeeping is profitable by using locally made transitional beehive. Table 1 also summarizes that the incremental net benefit of locally made transitional beehive 462.12 ETB. This shows that the beekeepers increased their benefit from locally made transitional beehive more than 2.9-fold compared to traditional hive. Melaku (2005) also came with similar conclusion in his study using partial budgeting analysis that timber made Kenyan top bar hive was beneficial and remunerative.

Conclusion and recommendations

In conclusion, beekeeping is potential income-generating activity with relatively low investment and operation costs. It could be an effective business for farmers who have little business capital and land resource. Income from a single bee colony at beekeeper's backyard can be improved with minimum cost if locally made transitional top bar beehive with its package used. This study underlines the importance of extension support to beekeepers in the use of this hive technology.

ACKNOWLEDGEMENTS

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Feed quality of some populations and varieties of sorghum

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Abstract. *Alane F, Rahale-Bouziane H, Fadjer Z. 2020. Feed quality of some populations and varieties of sorghum. Asian J Agric 4: 5-13.* This research aimed to characterize some local populations and those from ACSAD through two trials. The Sudan population is believed to be better than the other two local populations. There is one with an average height of 55.07 cm and an average number of tillers per square meter 128/m², that offers a green and dry yield of 5311.47 g/m² and 497.2 g/m² respectively. In the second test, the comparison of the chemical composition of the pasty milky and hard grain stages did not register a big difference. CP was almost the same (6.66% and 6.1%). The varieties Masser 17, Ezzera, and Biofuel from milky-pasty stage to hard grain stage has their cellulose rate decrease. The maturity of the seed is reached between 82 and 92 days after sowing and their sizes vary between 71 cm and 1.26 m. The average cellulose level of milky-pasty populations is 26.08% and the hard-grain stage is 28.44%. The analysis of the principal components at the hard grain stage has constituted two groups: Kherssi, Beida Adrar, Tougourte, Ain Sallah. Ezzera, Biofuel, Masser17, El Menea. Greater diversity in the chemical composition of the stems, than in those of the leaves more digested was observed.

Keywords: Biometrics, chemical composition, sorghum

INTRODUCTION

Sorghum cultivation covers 41.57 million hectares, in the tropical region from which it originates, but also in the temperate region (FAO 2001). *Sorghum* ranks are fifth in cereals, after wheat, rice, maize, and barley. But it comes second, after maize in Africa (Chantereau et al. 1997), where production reaches 18.78 million tons. The main producing countries on this continent are Nigeria (7.71 million tons) and Sudan (over 2.49 million tons). Asia produces 11.43 million tons of grain, of which 7.42 million tons in India. Globally, the United States is the leading producer with 13.61 million tons (FAO 2001).

In the United States and other developed countries in general, grain *sorghum* is reserved for animal feed. It has experienced a revival of interest with the spread of varieties without tannins, and therefore better food value. New industrial opportunities have emerged: *sorghum* fiber for paper mills and sweet sorghums for biofuel production (Chantereau et al.1997).

In 1753, Linnaeus described in Species Plantanum three species of cultivated *Sorghum: Holcus sorghum, Holcus saccaratus,* and *Holcus bicolor*. In 1805, Person suggested the name *Sorghum* for *Holcus sorghum* (L). In 1961, Clayton proposed the name of *Sorghum bicolor(L.) Moench* for cultivated *sorghum* and it is this name that is currently used (FAO 1995).

Sorghum cultivation had occupied a significant place in Algeria. Around the middle of this century (1943-47), grain *sorghum* was heavily used by local people to fight famine. The common white *sorghum* of Kabylia: these forms fall into the group of loose-tailed *sorghum* (*Sorghum vulgare L.* var.effusus Korn.) Which includes broom (*Sorghum vulgare* var technicus Korn.) and *sweet sorghum* (*Sorghum vulgare var.saccharatus* L.). Both were poorly cultivated in colonial times in Algeria; the first for its tassel and the second for its stalks and leaves which constitute an excellent forage after silage. In addition to the above varieties, *sorghum* from Sudan or *sorghum menu* (*Sorghum exiguum Forsk.*) were introduced to Algeria by Trabut (INRAA 2006).

The family of *sorghum* (Sudan grass) has stems and fine leaves. The sweet *sorghum* family has big stems (hence the name *cane*) and broad leaves. The hybrids (*Sudan grass* x *sweet sorghums*) are more or less different depending on whether the character Sudan grass or sweet sorghum prevails in the hybridization (Grenet et Cie, ND). The fineness of the stalks of the Sudan-grass allows it to be widely used: in green, in tedding (after pre-wilting of a day), or in silage.

The *sweet sorghum* whose stem size gives these plants the name "cane", has importance in presence of sugar. The range of precocity is quite broad. Hence two essential vocations, silage, and consumption in green at the barn (Grenet et Cie, ND).

In 1948, Laument spoke of the interest of forage *sorghum* as great, if neither corn can come without irrigation in fresh land (*sorghum menu*) or irrigated *sweet sorghum*. Both sweet and *sweet sorghums* are susceptible to irrigated crops of high simultaneous yields in grain and hardwood stems. The mowing is done at the formation of grains (before the risk of an accident by hydrocyanic acid). According to Borghi et al. (2013), *sorghum* is an excellent intercropping in poor soil where corn does not grow very well and provides large amounts of biomass.

Sorghum sudanense originates from Egyptian Sudan and is cultivated largely as a forage plant (*Sorghum menu*, Sudan-grass of the Americans). It is slightly slender and ranges from 2 to 3m long, with slightly hazy glabrous under the nodes (Maire et al. 1952).

Sorghum autogamy was exploited to obtain selected, short-cut, mechanically-harvested lines that quickly conquered the great plains of the south and central Midwestern states of America. Subsequently, these lines were replaced by hybrids (Chantereau et al. 1997). This improved the digestibility of *sorghum* matter, which contributes to higher productivity of livestock consuming dual-use sorghum (*Sorghum bicolor* L. Moench) (Vietor et al. 2010).

The names given by the farmers to the five Ethiopian local varieties among the 117 varieties are consistent with the Canonical Discriminant Analysis Study (CDA) and the Modeclus Cluster Analysis. Indeed, they formed dissimilar groups. The color of the median vein, the color of the grains, the size of the grains, the color of the glume, the hairiness of the glume and the shape of the grains were the morphological characters used by the farmers to name these local varieties of *sorghum*. These local varieties are to be protected.

The biometric parameters are determined of local populations and from ACSAD (Arab Center for the Studies of Arid Zones and Dry Lands, Syria) variety; in this work, we have determined the chemical composition of these same populations at two different stages: milky-pasty and hard grains of the whole plant, and organs leaves and stems separately.

MATERIALS AND METHOD

The plant material

The local seed was collected in 2012 from farmers in Algeria (Adrar wilaya) located 1330 km south of the capital. The first test was installed on 30/06/2013 at a dose of 3 seeds per pouch.

A second trial was conducted in 2015, with the introduction of eight ACSAD seed varieties (Syria). The origin, color, and weight of 1000 seeds of these populations are shown in Table 1.

The site and experimental device

The experimental device is composed of a completely random block type with three repetitions covering an area of 640,5 m². Seeding took place in open fields in a greenhouse covered by a net, at the Baraki experimental station of the National Institute of Agronomic Research of Algeria (INRAA). Located in Algeria, whose geographical coordinates are: 36 ° 41'4,35''Nord and at 3 ° 06'24,84''Est and an altitude of 18,5m. The seedling dose was 3 grams per bag. Fifteen days after germination, we left one plant per pouch. The first mowing was done 52 days after planting at the vegetative stage at an average of 40 cm.

The climate and irrigation

The climate of the region is like the Mediterranean. As it is located in the subhumid bioclimatic stage, it is characterized by a rainy season going on average from September to May, and by a sunny summer (Seltzer 1949).

Mean and maximum temperatures during the first week of the first test were 15 ° C and 23.3 ° C, respectively. The maximum rainfall recorded during the experimental period was 16 mm in October.

Mean temperatures during the first month of the second test installed on 4/05/2015 were 19.9°C. The rainfall accumulated at the climatic station during the experimental period is 245.2 mm. The maximum rainfall recorded during the experimental period was during October and November, with 91.6 mm, and 92.4 mm respectively (Table 2). The average temperature of the seven months of experimentation was 22.39°C, the maximum temperature was 27.2 recorded in August and the minimum was 19.9°C in April. The humidity of the air was high throughout the experimental period with an average of 72.13%.

Irrigation water was added to supplement the water deficit at the beginning of vegetation twice a week and at the end of vegetation once a week.

The soil

The physicochemical analysis of the soil shows a loamy clay structure (triangle of soil structure), a basic pH (7.87), and an electrical conductivity of 0.28 d/m which shows that the soil is not very salty. The nitrogen content is 0.034%. On the other hand, the phosphorus content is very high at 53.15 ppm and high for potassium 0.67 meq/g.

Table1. Origins, color, and weight of 1000 seeds of studied populations

Populations	Colors	Origins	Weight 1000 seeds
Hamra	Red	Algeria	40.16
Beïda	White	Algeria	31.13
Biofuel	White	ACSAD	40.4
Insallah	White	Algria	39.3
Ezraa	White	ACSAD	36.8
Tougourte	Black	Algeria	31.4
Masser 17	White	ACSAD	34.6
El Ménea	Black	Algeria	42.4
Dorado9	White	ACSAD	37.5
Kharsi	Red	Algeria	47

Table 2. Climatic data of the period of the test

Year Months	Precipitation (mm) Amount	Temperature (°C) Average	humidity (%) Average
2015-05	14	19.9	69.3
2015-06	14.4	23	70
2015-07	13.2	27.2	70.4
2015-08	7.2	27.3	70.9
2015-09	12.4	24	67.8
2015-10	91.6	20.6	74.5
2015-11	92.4	14.7	82
2015-12	245.20	22.39	72.13

RESULTS AND DISCUSSION

All the forage yield parameters in Table 3 show that the Sudan population is better than the other two populations. Indeed, this one with an average height of 55.07 cm and an average number of tiller per square meter 128/m², offers a green and dry yield of 5311.47 and 497.2 respectively.

In the milky-pasty stage, the chemical composition of these three populations, i.e., Hamra, Sudan, and Beida Adrar are different for the whole plant. The dry matter content is 92.5%, 94.5% and 94.85% respectively. Mean is 93.95% relatively the same in both leaf and stem organs separately (Table 4). The highest average mineral content is in the leaves (10.56% DM), the Hamra rate exceeds that of the other two populations. The average rate of mineral matter in the stems of the three populations is 5.98 %, not far from the average of the plants of the three populations (7.34%) (Table 4). The result of the ash content is greater than the average found in the organs and the straw of 194 Burkina Faso lineages, the straw offers 7% DM, the leaves 6.5% DM, and the stems 2.5% MS (Kondombo 2001).

The two other total nitrogen and crude cellulose antagonist parameters are determinants of digestibility in ruminant animals, showing a high average total leaf nitrogen content of the three populations (11.73%) (Table 5) with a higher Hamra leaf (12.92). The stems of the three populations record an average of 4.95% whereas the whole plant at this stage offers an average in all three populations of 7.66%. Burkinabé lines offer lower rates: leaves between 4.3-7%, which stems between 2-5.6% straw between 2.8-5.7% dry matter (Kondombo 2001). In Tunisia, in the milky-pasty stage, the content was 5.2% (Znaidi et al. 2010).

The level of fiber is high in the Sudan population, either in the organs or the whole plant (Tables 4 and 5), even though the population has fine stems and is therefore mainly intended for fodder production. Its pasty milky-pasty stage has 32.73% lower than the average gross yield at the same stage in Znaidi et al. (2010) 38.51%. The average of the plants of the three populations is 28.57% CF, the same average of the stems of the three populations 28.10% higher than the average of the leaves 24.85% (Table 5). The leaves of the Hamra offer a cellulose level of 23.31% lower than the rate of the other two populations. The average leaf rate in the green of this population is 28.91% and a dry leaf-to-stem ratio of 0.38% slightly lower than the Beida Adrar population ratio of 0.42% (Table 6). The average ratio of Burkinabe lineages is 0.49, the minimum is 0.25 and the maximum is 0.8 (Kondombo 2001).

The analysis of the variance of the chemical composition of the plants of the three populations, shows a very highly significant difference in the two parameters DM and CF (Table 7). On the other hand, at the stem level, the chemical composition shows a significant difference in the DM and the MM and a very significant difference in CF and none in CF (Table 9). At the leaf level, the analysis of variance is highly significant for MS and MM and very highly significant for CP and CF (Table 11).

Thus, to compare the populations, the moisture content and the crude cellulose content of the whole plant are examined. To compare the digestibility of the populations, we checked the content of CP and CF of the leaves (the leaf to stems ratio).

There was a very high positive correlation between CP and MM, a very high negative correlation between CF and MM in the whole plant of the three populations. A high negative correlation between crude cellulose and CP (Table 8). There was no correlation between the chemical parameters of leaves and stems (Tables 10 and 12).

Table 3. Green and dry yield and average height and number of talle/m² of local populations

Populations	GM g/m²	DM g/m²	Height in cm	Number of tillers/m²
Hamra	2517,19	258,53	35,97	81,33
Beida Adrar	2707,2	315,73	31,03	98,67
Sudan	5311,47	497,2	55,07	128

Table 4. Chemical composition of the *sorghum* plant of 2013-2014 at milky-pasty milk stage

Populations	In% of dry matter			
	DM%	MM%	CP%	CF%
Hamra	92.51	8.36	8.08	27.07
Sudane	94.49	6.10	7.001	32.73
Beida Adrar	94.85	7.55	7.91	25.92
Average	93.95	7.34	7.66	28.57

Table 6. Proportion of leaves and stems in the plant population and the ratio of leaves to stems

Populations	% of leaves in green	% of the stems in green	Report leaf on stems in sec
Hamra	28.91	69.42	0.38
Beida Adrar	35.32	78.037	0.42

Table 5. Chemical composition of stems and leaves of *sorghum* of companion 2013/2014 at milky-paste stage

Populations	Rods				Leaves			
	DM%	MM %DM	CP %DM	CF %DM	DM%	MM %DM	CP %DM	CF %DM
Sudan	93.74	6.29	5.28	31.16	92.89	10.50	10.93	26.57
Beida Adrar	93.36	5.63	4.70	26.38	93.36	9.67	11.35	24.67
Hamra	93.32	6.03	4.88	26.77	92.97	11.51	12.92	23.31
Average	93.47	5.98	4.95	28.10	93.08	10.56	11.73	24.85

Table 7. Analysis of the variance of the chemical parameters of the plants of the three local populations at the milky-pasty stage.

	Variance	DDL	Chi2	P	Meaning
MS%	3.507	8	28. 056	0.0009	***
MM%	1.360	8	10.883	0.4169	NM
MAT%	0.702	8	5.613	0.6191	NM
CB%	16.761	8	134.091	0<0.0001	***

NM: no significant ; *: significant; ***: very highly significant

Table 8. Correlation matrix of the chemical parameters of the plants of the three local populations at the milky-pasty stage

	MS%	MM%	MAT%	CB%
MS%	1			
MM%	-0. 164	1		
MAT%	-0.280	0.826	1	
CB%	-0.243	-0.882	-0.744	1

Dd1 = 9-2 = 7 α = 0.1 rth = 0.58 (*). α = 0.05 rth = 0.66 (**). α = 0.01 rth = 0.79 (***)

Table 9. Analysis of the variance of the chemical parameters of the stems of the three local populations at the milky-pasty stage

	Variance	DDL	Chi2	P	Meaning
MS%	0.249	8	1.994	0.0376	*
MM%	0.260	8	2.044	0.0429	*
MAT%	0.404	8	3.229	0.1617	NM
CB%	22.430	8	179.442	0<0.0001	***

NM: no significant ; *: significant; ***: very highly significant

Table 10. Correlation matrix of the chemical parameters of the stems of the three local populations at the milky-pasty stage

	MS%	MM%	MAT%	CB%
MS%	1			
MM%	-0. 512	1		
MAT%	0.012	-0.131	1	
CB%	-0.404	0.294	0.429	1

Dd1 = 9-2 = 7 α = 0.1 rth = 0.58 (*). α = 0.05 rth = 0.66 (**). α = 0.01 rth = 0.79 (***)

Table 11. Analysis of the variance of the chemical parameters of the leaves of the three local populations in the milky-pasty stage

	Variance	DDL	Chi2	P	Meaning
MS%	0.118	8	0.948	0.0029	**
MM%	3.259	8	26.070	0.0020	**
MAT%	5.379	8	43.033	0<0.0001	***
CB%	4.276	8	34.209	0<0.0001	***

*: significant; ** : very significant. ***: very highly significant

Table 12. Correlation matrix of the chemical parameters of the leaves of the three local populations in the milky-pasty stage

	MS%	MM%	MAT%	CB%
MS%	1			
MM%	-0. 182	1		
MAT%	0.385	0.131	1	
CB%	0.005	0.375	-0.191	1

Dd1 = 9-2 = 7 α = 0.1 rth = 0.58 (*), α = 0.05 rth = 0.66 (**), α = 0.01 rth = 0.79 (***)

In the second trial of 2014/2015, in addition to the three populations of the first trial, we added ten populations (local) and varieties (ACSAD). Two physiological stages were to be analyzed: the milky-pasty stage and the hard grains stage. The comparison of the chemical composition of these two stages did not register a big difference. In fact, the average level of CP was almost the same at both stages (6.656% and 6.089%), with a slight increase in the other chemical parameters of the milky-pasty stage at the hard grain stage (Table 13).

In addition, some varieties and populations did not follow this progression. Since the varieties, Masser 17, Ezzera, and Biofuel of the milky-pasty stage at the hard grain stage, their cellulose content began to decrease (Table 13). This can be explained by the decrease in the bark-to-marrow ratio of their stems (Table 14), so at the hard-grain stage, there was a synthesis of parenchymal tissues at the central level of the stem. Also, these varieties are characterized by a short cycle and a short size. The maturity of the seed is reached between 82 and 92 days after sowing and their sizes vary between 71cm and 1.26m (Table 14). The average cellulose content of milky-pasty populations was 26.08% and the hard-grain stage was 28.44% (Table 14), which is lower than the values given by the Jarrige tables (1988) at the milky-pasty stage. 30.7% CF and 6.9% CP.

The analysis of the variance shows a highly significant difference between populations and varieties for the DM parameter and a very highly significant difference between the CP and CF variables (Table 15).

Table 16 shows that milky-pasty DM is very highly negatively correlated with CP and highly positively correlated with MM and CB. The two chemical parameters MM and CF are very highly correlated.

Table 17 shows a correlation at the very high positive hard grain stage between CF and MM, very highly negative between CF and CP The correlation between MM and DM is highly positive.

The average content or the highest requirements for minerals in the milky-pasty stage are marked in the Tougourte population and the lowest is in El Menea, Biofuel, and Ezzera (Figure 1). While the needs are high at the hard grain stage at El Menea, Tougourte, Kharssi, and Beida Adrar. The first two populations are hybrids with fine stems <2mm and high height of about 1.5m (Table 14), easy to fade select for forage production. The lowest need for MM is recorded in biofuel (Figure 2).

The highest CP content in the two stages was studied. was marked in the Biofuel variety (9.1%) at the milky-pasty stage and 7.03% at the hard grains stage (Figures 3-4, Table 13). The lowest was at Ain sallah at the milky-pasty stage, and Ain sallah and Kherssi at the hard grain stage. They are two populations that are exploited mainly to produce seeds at the level of Oases and arid zones. The population of Ain sallah is taller and with big stems (1.72cm), with broad leaves and a very long cycle of 128.5 days, its panicles are large and compact. After harvesting the plant, it can feed the animals with a supply of nitrogen concentrate. The population Kherssi, is shorter than Ain Sallah, but richer in nitrogen at the milky-pasty stage, at the hard grain stage it produces vegetables, but it is characterized by the presence of the pigments that oasis women use as dyeing.

In the milky-pasty stage (Figure 5), the highest crude fiber content is found in the Tougourte population and the lowest in the El Menéa population respectively 30.59%, 21.57% (Table 13). 30% of content is good (Jarrige 1988, 1995).

Table 13. Chemical composition of ten populations and varieties of sorghum at milky-pasty and hard grain stages

Populations and varieties	Milky-pasty stage in %DM				Hard grain stage in %DM			
	DM%	MM %	CP%	CF %	DM%	MM%	CP%	CF
AinSallah	93.751	6.978	4.133	27.67	94.476	7.776	4.712	32.07
Khersi	93.575	7.245	8.121	26.28	92.738	8.930	3.886	32.69
Masser17	93.400	7.135	8.652	27.38	94.437	8.143	7.113	24.53
Ezzera	92.696	6.264	8.583	23.62	94.310	7.906	7.576	21.78
Tougourte	93.974	8.868	6.289	30.59	94.475	9.019	4.956	32.90
Biofiuel	92.904	6.692	9.089	24.67	94.759	7.641	7.027	21.39
El Menea	93.474	6.094	6.085	21.57	94.626	9.341	8.082	32.33
Beida Adrar	93.190	7.131	5.563	26.43	94.511	8.288	5.357	29.88
Dorado9	94.638	7.178	3.388	26.52	-	-	-	-
Average	93.511	7.065	6.656	26.08	94.291	8.381	6.089	28.444

Table 14. Biometric parameters of ten populations and varieties of sorghum at milky-pasty and hard grain stages

Populations and varieties	Milky-pasty stage					Hard grain stage	
	Cycle (day)	Height (cm or m)	Diameter of stems(mm or cm)	Bark /Moelle ratio (sec.)	The report sheets/stems	Cycle (day)	Bark/Moelle ratio (sec.)
In Sallah	60	1.4667m	1.72 cm	2.84	0.29	128.5	1.560
Khersi	35	1.2975m	12.35mm	2.50	0.5	111.33	4.125
Masser17	35	1.2225m	11.275mm	3.92	0.24	92.33	2.800
Ezzera	35	1.2575m	10.385mm	3.63	0.23	85.33	2.475
Tougourte	39	1.43m	1.77mm	3.75	0.30	106	1.980
Biofuel	35	70.67cm	18.63mm	2.53	-	82	2.270
El Ménéa	60	1.5367m	1.525mm	5.25	0.2	105.67	3.33
Beida Adrar	35	1.3525m	17.2mm	3.13	0.3	81	1.60
Dorado 9	60	96.33cm	2.22mm	2.95	0.4	106	2.32
Hamra	-	-	-	-	-	115	1.48

Table 15. Variance analysis of the ten milky-pasty populations and varieties of *sorghum*

	Variance	DDL	Chi2	P	Meaning
MS%	0.373	26	9.701	0.0031	**
MM%	0.675	26	17.557	0.2174	NM
MAT%	4.491	26	116.459	0<0.0001	***
CB%	8.527	26	221.706	0<0.0001	***

NM: no significant; **: very significant. ***: very highly significant

Table 16. Correlation matrix of the ten milky-pasty populations and varieties of *sorghum*

	DM%	MM%	CP%	CF%
DM%	1			
MM%	0.428**	1		
CP%	-0.658***	-0.085	1	
CF%	0.358**	0.721***	-0.179	1

Ddl=15-2=25 α=0.1 rth=0.323 (*), α=0.05 rth=0.381 (**), α=0.01 rth=0.487(***)

Table 17. Correlation matrix of ten populations and varieties of hard grain *sorghum*

	DM%	MM%	CP%	CF%
DM%	1			
MM%	0.467**	1		
CP%	0.198	-0.157	1	
CF%	0.110	0.523***	-0.542***	1

Ddl=24-2=22 α=0.1 rth=0.344 (*), α=0.05 rth=0.404 (**), α=0.01 rth=0.515(***)

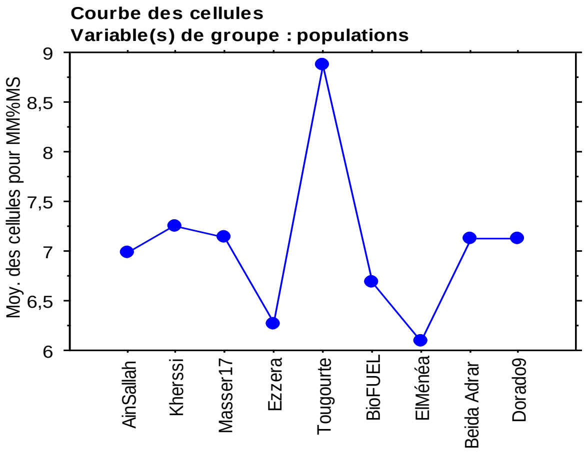


Figure 1. The average content of MM populations and varieties at milky-pasty stage

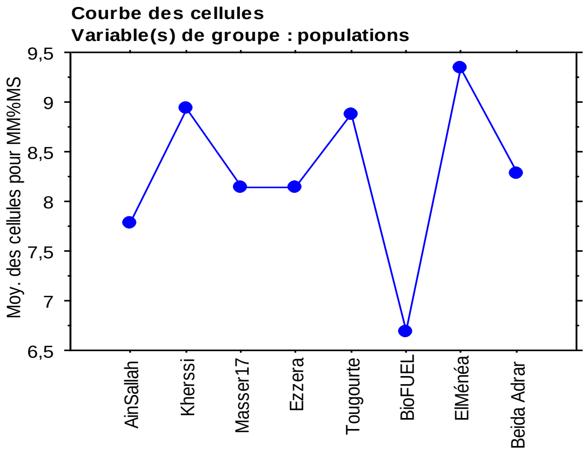


Figure 2. The average grade of MM populations and varieties at hard grain stage

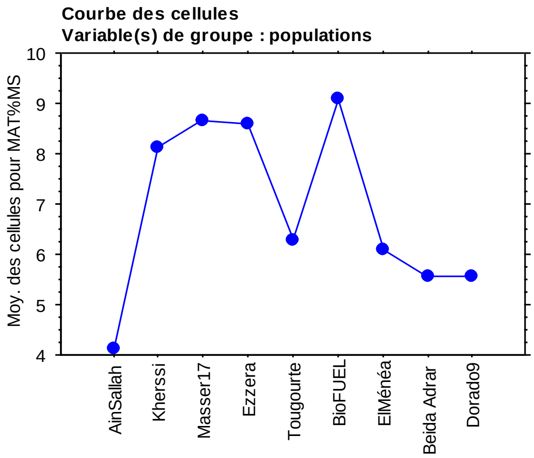


Figure 3. Population and variety counts in CP at milky-pasty stage

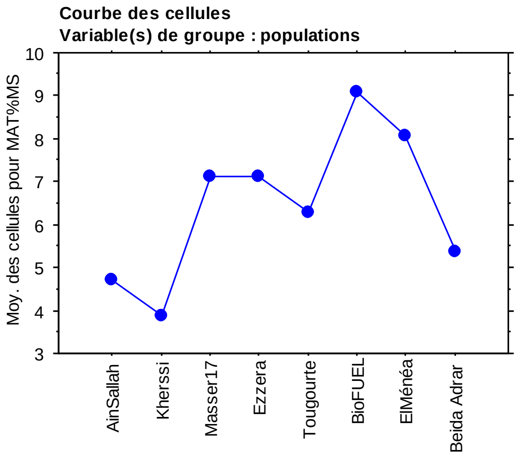


Figure 4. Population and variety counts in CP at hard grain stage

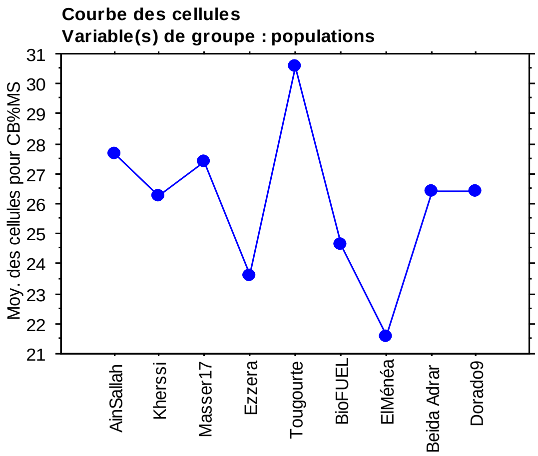


Figure 5. Populations and varieties counts in CF at milky-pasty stage

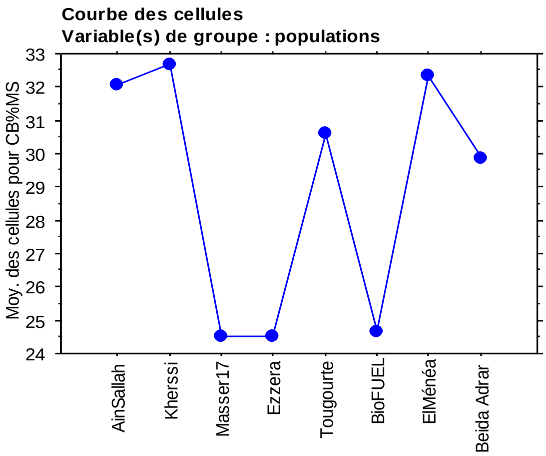


Figure 6. Populations and varieties counts in CF at hard grain stage

At the hard grain stage, according to Figure 6, the populations Ain Sallah, Kherssi, El Menea, and Tougourte recorded high levels in crude cellulose, but did not exceed 33%. All these populations are characterized by a long cycle greater than 100 days and a high to medium height, this rate of fiber is necessary to resist the lodging and the weight of the panicles.

In the analysis of the principal constituents at the milky-pasty stage and according to the Kaiser criterion (Figure 7), four axes can be chosen. In the elbow criterion, a significant fall is observed from the first axis (from 42.3% to 6.01% of the inertia). In the foreground (Figure 8) all the parameters are close to the circle, so they are indeed well correlated with the two factors constituting this plane (F1 and F2) except the parameter height which approaches the center it tends to cancel itself. The first factorial axis (1x2) gives (42,3% + 28,37% = 70,67%), the axis according to which is preserved, by the projection of the maximum of the initial dispersion of the points of the cloud. All variables occupy a restricted area within the correlation circle. The maximum angle between the two variables is less than 90 °. This suggests that all variables are positively correlated with each other.

The parameters, cycle in the day, MS%, ratio bark on the marrow, MM%, CF%, and height of the stems occupy the same plane and include the populations and varieties, El Menea, Tougourte, Dorado9, Ain Sallah. The opposite plane is occupied by the parameters CP %, and diameter of the stems group the rest of the populations which are Ezzera, Biofuel, Masser17, Beida Adrar, Kherssi.

At the hard grain stage, the Kaiser Criterion (Figure 9) can retain 3 axes. In the elbow criterion, there is a significant drop from the first axis (from 46.68% to 20.96% of the inertia). In the foreground (Figure 10), all the parameters are close to the circle, so they are indeed well correlated with the two factors constituting this plane (F1 and F2). The first factorial axis (1x2) gives (46.68% + 27.9% = 74.58%), is the axis according to which is preserved, by the projection of the maximum of the initial dispersion of the points of the cloud. All variables occupy a restricted area within the correlation circle. The maximum angle between the two variables is less than 90°. This suggests that all variables are positively correlated with each other**.** The parameters, cycle in the day, ratio bark on the marrow, MM%, CF% occupy the same plan and include populations and varieties, Kherssi, Beida Adrar, Tougourte, Ain Sallah. The opposite plane is occupied by the parameters CP%, and DM% regroup the rest of the populations which are Ezzera, Biofuel, Masser17, El Menea.

The results showed a greater diversity in the chemical composition of the stems than in those of the leaves. This diversity is much greater with the total walls than with the other variables. The average total wall (CF) content is lower in leaves than in stems and whole straw. This variation is explained by the fact that the leaves are richer in chlorophyllin parenchyma (hemicelluloses) which are more digestible walls, whereas the stems are rich in fibrous and little digestible support tissues. In addition, the varieties Masser 17, Ezzera, and Biofuel from milky-pasty stage to hard grain stage their cellulose rate to decrease. This can be explained by the decrease of the bark-to-marrow ratio of their stems, so at the hard grain stage, there has been a synthesis of parenchymal tissues at the central level of the stem. Also, these varieties are characterized by a short cycle and a short size. But for other populations (short and long cycles) there is an increase in the wall rate. The same result was obtained by Kondombo (2001) who noticed that the wall contents are higher in the early lines than in the later ones. Therefore, the authors who say that beyond the heading stage, the chemical composition, especially the cellulose content evolves (Andrieu and Weiss 1981, Demarquilly and Andreieu 1987) is not true in our case in all populations.

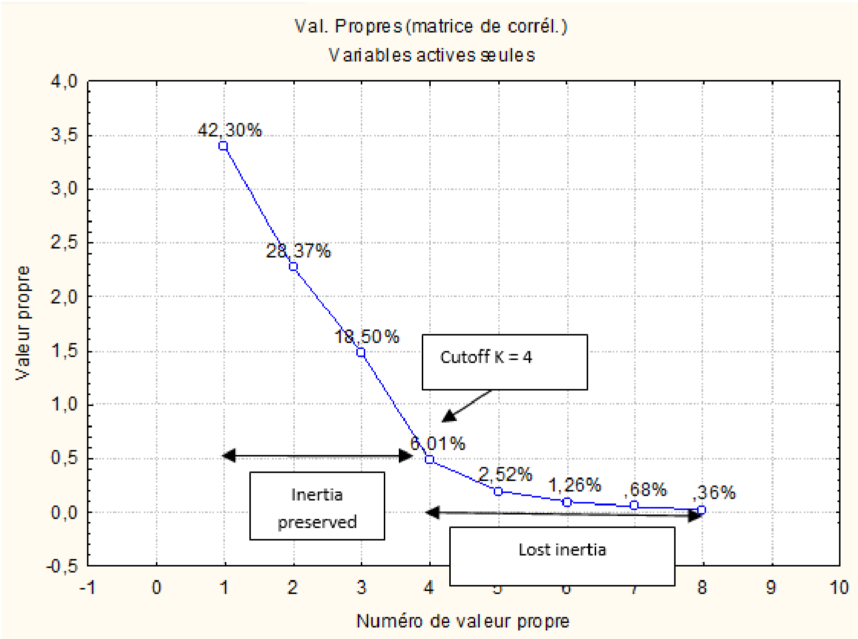


Figure 7. Graphical presentation of eigenvalues according to the Kaiser Criterion milky-pasty stage

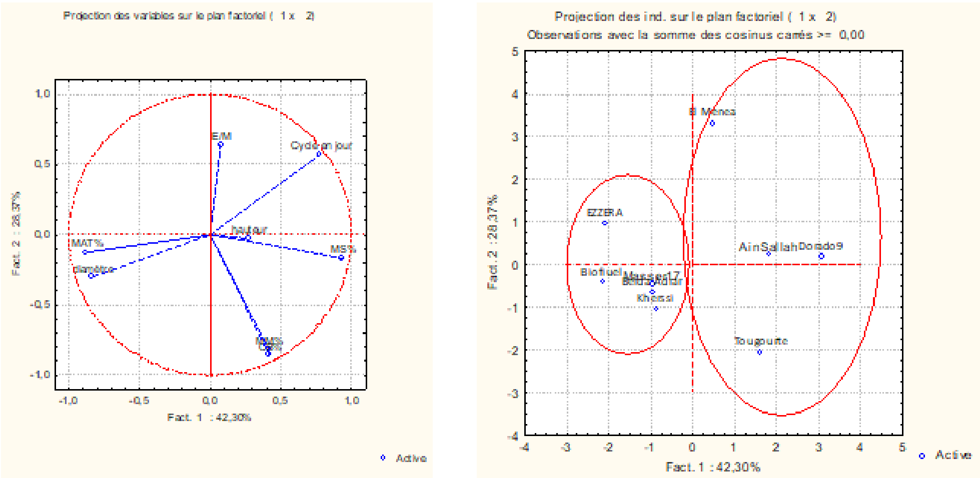
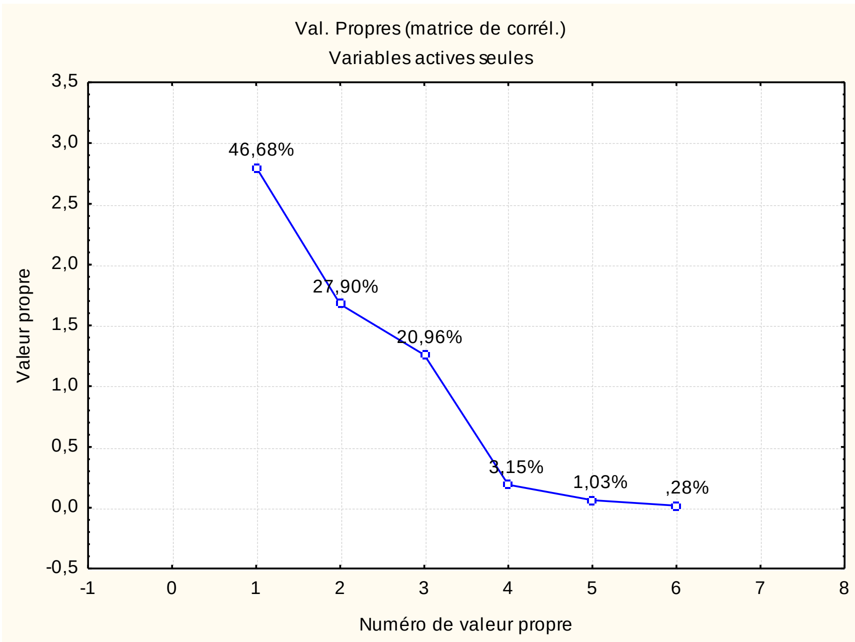


Figure 8. Analysis of the main components of the populations and varieties of sorghum studied according to plan1 at the milky-pasty stage



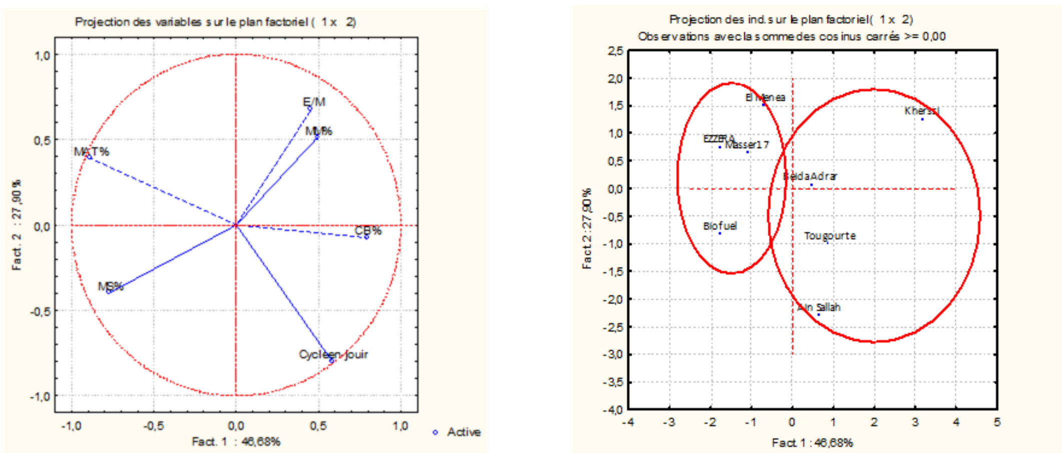


Figure 10. Analysis of the main components of the populations and varieties of *sorghum* studied according to plan1 at the hard grain stage

In this test, the average total nitrogenous matter (TMA) and total mineral contents are on average higher in the leaves than in the whole stems and straws. As for the whole straw, the different contents of the chemical composition and the values of digestibility, follow the evolution of those of the stems, because of their greater contribution to the total dry matter.

For the foraging aspect of the study, the analysis of the result leaves, stems, and straws whole, allowed to highlight the relations that exist between the main constituents and their influence on the food value of the straws. This analysis showed that many characters segregate independently. The use of fodder by animals is faced with the sorting problem, explaining that often the ingested values are higher than those distributed. The best populations and varieties for the content of CP, were: Khersi, Masser17, Ezzera, Biofuel at the milky-pasty stage. With El Menea, Ezzera, Biofuel, and Maser17 at the hard grain stage being the best for the content of CP. The low levels of cellulose in the milky-pasty stage are present in El Menea, Ezzera, and Biofuel whereas at the hard grain stage the low levels are at Biofuel, Ezzera, and Masser17.

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Phenotypic screening of cowpea (*Vigna unguiculata*) genotypes in response to parasitic weed *Alectra vogelii*

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Abstract. *Ugbaa M, Omoigui L, Bello L. 2020. Phenotypic screening of cowpea (*Vigna unguiculata*) genotypes in response to parasitic weed* Alectra vogelii. *Asian J Agric 4: 14-17.* Cowpea (*Vigna unguiculata* (L.) Walp) production is constrained by several abiotic and biotic factors. Among the biotic constraints, the parasitic flowering plant *Alectra vogelii* Benth. is one of the most formidable limitations in the dry savannas of West and Central Africa, a region that accounts for over 64% of world production. *Alectra* causes yield losses estimated between 41 and 100% in susceptible cultivars. Several control measures have been suggested for the control of the parasite. These include cultural practices, application of ethylene chemicals, and host plant resistance. Among these control measures, the use of resistance cultivars appears to be the most attractive option to the resource poor farmers in sub-Saharan Africa. This study was designed to revalidate and determine the reaction status of some improved and local cowpea genotypes to *Alectra vogelii*. Two screening experiments using pot culture technique and arranged in a Completely Randomized Design, were carried out in the screen house. Pot culture was comprised of sand and topsoil mixed in a 2:1 ratio and inoculum of 5000 *Alectra vogelii* seeds. To enhance effective parasite seed germination, the pot culture was watered twice a day for seven (7) days before planting of test cowpea genotypes. At 30 days after planting (DAP) *A. vogelii* shoots emerged from pots planted to susceptible cowpea genotypes, although some had delayed emergence up to 40 DAP. Susceptible cowpea showed leaf chlorosis, stunted growth, and partial leaf senescence. Some developed symptoms but *A. vogelii* shoots did not emerge. In both experiments, cowpea genotypes of B301, IT98K-573-1-1 and IT98K-205-8 were consistently resistant. They showed no attachment or emergence of the parasite. The absence of attachment on resistant cowpea genotypes suggests hyperactive mechanism of resistance to *A. vogelii*. This is a localized necrotic response that killed off attached parasite at the point of contact, a form of programmed cell death (PCD). This response strongly indicates dominant action in the resistance to *A. vogelii* in these genotypes. Resistant genotypes can be used as sources of resistance genes to develop improved resistant cowpea varieties.

Keywords: *Alectra*, cowpea, pot culture, resistance, screening

INTRODUCTION

Cowpea (*Vigna unguiculata* (L.) Walp) is an important food legume grown in tropical and subtropical regions of the world, primarily in sub-Saharan Africa. The largest areas under cultivation are in the savannas of West and Central Africa (WECA) where cowpea is an important crop. Cowpea is grown on about 12, 577, 845 million ha worldwide, with an annual grain production of about 7, 407, 924 tons (FAO 2017). Nigeria is the largest cowpea producer in the world and accounts for over 3.4 million tons of grain production from an estimated 3.7 million ha land area (FAO 2017). Nigeria is also the largest consumer of cowpea (FAO 2017) with consumption per capita is about 25-30 kg annually (FAO 2010).

Cowpea forms a major staple in the diet in Africa and Asian continents and is an important source of nutritious food and fodder in West Africa (Awe 2008). Cowpea contains 23-35% protein in its grains, about twice the protein content of most cereals, and is a rich source of B-vitamins, 62% soluble carbohydrates, small amounts of other nutrients, and relatively free from anti-nutritive factors (Brader 2002). Their amino acid also complements those of cereals (Asumugha 2002). Islam et al. (2006) emphasized that all parts of the plant that are used as food are nutritious, providing both protein and vitamins. Immature pods and peas are used as vegetables while several snacks and main dishes are prepared from the grains (Bittenbender et al. 1984). The use of cowpea haulms as fodder is attractive in mixed crop/livestock systems where both grain and fodder can be obtained from the same crop (Tarawali et al. 1997). The crop is also a valuable and dependable commodity that produces income for many smallholder farmers and traders in sub-Saharan Africa (Langyintuo 2003).

Despite the importance of cowpea, yield on farmers' field is still low and significant national production deficit exists due to a variety of abiotic and biotic stresses that constrain its production and productivity. These constraints include insect pests, diseases, parasitic weeds nematodes, drought, and heat (Omoigui et al. 2007). One of the serious biological constraints of increasing cowpea productivity in semi-arid regions of West and Central Africa (WECA) is the threat of being attacked by parasitic weeds *Strigages nerioides* and *Alectra vogelii* (Ehlers and Hall 1997). Fields infested by these parasitic weeds are difficult to clean due to considerable number of seeds produced and the dormancy mechanisms, which enable seeds to survive in the soil for several years. *A. vogelii* for instance, can produce as many as 600,000 seeds per plant (SP-IPM 2003). In an infested field, up to 75% of the crop damage occurs underground before shoots emerge above ground

(Emechebe and Singh 1989). According to Lagoke et al. (1991), several cultivated lands have been abandoned due to high infestations with these noxious parasitic weeds. Although attacks by *Alectra* are less severe than that of *Striga*, total yield loss is not uncommon in fields heavily infested. Yield loss resulting from *A. vogelii* attack ranges from 41-100% on susceptible cultivars (Lagoke et al. 1997). Development and use of cowpea varieties resistant to *A. vogelii* are one of the viable options to curb crop losses caused by this parasite. But equally important to prevent the spread of the parasite to new regions and be affordable for poor farmers and small-holder farming systems prevalent in Africa. Resistant varieties are particularly needed in the northern Guinea savannah zone of Nigeria where *A. vogelii* has become a major challenge in cowpea production (Mangani et al. 2008). Therefore, this study was designed to phenotype selected cowpea genotypes for their response to *A. vogelii* infestation.

MATERIALS AND METHODS

Planting materials/inoculum source

The plant materials used in this study were obtained from the International Institute of Tropical Agriculture (IITA), Kano Station, and the Molecular Biology Laboratory of the University of Agriculture, Makurdi, Nigeria. The eleven cowpea genotypes and their resistance/susceptibility phenotypes previously reported are presented in Table 1. IT03K-338-1 and IT98K-205-8 are advanced breeding lines developed at IITA earlier reported having resistance to *Striga* and *Alectra*. IT97K-499-35 and IT98K-573-1-1 are improved varieties developed also by IITA. B301 is a landrace from Botswana previously reported to be resistant to *Striga* and *Alectra*. Borno Brown, BOSADP brown, Golam white, Banjar, Kanannando Brown, and Yamisra are local cultivars grown in the Northeast, and previously reported to be susceptible to *Alectra*. Seeds of the parasitic weed *Alectra vogelii* Benth. used in this research were obtained from Borno State, through the Molecular Biology Laboratory of the University of Agriculture, Makurdi, Nigeria.

Experimental location and design

The plant materials were screened to re-validate their response to *Alectra vogelii*. Plants were grown with the pot-culture technique. Plastic pots with size of 15 cm in diameter and 14 cm in length containing about 1.8 kg of soil were used. Each pot was filled with a mixture of top-soil and sand in a ratio of 2:1 and inoculated with five thousand *Alectra vogelii* seeds. The experiment was carried out in the screen house of the Molecular Biology Laboratory, Department of Plant Breeding and Seed Science, University of Agriculture Makurdi (Lat 7.8°N, Long 8.6° E, and Alt 115 m above sea level). The experiment was laid out in Completely Randomized Design (CRD) with three replications. Each cowpea genotype was planted to two (2) pots in each replication. 5000 *Alectra* seeds were inoculated into each pot. Estimation of 5000 *Alectra* seeds was done as shown below.

Estimation of *Alectra* seeds

One thousand (1000) *Alectra* seeds were counted using a compound microscope (Brunel Microscope 0723219. X400 magnification) and weighed with a digital sensitive balance (Denver Instruments TP-3002). The weight of 5000 *Alectra* seeds was extrapolated as shown below.

Weight of 1000 seeds = 0.125g

Therefore, weight of 5000 seeds = 0.125g × 5 = 0.625g

Inoculated pot culture was conditioned prior to plant the test materials by watering for seven days to enhance germination of *Alectra* seeds.

Cultural practices

Three cowpea seeds were sown per pot at a depth of 2 cm and later thinned to two at 14 days after planting (DAP). One gram of NPK (15:15:15) fertilizer was applied per pot at 7 DAP at a rate of 15kg ai/ha. Weeds were hand-pulled while still tender.

Data collection and scoring

At about 30 DAP, pots were examined for *Alectra* shoot emergence. Thereafter, *Alectra* shoot count was done on a daily basis until the experiment finished at 65 DAP. At the end of experiment, the soil was washed off the plant root after submerging each pot in a 20-L bucket of water for about 5 min. The roots of each plant were gently separated and carefully freed from any remaining soil and examined for *Alectra* tubercle attachment. Plants allowing attachment and emergence of *Alectra* were classified as susceptible. Those without any attachment and free of infestation were categorized as resistant.

Table 1. Plant materials used and their resistance response to *Alectra*.

Cowpea cultivars/landraces/lines	Pedigree	Response to *Alectra*
IT03K-338-1	IT87D-941-1 × IT90K-59	R
IT97K-499-35	IT93K-596-9-12 × IT93K-2046-1	MR
IT98K-573-1-1	IT93K-596-9-12 × IT86D-880	R
IT98K-205-8	IT93K-596-9-12 × IT93-2046-1	R
B301	Landrace from Botswana	R
Borno Brown	Commercial cultivar in Borno	S
Golam white	Commercial cultivar in Borno	S
BOSADP Brown	Commercial cultivar in Borno	S
Banjar	Commercial cultivar in Borno	S
Kanannado Brown	Commercial cultivar in Borno	S
Yamisra	Commercial cultivar in Borno	S

Note: S: susceptible, R: resistance, MR: moderately resistant (Omoigui et al. (2012)

RESULTS AND DISCUSSION

At about 30 DAP, *A. vogelii* started emerging from the susceptible cowpea plants. While some genotypes had delayed emergence until 8 weeks. The susceptible cowpea plants showed leaf chlorosis, stunted growth, and partial leaf senescence. Some plants developed the symptom but *A. vogelii* did not emerge from the soil.

In both experiments, the cowpea cultivars: B301, IT98K-573-1-1, and IT98K-205-8 were consistently resistant to *A. vogelii* (Table 2). These cowpea lines showed no attachment or emergence of the parasite and were classified as resistance while the remaining cowpea lines either demonstrated attachment or emerged *Alectra* and were classified as susceptible (Figure 1).

It has been shown by researchers that studies on parasitic weeds and their interactions with host plants can be successfully carried out in screen house using pot trials. For example, screening for varietal resistance, conditioning trials, nutritional interrelationships of host and parasite, effect of herbicides, germination stimulants, and other aspects have been approached effectively using pot and buried seed studies (Sand et al. 1990). One of the major advantages of pot trials is that they can be carried out year-round, while field experimentation is limited to one cycle per year. In cowpea, pot screening using pot culture inoculated with parasite seeds has been successfully employed in screening for varietal resistance to *Striga* (Omoigui et al. 2012) and *Alectra* (Kureh et al. 1994; Magani et al. 2008).

In the present study, pot screening of cowpea under screen house condition using pot mixture of topsoil and sand (2:1 vol/vol) with inoculum of 5000 *Alectra vogelii* seeds proved a reliable method for assaying resistance to the parasitic weed. Magani et al. (2008) who reported similar success also stated that chances of success improve with increased density of parasite seeds in pot mixture.

This study showed that cowpea genotypes of B301, IT98K-573-1-1, and IT98K-205-8 were resistant to *Alectra vogelii* as they supported no attachment of the parasite tubercle across two experiments. This result confirms the work of previous researchers who also reported resistance of B301 (Atokple et al. 1995; Omoigui et al. 2012) and IT98K-573-1-1 and IT98K-205-8 (Omoigui et al. 2012) to *Alectra vogelii.*

Table 2. Response of cowpea cultivars to *Alectra vogelii* averaged across two (2) experiments

Cowpea genotype	No. of *Alectra* count/plant	Response
Borno Brown	11	S
BOSADP Brown	12	S
Golam white	8	S
Banjar	12	S
Kanannado brown	10	S
Yamisra	11	S
IT03K-338-1	1	MR
B301	0	R
IT97K-499-35	1	MR
IT98K-573-1-1	0	R
IT98K 205-8	0	R

Note: R: Resistant, S: Susceptible, MR: Moderately resistant

Figure 1. Chlorosis and partial leaf senescence of susceptible cowpea plant due to Alectra infestation

Figure 2. A. *Alectra vogelii* infected plants showing numerous hairy attachments. B. *Alectra vogelii* shoots and attachments on susceptible plant exposed after washing away the pot culture.

The absence of attachment on the cowpea genotype suggests an active mechanism of resistance that prevented successful attachment of parasite. This response seems to be a localized necrotic response that killed off attached parasites at the point of contact (hyperactive response, HR), a form of programmed cell death (PCD) in the host (Zhang and Zaitlin 2008). This implies that resistant cowpea cultivars did not allow the parasite haustorium to penetrate and establish connection with its vascular system to derive nutrients for its survival. This response strongly indicates dominance interaction in the resistance to *Alectra* in these genotypes. Berner et al. (1993) stated that the mechanism of resistance of cowpea to *Alectra* appears to be post-attachment mortality of the parasite with no reduction in parasite seed germination. Borg (1999) also considers that low induction of germination of parasite seeds, which seems to be the resistance response to parasitic plants in some other crops, plays a little role in resistance of *Orobanche* in legumes. In *Nicotiana langsdorffii*, one of two species of *Nicotiana* known to express an incompatible interaction with the oomycete *Perono sporatabacina* (the causal agent of tobacco blue mold disease), Zhang and Zaitlin (2008) showed that incompatibility was due to the hypersensitive response (HR), and plants expressing the HR are resistant to *P. tabacina* at all stages of growth. Resistance was due to a single dominant gene in *N. langsdorffii* accession S-4-4 named *NlRPT*.

The resistance of B301, IT98K-573-1-1, and IT98K-205-8 to *Alectra vogelii* also implies that these genotypes can be cultivated in Borno State where the *Alectra vogelii* seeds used in this work were obtained. Omoigui et al. (2012), who conducted a two-year field screening in this region, stated that even though some of the genotypes, especially B301, were low yielding the genotypes would be useful sources of cowpea *Alectra* resistance genes for incorporation into high yielding and adapted cultivars for host-specific race in the region. The use of resistant cultivar integrated with other cultural practices appears to be the only viable management option for effective control of *Alectra* infestation on cowpea.

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The growth and yield performance of true shallot seed production in Central Sulawesi, Indonesia

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Abstract. *Saidah, Wahyuni AN, Muchtar, Padang IS, Sutardi. 2020. The growth and yield performance of true shallot seed production in Central Sulawesi, Indonesia. Asian J Agric 4: 18-22.* Shallot (*Allium ascalonicum* L.) is a vegetable commodity that has high economic value and market prospects. So far, planting shallots using seeds from tubers, but planting onions with seeds is better and more cost efficient, with the seeds being healthier. The aim of this study was to determine the growth and yield performance of true shallot seed production in Central Sulawesi, Indonesia. The study was conducted from May to September 2018 in Wuasa Village, North Lore Sub-district, Poso District at an altitude of 1083.7 meters above sea level. Research with an on-farm research approach on farmer's land as a cooperator covering an area of 3000 m2. The study used the Probability Sampling Method with 10 replications. The results showed that the growth and yield performance of true shallot seed in Central Sulawesi was quite high. This is due to the suitability of the climatic conditions and cultivation technology that has been applied. In the yield component, the percentage of flowering plants was quite high at 78% with several capsules/flower at 80.67. The success of true shallot seed production is expected to be able to replace the source of seed from the tuber, which may contain viruses and diseases carried by the tuber seeds.

Keywords: Growth, shallot, true shallot seed, yield

INTRODUCTION

Shallot is one of the leading national horticultural commodities that is widely cultivated by farmers because of the good market prospects with high selling prices. Many attempts have been made to support the development of shallot production. One of them is the use of good quality seeds. Seed is one of the aspects that has an important role in supporting the success of plant cultivation. The use of good quality seeds is an initial step that plays an important role in supporting increased production and productivity of shallots.

The seeds used in shallot cultivation can be derived from tuber and seed of shallot. In general, the seeds used by farmers in shallot cultivation derived from the tubers of consumption from the previous planting season. Other than that, according to Maemunah and Nurhayati (2012), the farmers do not carry out the seed selection stage, and the use of tubers is carried out continuously from previous production, so that it can affect the degradation of shallot tuber production, both in terms of quality and quantity. The use of consumption tubers as seeds is done because of the lack of availability of quality seeds around the farmer's location. The farmers must bring seeds from seed production centers which require farmers to incur additional costs for distribution costs that are quite high due to the large volume of tubers to be distributed. One alternative to meet the needs of good quality seeds with relatively lower costs is the use of True Seed of Shallot (TSS).

The use of TSS has many advantages, especially related to the availability of good quality seeds and among others, have a longer storage time, low variations in seed quality, high productivity, and rarely contamination by seed-borne pathogens (Pangestuti and Sulistyaningsih, 2011). By looking at the opportunities and advantages of TSS, it is necessary to develop seed production of shallot to fulfill the lack of good quality seeds and can be available all the time, especially in Central Sulawesi. Based on the above, it is important to conduct a direct study on the farmers' land to determine the growth and yield of true shallot seed of production.

MATERIALS AND METHODS

Time and place

True shallot seed production activities were carried out from May to September 2018 in Wuasa Village, North Lore District, Poso District (-1,42678, 120,32484, 1155,9 m) at an altitude of 1083.7 meters above sea level. Location was selected in accordance with true shallot seed production requirements, which are: highlands> 900 m above sea level, not foggy, and not windy, and there are no shallot plants that flower in the vicinity for a minimum of 1 km (in accordance with the requirements in the Ministerial Regulation No. 131/Kpts/SP.130/D/11/2015).

Materials and tools

The materials used in this study were Bima Brebes Variety, Apis cerana, Benzyl Amino Purine (BAP), Boron, organic fertilizer, inorganic fertilizer, *Tagetes erecta* flowers, herbicides, fungicides, and insecticides. The equipment used is analytical scales, hoes, mulch, plastic shade, bamboo, mild steel, nails, hand sprayers, gauges, and other support tools.

Research methods

Research with an on-farm research approach of 3000 m^2 on farmer's land. The study used the Probability Sampling method with 10 replications. The samples were 10 plants in each replication, so that there were 100 observation units.

Research implementation

The stages of the research implementation consisted of planting preparation (including *Tagetes erecta* planting, land management, application of organic and basic fertilizers, mulch installation, and tuber vernalization), tuber planting, shade installation, application of supplementary fertilizer, application of boron, pest, and disease control, harvesting, and data collection and analysis.

Data collection and analysis

Analysis of the data included the average tabulation of growth components (plant height, number of tillers, and number of leaves), flowering components (percentage of flowering plants, number of flowers/clumps, flower stem height, flower diameter, percentage of capsules formed), and yield components (number of capsules/flowers, weight of 1000 seeds, number of tubers/clumps, weight of tubers/clumps, diameter of tubers, and weight of tuber).

RESULTS AND DISCUSSION

Observations on the growth components of the true shallot seed production development activities were carried out on the parameters of plant height, number of tillers, and number of leaves. Based on the observations shown in Table 1 and Figure 1, the average plant height produced reached 44.08 cm. These results indicate the growth of plant height is included in the optimum category because it can reach the maximum height of shallots based on the description of the Bima Brebes variety, which is 25-44 cm. Optimum results were also obtained in the number of leaves parameters, which amounted to 43.6 leaves with a description of the variety of 14-50 leaves. In the parameter number of tillers, the number of tillers is 8.02. Although these results are not optimum, they are still in the potential number of tillers based on varieties description, namely 7-12 tillers.

The high result of the growth components was influenced by genetic factors and environmental suitability that support the expression of the superior character of Bima Brebes Variety. This is in line with Sinaga, Bayu, and Nuriadi (2013) which states that a plant's genetic traits cannot emerge a certain character unless it is in the appropriate environmental conditions. In addition, Kurniawan, Kusmana, and Basuki (2009) stated that phenotypic characteristics of varieties are not always determined by differences in genotype, but also because of differences in environmental conditions or interactions of that factors. the provision of plastic shade also affects the rate of shallots growth, especially on plant height and number of leaves parameters. This is in line with research conducted by Sumarni et al. (2010), which states that the provision of transparent plastic shading significantly affects plant height and number of leaves per plant but does not affect the number of tillers per plant.

Table 1. The average of growth components in the development of true shallot seed production in Wuasa Village, North Lore Sub-district, Poso District, 2018.

Growth components	Value	Description*
Plant height (cm)	44,08	34,5 (25-44)
Number of tillers	8,02	7-12
Number of leaves	43,6	14-50

Note: * Description of Bima Brebes Variety (Balitsa, 2019).

Figure 1. The performance growth components in the development of true shallot seed production in Wuasa Village, North Lore Sub-district, Poso District, Central Sulawesi, Indonesia, 2018.

Table 2 and Figure 2 show the results analysis of the flowering components of shallots which include the percentage of flowering plants, number of flowers per clump, flower height, flower diameter, and percentage of capsules. In the number of flowers per clump, parameter obtained 2.83 flowers, while the height of the flower stalk parameter obtained 48 cm. According to Wibowo (2007), the height of the flower stalk ranges between 30-50 cm. Based on this, the average height of the flower stalk obtained was at maximum high potential. In the parameters of the percentage of flowering plants, the result obtained was 78%. The results of the percentage of flowering plants obtained are relatively high compared to the results of other studies. According to Pandiangan et al. (2015), a good category of the percentage of flowering is 50%. The high percentage of flowering plants is influenced by the application of BAP (Benzyl Amino Purine). BAP is a growth regulator that plays a role in increasing cell division which can ultimately initiate flower formation.

Based on research by Kurniasari et al. (2017), the use of BAP can increase the percentage of flowering up to 50% with the number of flowers produced is 2 flowers per clump. Application of growth regulators aims to stimulate the formation of flowers, so the use of growth regulators can affect the number of flowers per clump. The application of boron also influences the formation of flowers because it functions in stimulating the formation of flowers and it is a microelement related to the metabolism of auxin hormone (Amanullah et al., 2010). Based on Table 2, the average number of flowers per clump obtained 2.83 flowers. However, although the percentage of flowering plants obtained was quite high (78%), the percentage of capsules formed was only 35%. High or low percentage of capsule formation obtained is influenced by the level of pollination factor.

Pollination of shallots is strongly influenced by the presence of pollinators. In general, one of the obstacles in the development of true shallot seed production is in the pollination process. According to Curran and Proctor (1990), male and female organs of shallot are not ripe at the same time, so the shallot plants are included in the cross-pollinating plant. Male organs in shallot flowers mature 2-3 days earlier than female organs (Chandel et al., 2004). Based on this, the role of pollinating insects to assist in the process of cross-pollination is needed, to increase pollination and seed formation. Like cross-pollinating shallots, Gure et al (2009) state that natural pollination without the introduction of pollinators only reaches 9% in onions. In the development of the production of true shallot seed in Central Sulawesi, the presence of pollinators in the production location that helped the process of pollination of shallots include *Apis cerana* and green flies. According to Palupi et al. (2015), *Apis cerana* is one of the most effective pollinators in shallot pollination compared to other pollinators such as *Apis mellifera, Trigona* sp., and *Lucilia* sp. While the advantages of greenfly pollinators are abundant population numbers and rapid movement from one flower to another (Kunast, 2013).

Table 3 and Figure 3 show the observations on the yield components which include parameters of number of capsules/flower, weight of 1000 seeds, number of tubers/clump, weight of tubers/clump, weight of tubers, and diameter of tubers. Based on the analysis, the number of capsules/flower produced is 80.67 capsules.

Table 2. The Average of Flowering Components in the Development of True Shallot Seed Production in Wuasa Village, North Lore Sub-district, Poso District, 2018

Flowering components	Value
Percentage of flowering plants (%)	78
Number of flowers per clump	2.83
Flower height (cm)	48
Flower diameter (mm)	46.40
Percentage of capsules (%)	35

Table 3. The Average of Yield Components in the Development of True Shallot Seed Production in Wuasa Village, North Lore Sub-district, Poso District, 2018.

Yield components	Value
Number of capsules/flower	80.67
Weight of 1000 seeds (g)	1.97
Number of tubers/clump	14.17
Weight of tubers/clump (g)	70.18
Diameter of tuber (mm)	21.56
Weight of tuber (g)	5.07

Figure 2. The performance flowering components in the development of true shallot seed production in Wuasa Village, North Lore Sub-district, Poso District, 2018.

Figure 3. The Performance Yield of True Shallot Seed Production in Wuasa Village, North Lore Sub-district, Poso District, 2018.

These results are relatively high compared to the results of research on true shallot seeds production by Kurniasari et al., (2017) who obtained 32 capsules/flowers by using BAP solution concentrating 50 ppm. It is known that the optimum number of capsules is obtained at BAP solution concentration of 37.5 ppm. This is in line with the research conducted by Rosliani et al. (2012), which states that BAP treatment at a concentration of 37.5 ppm gives optimum results both on increasing flowering and the number of capsules produced. From this statement, it was concluded that one of the factors that can affect the number of capsules produced in the production of true shallot seeds is the concentration of BAP given.

On the weight of 1000 seeds parameter, it was obtained of 1.97 g. These results are still below the weight of 1000 seeds obtained by Rosliani et al. (2013) which is 2.18 g. the factor that affects the difference of the results obtained is the time of application of BAP solution. According to the results of research by Kurniasari et al. (2017) on observing the weight of 100 seeds, the increase in weight of 100 seeds was influenced by the time of application of the BAP solution. BAP application at ages 2, 4, and 6 Weeks After Planting gave a weight of 100 seeds by 0.317 g, while the application of BAP at ages 1, 3, and 5 Weeks After Planting gives 0.237 g (Kurniasari, et al., 2017).

Observation of yield components was also carried out on the shallot tubers production, including the number of tubers/clump, weight of tuber, and diameter of tuber. Based on observations, the number of tubers/clump obtained was 14.17 tubers. The yield of tuber obtained is quite high. Meanwhile, the weight of tuber obtained was 5.07 grams with a tuber diameter of 21.56 mm. Based on the results of research by Ayu et al. (2016), the average number of tubers per clump produced by the Bima Brebes variety was 4.52 tubers with a tuber weight per clump of 24.02 gr and the average weight per tuber was 5.31 gr. The number of tubers and the diameter of the tubers are influenced by factors of phosphorus (P) and Potassium (K) nutrients. In addition, soil texture is also very influential in the formation of tubers. P nutrient deficiency in shallot production activities will inhibit the growth of roots, leaves, and tubers. Type of soil containing high clay fraction will affect the process of tuber development which results in small tubers produced. While loose and fertile soil will encourage the development of large tubers and produce a high number of tubers (Napitupulu and Winarto, 2010; Brewster, 1994).

In conclusion, growth, and yield performance for true shallot seed production in Wuasa Village, North Lore Sub-district, Poso District, Central Sulawesi showed high results. The suitability of agro-climate and the right production technique is the success factor in expressing the optimum growth and yield of shallot that is obtained. The percentage of flower formation showed a high result of 78% with the number of capsules formed at 80.67 capsules per flower. Shallot variety of Bima Brebes has a good chance to produce seed from seed because it has > 75% flower in the highlands.

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Simple carbon and organic matter addition in acid sulfate soils and time-dependent changes in pH and redox under varying moisture regimes

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Abstract. *Michael PS. 2020. Simple carbon and organic matter addition in acid sulfate soils and time-dependent changes in pH and redox under varying moisture regimes. Asian J Agric 4: 23-29.* It has been recently shown that the addition of dead plant materials as organic matter in acid sulfate soils (ASS) creates microenvironments conducive for soil microbes to reduce the sulfate content and redox potential (Eh), thereby increasing the pH of sulfuric soil and sustaining the pH of sulfidic soil, respectively. The time course of effects of addition of organic matter however was not clearly established. This was study conducted to assess the time course of the effect of soil carbon and organic matter following addition of glucose and acetate as carbon and lucerne hay as organic matter sources, respectively. The conditions of the treatments were either anaerobic (flooded) or aerobic (under 75% field capacity). The results showed that the effects of addition of amendments on pH and Eh of ASS are immediate, but treatment dependent. Organic matter as a multiple food sources for soil microbes was more effective in reducing the soil and increasing the pH of the sulfuric and the sulfidic soil, respectively. The effects of acetate were comparatively higher than glucose, dependent on the type of microbial ecology that was engaged by these resources being simple carbon sources. The overall increase in pH and reduction in Eh was immediate within the first three days, but the changes in the soil properties measured were reversed over time. The reversal in the mechanism inducing the changes in pH and Eh ceased as the microbial metabolic resources used as metabolic substrate to generate alkalinity got depleted.

Keywords: Acid sulfate soil, Eh, organic matter, pH, simple carbon, time-course

INTRODUCTION

Acid sulfate soils (ASS) are naturally occurring soils that either contain actual sulfuric soil (pH<4) or sulfidic soil (pH>4) that has the potential to form it (Pons 1973) in an amount that can have serious negative impacts on other soil properties (Fitzpatrick et al. 2009). Under inundated soil conditions, sulfidic soils pose no threat unless exposed to atmospheric oxygen and the sulfidic minerals (e.g., pyrite, FeS_2) are oxidized, resulting in production of sulfuric soil (acidity). Release of sulfuric acid, in turn, leads to solubilization of acidic minerals and toxic constituents from the soil matric in which these are held (Michael et al. 2015).

Generation of sulfuric acidity, mobilization of acidic minerals (e.g., Fe^{3+} and Al^{3+}), and accumulation of potentially other toxic soil constituents (e.g., As) are responsible for the negative environmental and health impacts (Michael 2013). Unless successfully managed, the negative impacts on soil and water characteristics, human health, commercial and recreational fisheries, engineered and community infrastructure, scenic amenity and tourism, and agricultural productivity are all seriously compromised (Sammut et al. 1996; Sammut 2004; Powell and Martens 2005; Michael 2013).

The seriousness of the negative impacts of ASS has led to the development of several management strategies, focusing on two key principles. The first principle is to neutralize the sulfuric acidity by application of alkaline material, e.g., agricultural lime. The second principle is to curtail sulfidic soil oxidation by inundation through flooding (Fitzpatrick et al. 2009). In some places, however, lime availability is a problem (Powell and Marten 2005) and under many general soil use conditions, flooding is undesirable (Michael et al. 2015b).

We have shown recently that addition of plant materials in the form of complex organic matter containing varying carbon and nitrogen content creates alkalinity using microbial processes (Michael et al. 2012; Michael, 2018a,b,c). Addition of the organic matter creates anoxic environment by microbial depletion of oxygen, conducive for microbes, such as sulfate-reducing bacteria (SRB) to flourish and generate biogenic alkalinity (Michael et al. 2015a). It further found that the biogenic alkalinity generated resulted in amelioration of sulfuric soil (increased pH) and prevented oxidation of the sulfidic soil (Michael et al. 2016; Michael and Reid 2018). In comparison to lime, our findings are significant contributions towards ASS management with organic matter being relatively cheap and readily available. The effects of live plants on acidification, redox potential, and sulfate content have been recently reported(Michael et al. 2017; Michael 2020).

In our studies (e.g., Michael et al. 2015), organic matter was either applied as surface mulch or incorporated in the ASS and incubated for "six months" prior to the measurements. As the changes in soil chemistry (pH, Eh, and sulfate content) were measured based on "long-term studies" of six months, the short-term changes in soil chemistry induced by the organic matter applied (important information needed for timely management of ASS negative impacts) was not clear. Therefore, this study was conducted to assess the time-dependent changes in ASS chemistry (pH and Eh) induced following addition of amendments over a shorter period (12 weeks).

MATERIALS AND METHODS

Soil materials

The ASS used in this study was well described in Michael et al. (2016). The sulfidic soil was collected from a "sulfuric subaqueous clayey soil" (Fitzpatrick 2013) at a depth of approximately 1 m in the Finniss River in South Australia (35˚24'28.28''S; 138˚49'54.37''E). Figure 1 of Michael et al. (2015a) shows the map of the sampling site. Detailed information on soil classification using the Australian ASS Identification Key (Fitzpatrick et al. 2008) and Soil Taxonomy (Soil Survey Staff 2014) are given in Table 1 of Michael et al. (2016). In addition, a comprehensive list of references containing additional information on soil morphology and geochemistry prior to rewetting (i.e., sites AA26.3 and FIN26 in Fitzpatrick et al. (2009a)) and after reflooding are given in the same table.

The pH of the freshly collected sulfidic material measured in water 1:5 (pH_w) was 6.7 and the pH following peroxide treatment (pH_{ox}) was 1.4, with the water holding capacity estimated to be 49%. The residual organic matter content, estimated using the weight loss-on-ignition method (Schulte and Hopkins 1996) was 10.6%. To manufacture "sulfuric horizon material" by oxidizing the sulfides, the sulfidic soil material was spread thinly on plastic sheets and kept moist until $pH_w<4$. The manufactured sample and freshly sampled sulfidic material are henceforth referred to as "sulfuric soil" ($pH_w<4$) and "sulfidic soil" ($pH_w>4$), respectively.

Carbon and organic matter sources

Laboratory grade glucose and acetate, obtained from a chemical store within the School of Biological Sciences, The University of Adelaide, South Australia were used as simple carbon sources. To use as organic matter, bales of lucerne hay were purchased from a supplier in Adelaide, South Australia, and prepared as described by Michael et al. (2016). The plant materials were chopped into pieces and then oven-dried at 60 ºC for three days. The dry pieces were finely chopped using an electric blender to ≈0.5 mm. The nitrogen content of the organic matter analyzed by ICP-OES using 0.5 g samples (n=3) was estimated to be 3.2%. The carbon content can be approximated from the data in Kamp et al. (1992).

Experimental treatments

Three experiments were conducted, as described below, with glucose and acetate as simple carbon sources and lucerne hay as organic matter incorporated in the soils by bulk mixing in proportions of 25:2, 10:1, and 80:1 (amendments: soils w/w). Set in 80 ml Falcon tubes, all the treatments were replicated three times and set out in a complete randomized design under glasshouse conditions. Although the 'aerobic treatments' were regularly watered, it was probable that the moisture was unevenly distributed over time, with the upper parts being aerobic and the lower parts of the profile becoming waterlogged. The anaerobic treatments were always under flooded conditions with adequate amount of water ponding on the surfaces (Michael et al. 2016).

Experiment 1. This was conducted under flooded conditions using sulfuric soil to assess the changes in soil chemistry induced by organic matter as a function of time over 5 weeks.

Experiment 2. Two sets of treatments with glucose, acetate, or organic matter mixed in sulfuric soil each were prepared. The first sets of the treatments were maintained under aerobic soil (regular watering) conditions while the second sets were maintained under anaerobic (flooded) soil conditions, respectively.

Experiment 3. The description of this experiment is like that of experiment 2, except that sulfidic soil was used instead.

Measurements

In experiment 1, pH and Eh were logged daily from within the top 20 mm of the soils. In experiments 2 and 3, changes in Eh and pH were measured from the surface (0-10 mm), middle (10-20 mm), and deep (30-40 mm) profiles as described previously (Michael et al. 2015). Redox was measured using a single Ag/AgCl reference and platinum (Pt) electrode combination using an automated data logger (Dowley et al. 1998; Merry et al. 2002). To measure the Eh, the frame of the redox probe was marked as per the targeted profiles from the tip (e.g., 0-10 mm, etc.).

During measurement, the Pt and the reference electrodes were both inserted into the soil from the surface. The reference electrode remained inserted whilst the Pt electrode was driven invariably once at a time into the soil. This allowed to equilibrate for 10 min and then Eh logged at 1 min intervals for the next 10 min and averaged (Rabenhorst et al. 2009). These values were corrected for the reference offset to be relative to the potential of a standard hydrogen electrode by adding 200 mV (Fiedler et al. 2007). The stability and accuracy of the electrodes were maintained as per Fiedler et al. (2007). pH was measured using a 2 g soil (1:5 water) with a pre-calibrated Orion pH meter (720SA model) (Michael et al. 2012).

Statistical analysis

The Eh values obtained over a 10 min period were averaged and a treatment average was obtained by taking the mean of the three replicates. Similarly, treatment average pH was obtained by taking the mean of the three replicates. To compare the treatment means, significant differences ($p<0.05$) between treatment means of each profile were determined by two-way ANOVA using statistical software JMPIN, AS Institute Inc., SAS Campus Drive, Cary, NC, USA 27513. If an interaction between the treatments and profile depths was found, one-way ANOVA with all combinations was performed using Tukey's HSD (honestly significant difference) and pairwise comparisons.

RESULTS AND DISCUSSION

Effects of organic matter on sulfuric soil pH and Eh

The changes in pH and Eh of the sulfuric soil as a function of time initially measured over 5 weeks in an 80 g of flooded soil with or without incorporation of 1 g of lucerne hay are shown in Figure 1. Within the first 3 days, an immediate increase in pH by 1.1 units from an initial pH of 4.4 and a decrease in Eh was observed in the lucerne hay treatment. Changes in the control treatment were slower. Over the next 14 days, pH continued to increase and Eh declined. Between 18 and 38 days, the pH of the control soil fell, corresponding to an increase in Eh, while the pH of the lucerne hay treatment continued to rise and Eh was relatively stable. The time-course of changes in pH and Eh following the addition of complex organic matter showed that the values obtained by these treatments are highly dependent on treatment time.

The highly oxidized sulfuric soil got reduced, as expected, due to the flooded condition which created anoxia in both the control and in the soil with organic matter. The changes in pH and Eh measured however were influenced by the added organic matter. The soil used contained 10.6% residual organic matter (Michael et al. 2015b), but the addition made significantly influenced the overall changes measured. For example, within the first 15 days, both the pH of the control and amended soils increased, and there was a concurrent reduction in Eh but the changes in the amended soil pH were higher (Figure 1.A), and reductions in the Eh were lower (Figure 1.B). As the small amount of residual organic matter in the control soil got depleted, the pH lowered to respond to a positive increase in Eh within the next 20 days. Comparatively, the pH continued to increase and Eh decreased in the complex organic matter amended soil parallel to the slow decomposition of the complex plant materials.

It has been reported that the close correlation between the generation of alkalinity and the decrease in soil sulfate concentration is a strong indication that the pH changes resulted from the action of sulfate-reducing bacteria (SRB) that used the organic matter as a carbon source (Michael et al. 2016; Michael 2018a). The time course of effects of the changes in pH, Eh, and sulfate content were not clearly established in ASS following the addition of amendments. The rapid changes in pH and Eh immediately following flooding (Figure 1) suggested that initially aerobic microbial activity was involved, and that the oxygen demand generated by these organisms drove the Eh into the range that was more suitable for anaerobic bacteria such as sulfate reducers. The pH of the control soil also responded to flooding and increased over the first 18 days but then began to decline. The changes in Eh mirrored the changes in pH. The differences between the control soil and the lucerne treatment were in the speed and magnitude of the changes, being much slower in the control and reversing after approximately 3 weeks (Figure 1), possibly due to the exhaustion of the residual carbon in the soil. Apart from the alkalinizing effects found, organic matter contains other essential metabolic substrates in addition to carbon and nitrogen (Jarvis and Robson 1983; Jarvis et al. 1996; Marschner and Noble 2000) that are beneficial to the sulfuric soil and the microbes in holding water and creating soil microenvironments.

Effects of organic matter and simple carbon on sulfuric soil pH and Eh

The results presented in Figure 1 showed that the daily changes in pH and Eh as a function of time following addition of organic matter were highly dependent on treatment time. Therefore, the effects of simple carbon compounds (as a major component of the complex organic matter) after 3, 6, and 12 weeks were monitored for comparison. Throughout the 12 weeks, the control soil pH remained strongly acidic under aerobic conditions (Figure 2.A). Among the amended treatments, the order of increase in pH was lucerne hay > acetate > glucose and the changes elicited were rapid and essentially complete by 3 weeks, although lucerne hay continued to increase it beyond the 3 weeks (Figure 2.C). The increase in pH strongly correlated with reductions in Eh, with large changes observed in the lucerne and acetate treatments.

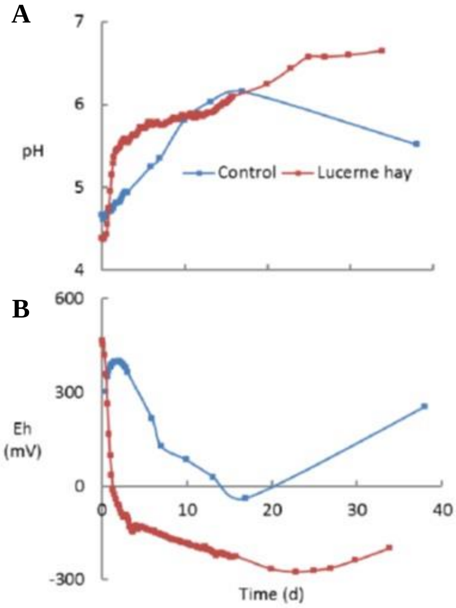

Figure 1. Short-term changes in (A) pH and (B) redox of sulfuric soil maintained under anaerobic conditions with or without addition of organic matter (lucerne hay). The initial pH was 4.4.

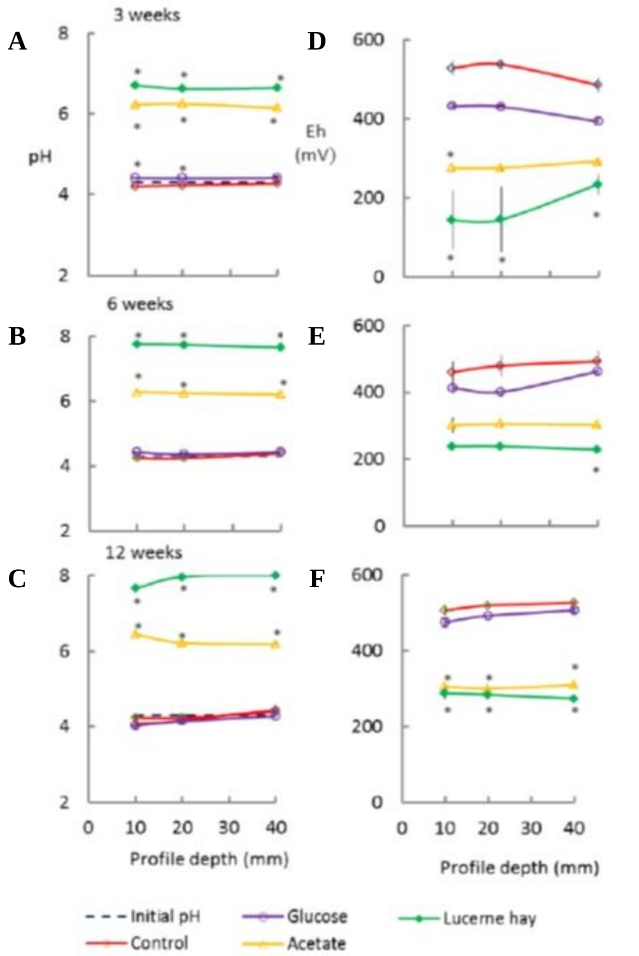

Figure 2. Timely changes on (A-C) pH and (D-F) redox of sulfuric soil maintained under aerobic conditions for 3-, 6-, and 12-weeks following addition of organic matter and simple carbon compound sources. The values are means ± s.e. of three measurements (n=3). Asterisks indicate significant differences ($p<0.05$) between treatment and control at the same depth.

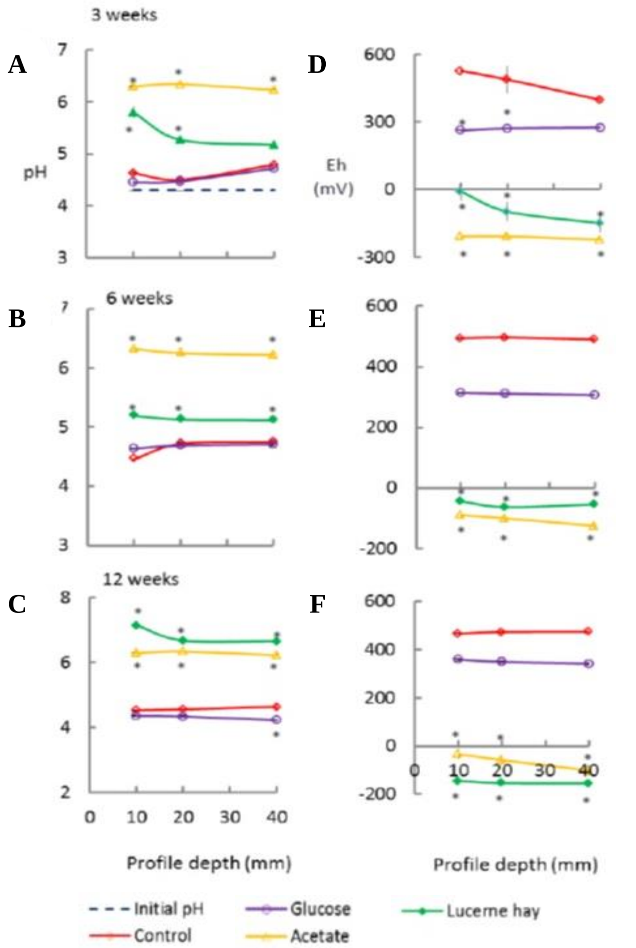

Figure 3. Timely changes on (A-C) pH and (D-F) redox of sulfuric soil maintained under anaerobic conditions for 3-, 6-, and 12-weeks following addition of organic matter and simple carbon compound sources. The values are means ± s.e. of three measurements (n=3). Asterisks indicate significant differences ($p<0.05$) between treatment and control at the same depth.

Under anaerobic conditions, the effect of acetate was immediate and lucerne hay with high nitrogen content was time-dependent (Figure 3). Acetate increased the pH to 6.3 units throughout the profiles within the first 6 weeks (Figure 3.A), with lesser changes from the lucerne hay. No responses in the control and glucose treatments were evident during the 6 weeks (Figure 3.B). After 12 weeks, the effects on soil pH of all the treatments remained like 6 weeks, except that lucerne hay increased it to units higher than 7 at the surface and 6.5 throughout the profile (Figure 3.C). pH of the soil amended with glucose remained lower than 5, close to the initial pH. The overall effects of the amendments on pH after 12 weeks were: lucerne hay > acetate > control > glucose.

A major difference between the aerobic and anaerobic treatments was in the response of Eh. As shown in Figures 2.D-F, the unamended control soils, and the soil amended with glucose remained highly oxidized at the end of three weeks, while the soils amended with acetate and lucerne hay were moderately reduced (150–300 mV) throughout under aerobic conditions. Under anaerobic conditions, the control and the glucose amended soils remained highly oxidized throughout the 12 weeks (Figures 3.D-F) but the Eh of the lucerne hay and acetate amended soils were 200 to 400 mV lower and remained below 0 mV throughout.

Under aerobic conditions, acetate and lucerne hay increased the pH of the sulfuric soil to near 7, within the first 3 weeks (Figure 2.A). Within the next 6 weeks, the effects of acetate remained stable but lucerne hay increased to 8 (Figures 2.B-C), a strong indication that decomposition of the organic matter was slow. In the control and glucose treatments, the pH gradually decreased over 12 weeks (Figures 2.A-C). The changes in pH were generally correlated with changes in Eh, with lucerne hay and acetate maintaining an Eh in the range of 100 to 300 mV at the surface within the first 3 weeks then stabilized within the range of 200 to 300 mV in the next 6 weeks (Figures 2.D-F). The Eh of the control and glucose treatments were generally high throughout.

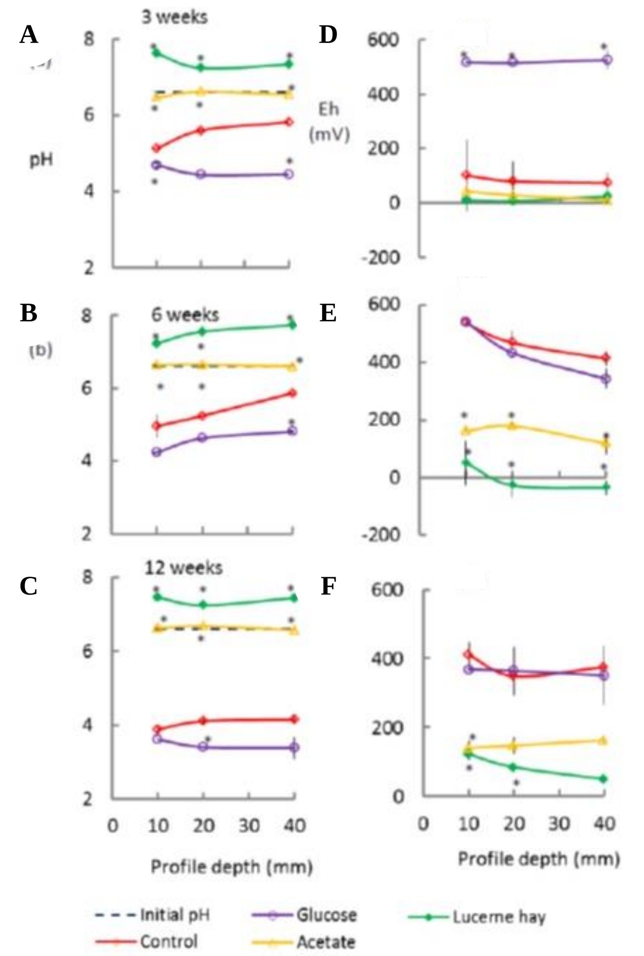

Figure 4. Timely changes on (A-C) pH and (D-F) redox of sulfidic soil maintained under aerobic conditions for 3-, 6-, and 12-weeks following addition of organic matter and simple carbon compound sources. The values are means ± s.e. of three measurements (n=3). Asterisks indicate significant differences (p<0.05) between treatment and control at the same depth.

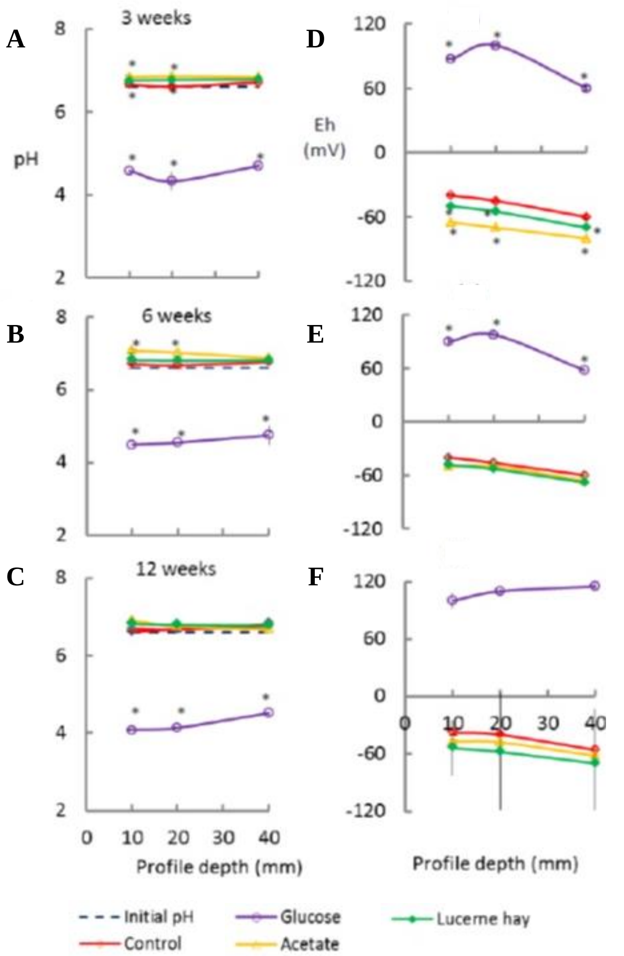

Figure 5. Timely changes on (A-C) pH and (D-F) redox of sulfidic soil maintained under anaerobic conditions for 3-, 6-, and 12-weeks following addition of organic matter and simple carbon compound sources. Asterisks indicate significant differences (p<0.05) between treatment and control at the same depth.

Effects of organic matter and simple carbon on sulfidic soil pH and Eh

A more detailed investigation of the effects of simple carbon compounds on sulfidic soil (pH >4) was undertaken by conducting a time course study over 12 weeks under both aerobic and anaerobic conditions, and the changes were compared with those induced by lucerne hay as complex organic matter. As shown in Figures 4.A-C, acetate stabilized the pH while lucerne hay increased it under aerobic conditions within the first 3 weeks then remained stable. The pH of the control and glucose treatments acidified after 12 weeks.

The changes in pH generally correlated with changes in Eh under aerobic conditions, with lucerne hay and acetate maintaining an Eh in the range of 0 to 200 mV at the surface (Figures 4.D-F). Within the first 3 weeks, Eh of the control treatment fell to around 100 mV, but then became quite aerobic, which corresponded to more rapid decrease in pH. The Eh remained high throughout the glucose treatment, corresponding to the low pH. Under anaerobic conditions (Figure 5), pH of the control sulfidic soil was stable as expected, and there was no significant effect of lucerne hay and acetate on pH of the amended treatments. Glucose, however, acidified the soil in the first 3 weeks but between the 6 and 12 weeks, the changes in pH remained relatively stable, between 4 and 5 units (Figures 5.B-C). The Eh in all treatments except glucose was around -60 mV at all sampling times (Figures 5.D-F). In the glucose treatment, Eh was also reduced but was mostly in the range of 60 to 100 mV.

Under anaerobic conditions, reduction reactions increased the pH of the control sulfuric soil to near 4.5, lucerne hay to near 6, and acetate to 6.5 within the first 3 weeks (Figure 3.A). However, glucose-induced a very small change in pH in the same period and remained relatively near 4 in the next 6 weeks (Figures 3.B-C). The Eh in all treatments except the control and glucose was around -100 mV at all sampling times. In the control and glucose treatments, Eh was mostly in the range of 300 to 500 mV. As shown in Figure 1 and based on our other recent related studies (e.g., Michael et al. 2015; Michael 2015b), flooding induces reduction reactions and results in lower Eh. The control treatment of this study was not reduced under the flooded soil condition as expected and remained aerobic. This was attributed to probable uneven distribution of residual organic matter content or poor microbial ecology of the sulfuric soil to begin the reduction processes. In addition, a strong smell of butyric acid was present during measurement in the soils amended with glucose, which may have been the product of a specific bacterial metabolism.

Soil microbes use a range of carbon compounds (e.g., glucose and acetate) for cellular respiration, with the main limitation for microbial activities to use them as a carbon source and alter the soil chemistry is the supply (Kuzyakov et al. 2000; Neff et al. 2002). However, there was a clear difference in the effects of glucose and acetate on soil pH, under both aerobic and anaerobic conditions (Figures 4 and 5). Under aerobic conditions, acetate was able to stabilize the pH of sulfidic soil (Figure 4.A) and had minimal effect under anaerobic conditions (Figure 5.A), and rapidly induce moderate reductions in Eh (Figures 4.D and 5.D).

Comparatively, glucose maintained a high Eh throughout and caused strong acidification under all the moisture conditions (Figs. 4 and 5). The obvious odor of butyric acid suggests that the acidification with glucose even in the sulfuric soil was mediated by fermentative microbes producing acidic metabolic end products. The effects of acetate were interesting, in relation to the apparent requirement for nitrogen when complex organic matter is added. The changes in pH and Eh following addition of acetate under aerobic conditions were almost as great as those of lucerne hay and equally rapid (Figure 4). Since acetate can be utilized as an energy source by some groups of bacteria under aerobic conditions and by sulfate and iron-reducing bacteria under anaerobic conditions (Kamura et al. 1963; Thauer et al. 1989), the observed ameliorative effects may have resulted from the biogenic alkalinity generated by a range of microbial ecology (Michael 2018a).

In conclusion, we have demonstrated through various recent long-term studies that the addition of a range of nutrient sources, under either aerobic or anaerobic soil conditions, engages a range of soil microbial ecology that alters the chemistry of ASS. The time course of effects of the additions, however, was not clearly established by our studies. This study demonstrated that the effects are time course and treatment dependent. The effects of addition of complex organic matter were immediate, however as the food sources got exhausted, the effects measured reversed over time. The effects of addition of glucose and acetate were variable, a strong indication that a range of soil microbes is engaged in altering the chemistry of ASS, dependent on amendments. This study also showed that addition of amendments that are multiple sources of food (e.g., organic matter) engages a range of soil microbes to alter the chemistry of ASS more effectively than alone food source (e.g., glucose), which would only attract a handful of microbes. In addition, it was found that the effects of the amendments on pH and Eh measured were stable up to six months, but the protection was lost after 12 months.

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