International Journal of Innovation Sciences and Research
Vol.8, No, 06, pp.1376-1382, June, 2019
***Available online at** http://www.ijisr.com*

RESEARCH ARTICLE

EFFECT OF TWO BIOFERTILIZERS BASED ON *AZOLLA FILICULOIDES* AND *AZOLLA CAROLINIANA* IN CABBAGE PRODUCTION (*BRASSICA OLERACEA*) HAUT SASSANDRA(DALOA, CÔTE D'IVOIRE)

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Accepted 28th May 2019; Published Online 30th June 2019

ABSTRACT

The present study aims to evaluate the interest of two forage species: *Azolla filiculoides, Azolla caroliniana* compared to the compost on cabbage productivity in Daloa.The test was conducted according to a device composed of a complete randomized block with 2 repetitions per treatment comprising 4 treatments. The establishment density of cabbage plants is at spacings of 40 cm x 40 cm at the rate of 27 plants per plank on an area of 4 m² per repetition. The treatments were compared, and evaluated by accounting the agronomic parameters: The results showed a greater growth and development of plants on plots amended with Azolla followed by compost compared to unamended plots. The highest yield has been obtained with *Azolla caroliniana* that produced 20.87 t / ha of cabbage. The study showed that *Azolla filiculoides* and *Azolla caroliniana,* under the conditions of this test had great potential for improving the availability of soil nutrients and could provide the needed nutrients amount for cultivation of cabbage without the use of mineral fertilizers.

Key words:Cabbage (*Brassica oleracea*), *Azolla Caroliniana, Azolla Filiculoides,* Compost, Agronomic Parameters, Yield.

INTRODUCTION

Market gardening, particularly *brassica Brassica*, is the most widely used food resource worldwide Gruarin S. (1998). Cabbage is the most consumed vegetable product in Asia but also in Africa and America (Talekar and Shelton, 1993). The agricultural sector plays a major role in the socio-economic life of Côte d'Ivoire. Market gardening, which occupies not only a significant place in the country's agriculture and economy, is an activity that responds to urban food preferences and demand (Singbo *et al.,* 2004). It is the main source of vitamins and trace elements for the populations (Sangaré *et al.,* 2009). Cabbage is a vegetable of great importance in the diet of man. It is an important source of vitamins A, C and E Depezay L. (2006). It brings the minerals and vitamins needed into a diet and generates substantial income for producers and others involved in the marketing system, thereby reducing poverty and creating jobs. In Côte d'Ivoire, demand for cabbage is increasing and prices are attractive except during periods of overproduction (October - November). The unit price of cabbage can vary from 100 to 400 FCFA / kg (Dao D., *et al.,* 2003).In an effort to increase agricultural production and productivity to meet the high demand for vegetables, producers are using various types of pesticides and synthetic fertilizers in a massive and abusive manner. This is to ensure the availability of market gardening products for both local markets and for export in case of surplus. The use of these chemical fertilizers, thanks to their immediate beneficial effect on the productivity of vegetable crops, is one of the solutions, but their high cost and unavailability make them almost inaccessible to small farmers (Useni *et al.,* 2013).

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In addition, the use of these chemicals pollutes the air, disturbs the soil balance, and contaminates the water of Cissé I. (2000). And causes a decline in soil organic matter (Mulaji KC 2011). In the face of these socio-economic and environmental issues, it becomes imperative to look for other sources of nutrients that can enable sustainable agriculture that respects the environment and human health. In such a context, organic fertilization should be an appropriate alternative for restoring soil fertility. Numerous studies have shown that the use of Azolla plays an important role in increasing rice grain yield and various soil properties (Becking, JH (1979)) (DJOGBEDE *et al.,* 2012). Given the importance of this aquatic fern of the genus Azolla containing a species of seaweed *Azollae anabaena* capable of producing nitrogen assimilable by the plant and following the high price of synthetic fertilizers that makes them inaccessible to the majority of farmers, The present study The present study tests the hypothesis that Azolla is an inexpensive alternative in cabbage culture under the edapho-climatic conditions of Daloa. This study was conducted to evaluate the interest of two forage species: *Azolla filiculoides, Azolla caroliniana* compared to compost on cabbage productivity in Daloa.

MATERIALS AND METHODS

Material

This study was conducted at the University Jean Lorougnon Guédé (UJLoG), located in the department of Daloa, region of Haut-Sassandra in the center-west of Côte d'Ivoire, located between 6 ° and 7 ° north latitude and 7 ° and 8 ° West Longitude. This region covers an area of 15,200 km² with a population estimated at 1,430,960 inhabitants (INS 2014). The climate of this region is characterized by an equatorial and subequatorial regime with two rainfall maxima with four seasons the long rainy season lasts from April to mid-July, with a peak in June. The short dry season runs from mid-July

to mid-September, the short rainy season runs from mid-September to November with a peak in September while the long dry season lasts from December to March (N'Guessan AH *et al.,* 2014). These two maximas are separated by one or two months more or less rainy (Brou, 2005). The wet and dry seasons alternate with temperatures ranging from 24.65 ° C to 27.75 ° C on average (N'Guessan *et al.,* 2014). Daloa has ferralitic soils with good agricultural qualities for all crop types (Zro FGB *et al.,* 2016). Almost all of the basin is in a tropical humid zone with a dense forest vegetation with a regressive evolution. The degradation of this forest is accelerated by the intensification of cash crops (cocoa, coffee, oil palm and rubber). Extensive and itinerant cultivation practices and the uncontrolled exploitation of forest tree species have notably pushed back the limits of this forest (Sangaré, A *et al.,* 2009). Annual rainfall has increased from 1868.5 mm in 1968 to 1200 mm in 2014, a decrease of about 40% (Ligban *et al.,* 2009). Hydrographically, the region is influenced by the Sassandra River and its tributaries (Lobo and Davo) and Buyo Dam Lake (Koffié-Bikpo and Kra, 2013). Material: The plant material consisted of cabbage seed (*Brassica oleracea L.*) of the variety Majesty. The agronomic characteristics of the variety are given in Table 1.Fertilization was provided in five fractions for all fertilizers. A two-month compost consisting of 100 kg of chicken manure, 50 kg of white sawdust, 35 kg of coffee pulp, and 100 kg of charcoal was used as organic manure. The compost was incorporated at 34.3 kg per board in five inputs and was applied in increasing doses. *Azolla filiculoides* and *Azolla caroliniana* are aquatic ferns producing ammonia from atmospheric nitrogen. They come from the Azolla culture tanks near the experimental plot and have been used as biological fertilizer in rice and tomato cultivation in previous experiments at the study site. *Azolla filiculoides* and *Azolla caroliniana* were brought in liquid form, except the first input that was in solid form.

Methods

Experimental device: The experimental device is a randomized complete block with 2 repetitions of 4 treatments each. The block is subdivided into 8 elementary plots of 4 m × 1 m each, ie 4 m2 of surface area. The spacing between the elementary plots was 1m. The test was conducted on a total area of 72 m2. The cabbage plants were placed at 40 cm x 40 cm spacings at the rate of 27 plants per plank per 4 m² per repetition (test) or 8 m² per treatment (Fig 2).

Installation of the trial: The experiment was conducted from the dry season of December 2017 to April 2018. A nursery of cabbage plants was set up previously to have young cabbage seedlings that will be transplanted later between 30 and 35 days of sowing. The transplant took place on 19/01/2018 with a spacing of 0.4m x 0.4m. The various maintenance operations consisting of watering, weeding, weeding and phytosanitary treatments on cabbage plants are normally done.Data collection:

$$T = \frac{\text{number of transplants}}{\text{number of surviving plants}} \times 100 \quad (1)$$

It consists of collecting the following data:

- the rate of recovery, which was at the beginning of vegetation by the ratio of the number of plants that survived a week after transplanting.
- the length and width of the leaves, the number of leaves, the wingspan of the plant
- the yield: The evaluation of the yields (T ha-1) was made at 84 days after transplanting, according to the following formula

$$R = M/S \quad (2)$$

The measurements made relate to 10 plants fixed and numbered per board, ie 20 plants identified by treatment. These plants were chosen randomly. They were done once a week from the 44th day after sowing to the 65th day after sowing or 4 times during the cycle corresponding to time T1, T2, T3 and T4.Data analysis: The data was encoded using the Excel spreadsheet. The different analyzes were done using STATISTCA 7.1 software and the data processing was based on the analysis of variance and multiple comparison of averages test. Shapiro-Wilk normality tests were applied to all variables before performing all analyzes. This analysis of variance test (ANOVA) made it possible to see the significance of treatment effects on growth and yield parameters. In this condition, the Tukey test was used to classify the two-to-two means when the variables displayed a difference ($p < 0.05$) between treatments. The differences are significant for a probability value less than 0.05.

RESULTS

The recovery rate achieved with the different fertilizations was maximal (100%) on all the treatments including the control (fig 3).Variance analysis of leaf length of cabbage plants indicates that the effect of fertilization on leaf length is significant at the 5% threshold from the first week for all treatments. Leaf length is greater with *A. filiculoides* (28.27 cm and 31.94 cm) and *A. caroliniana* (25.43 cm and 30.18 cm) than with compost (24.08 cm and 25.28 cm) and the control (13.84 cm and 14.56 cm) at times T3 and T4 (Fig4). The effect of fertilization on the length of cabbage leaves of different treatments shows that treatment with A. filiculoides gives the greatest length. The treatment of cabbage with compost reveals that the growth in leaf length of plants becomes increasingly weak (from 24.08 cm to 25.28 cm) from T3 to T4 while it continues to increase in fertilization with *A filiculoides* (from 28.27 cm to 31.94 cm) and *A. caroliniana* (from 25.43 cm to 30.18 cm) at constant dose over time. However this growth becomes weak from the head. The analysis of variance posited reveals that the fertilization has a significant effect at the threshold of 5% on the width of the cabbage leaves from the first week for all the treatments. Treatments with *Azolla filiculoides, Azolla caroliniana* and compost resulted in better leaf width growth, which was found to be significantly different from those induced by the control. With these three treatments the leaf widths were 32.41 cm for *Azolla filiculoides*, 30.12 cm for *Azolla caroliniana* and 26.33 cm for compost at T3, whereas at the same date, these widths with the control were 12, 35 cm. At T4, the average width of cabbage leaves treated with the two species of Azolla is wider (36.4 cm and 35.06 cm) and significantly different from that induced by compost (28.47 cm) and the control (13.2 cm). This difference is well observed in Fig 4 showing the evolution of leaf width as a function of time.The analysis of variance in the number of cabbage leaves reveals a significant effect at the 5% level in the first week. The number of leaves emitted by the plants is higher in fertilization with the three biofertilizers (Table I). 4 weeks after transplanting, the effect of fertilization on the number of leaves emitted by the cabbage plants of the various treatments shows that *A. filiculoides* (21.7 cm) induces a foliar emission significantly higher than the other organic treatments (19.9 cm for *A. caroliniana* and 20.10 cm for

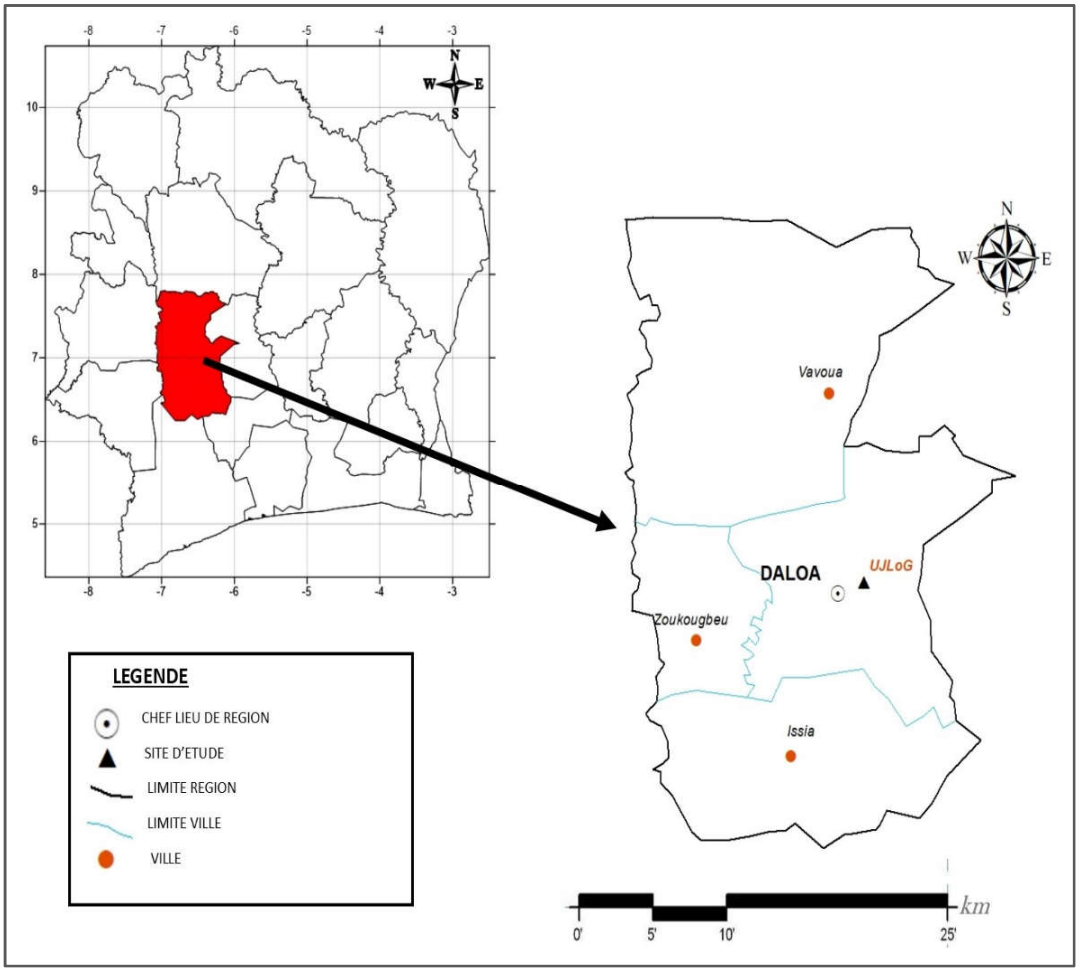

Fig. 1. Presentation of the study area

Table 1. Agronomic characteristics of the variety used (source: technical sheet)

Variety	Vegetative Cycle	Resistors	Average weight of the fruit	yield
Majesty	60 to 65 days after planting in the cool dry season.	• High: burst: 90 days after planting • Intermediates: Collar rot and powdery mildew	1 to 1.5 kg of weight	3 à 35 T /ha

Table 2. Fertilization of planks

		treatments			
Apports	Fertilization period	Witness	Compost	*A. filiculoides*	*A. caroliniana*
1er intake	1 semaine avant repiquage	0Kg/ board	10Kg/ board	10 Kg/ board	10 Kg/ board
2th intake	1 week after transplanting	0Kg/plant	150g/plant	30mL/plant	30mL/plant
3th intake	2 week after transplanting	0Kg/plant	150g/plant	30mL/plant	30mL/plant
4th intake	3 week after transplanting	0Kg/plant	300g/plant	30mL/plant	30mL/plant
5th intake	4 week after transplanting	0Kg/plant	300g/plant	30mL/plant	30mL/plant

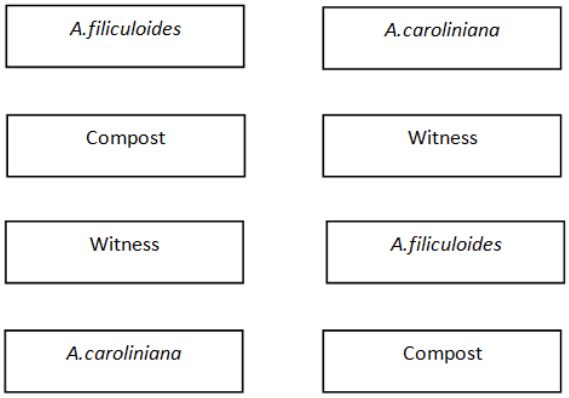

Fig. 2. Experimental device of the test

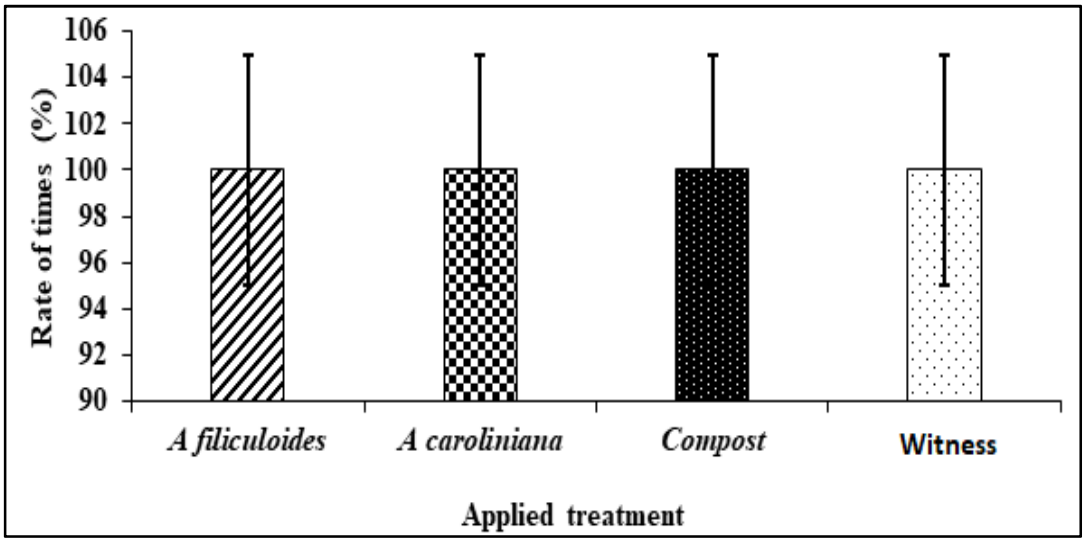

Fig. 3. Recovery rate of cabbage at 7 days after transplantation

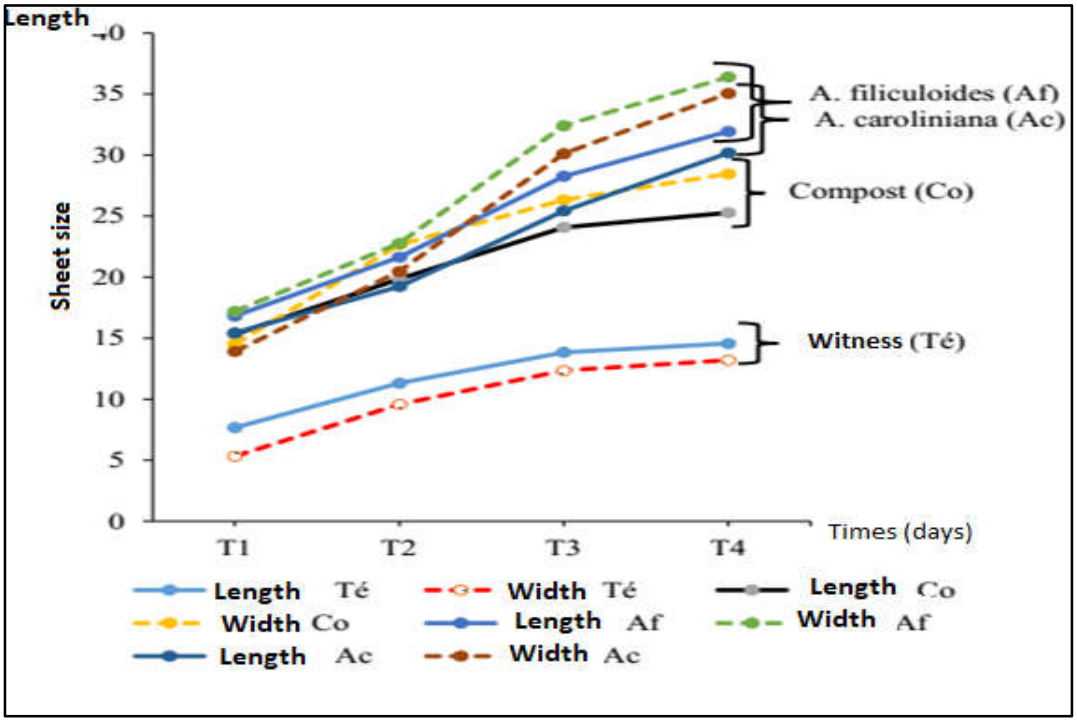

Fig. 4. Evolution of the length and width of cabbage leaves

Table 4. Evolution of the number of leaves of cabbage plants

Measurement time	Average number of cabbage leaves				
	Witness	Compost	*A. filiculoides*	*A. caroliniana*	*p-value*
T1	4,7 ± 1,8a	11,8 ± 2,4b	11,7 ± 1,8b	11,5 ± 2,1b	*1.89e-09*
T2	6,40 ± 2,2a	16,60 ± 3,5b	17,90 ± 2,4b	15,70 ± 1,4b	*4,14e-12*
T3	09,50 ± 2,3a	18,40 ± 3,5b	20,10 ± 3,1b	18,10 ± 2,2b	*2.31e-09*
T4	11,50 ± 2,3a	20,10 ± 3,2b	21,7 ± 2,7b	19,9 ± 1,9b	*3,32e-10*

For each average, values with the same letters (a, b, c) on the same line are statistically identical to the 5% threshold

Table 5. Spread of cabbage plants

Measurement time	Average size of cabbage plants				
	Witness	Compost	*A. filiculoides*	*A. caroliniana*	*p-value*
T1	12,85 ± 2,3a	34,75 ± 9,9b	34,79 ± 6,7b	32,07 ± 5,3b	*5.81e-09*
T2	22,07 ± 2,4a	48,42 ± 14,5b	49,93 ± 7,2b	41,34 ± 6,0b	*3,79e-08*
T3	22,80 ± 5,4a	51,28 ± 12,2c	59,07 ± 14,2bc	65,11 ± 6,2b	*1.36e-08*
T4	23,68 ± 5,3a	54,27 ± 11,7c	61,94 ± 13,3bc	67,35 ± 5,8b	*1,82e-09*

For each average, values with the same letters (a, b, c) on the same line are statistically identical to the 5% threshold

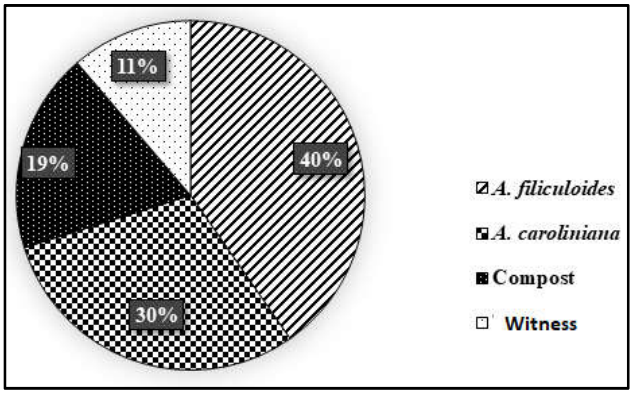

Fig. 5. Diagram of the number of apples per fertilization

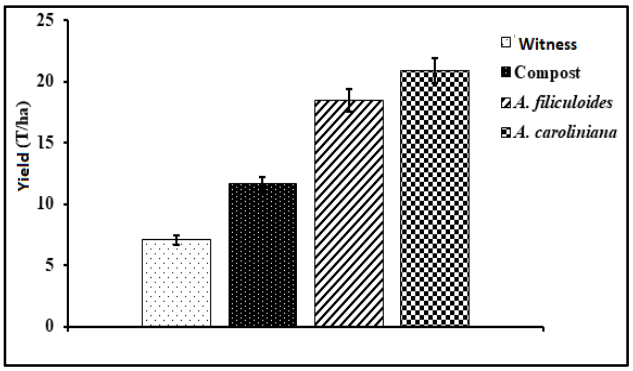

Fig. 6. Performance Evolution Diagram

compost) and without fertilizer (11.50 cm). Biofertilizers give more foliation than foliation in the absence of fertilizer. This leaf formation stops from the head for all treatments. The analysis of variance of soil cover by cabbage plants reveals a significant effect at the 5% threshold from the first week for all treatments. The wingspan of fertilized plants with both Azolla species and with compost is substantially equal to but larger than that of cabbage plants without fertilization (control) at T1 and T2 (Table II). The treatment with the compost reveals that the increase of the scale of the plants decreases with the increasing dose of compost and becomes weaker with 54,27 cm at T4 although this quantity of fertilizer increases. Moreover, on the same date, growth in the size of cabbage plants fertilized with *A. filiculoides* and *A. caroliniana* was higher, reaching 61.94 cm and 67.35 cm respectively. The increase in size ceases with the apples. Treatments with *Azolla filiculoides* and *Azolla caroliniana* yielded large proportions (40% and 30%) in front of compost (19%) and control (11%) (Fig 5).Treatments with Azolla yielded better and different from compost and control. Plants fertilized with *Azolla caroliniana* and *Azolla filiculoides* had high yields of 20.87 T / ha and 18.51 T / ha respectively, followed by compost treatments of 11.63 T / ha and control of 7 T / ha (Fig 6). Cabbage yields vary from one treatment to another

DISCUSSION

The recovery rate was maximal for all treatments, showing that the organic fertilizer inputs had no effect on this parameter. This situation would be justified by the selection of vigorous plants operated before transplantation. Studies, notably those of (Ojetayo *et al.,* 2011). And (Kimuni *et al.,* 2014). Showed that the recovery rate of cabbage, Chinese cabbage, was similar on soil without fertilization, fertilized with NPK 15-15-15 and various composts of manure.

In addition, higher effects of organic fertilizers were observed on the rest of the growth parameters compared to the control without fertilization. Nitrogen fertilization affects all parameters contributing to good yield (Doberman and Fairhurst, 2000). *A. filiculoides* and *A. caroliniana* allow good growth of cabbage. This is possible thanks to the permanent provision of Nitrogen assimilable to the cabbage plant by Azolla as shown by the works of (Maria Andrea and Paul, 2007) and this could be related significant mineralization of soil nutrients from Azolla-treated planks. The fern fixes and accumulates nitrogen which it transforms into ammonia thanks to the heterocysts contained in the cells of Azolla which sites are fixing the atmospheric nitrogen (Dupuy and Rabeharisola, 1987). This form of assimilable nitrogen and other minerals will be released gradually, which can ensure their availability at the time of actual need by the plant and allow its growth. In terms of the number of leaves and the spread of cabbage plants subjected to Azolla treatment, the superiority would also be related to a large amount of the nutrients available to the plant. Organic matter (Azolla) has a significant amount of elements such as nitrogen and phosphorus, which are essential for the growth and development of plants and act immediately on the development of foliage and on the production of plants in cultivation (Brasset and Couturier, 2005).

The compost provided did not achieve high plant growth compared to Azolla probably because of the low availability of their nutrients, particularly nitrogen. The difference in vegetative growth of cabbage treated with compost with cabbage treated with Azolla is explained by the fact that the nitrogen of the compost was mainly in organic form and its mineralization during the growing season was probably not sufficient to fill the nitrogen requirements of cabbage plants. The effect of compost through organic matter on vegetative growth, demonstrated by the results obtained in this trial, are confirmed by other authors such as (N'Dayegamiye *et al*., 2004). Which recorded significant increases in growth parameters and barley yields compared to the control. The witness without fertilization at the smallest growth. The poor performance of cabbage plants observed on control planks can be attributed to the physico-chemical conditions of the soil (Mukalay *et al.,* 2008); (Kasongo, 2008).

The fertilization is therefore correlated with the vegetative parameters of the plants which is consistent with the results of (Kimuni *et al.,* 2014). Who recalled that the growth and yield of Chinese cabbage have been significantly improved following the contributions of different doses of NPK composts and mineral fertilizers. Finally these studies show that a good growth of the cabbage is one of the conditions sine qua non for a very good yield of the cabbage. The results show that under the organic fertilization of cabbage with Azolla caroliniana and Azolla filiculoides, the number of apples obtained is considerable. The yield of cabbage leaves increases by 13.81 t / ha, ie 24% for Azolla caroliniana, 11.45 t / ha or 20% for *Azolla filiculoides* compared to the control without fertilization. This is in line with the results of (Diara, 2000) which showed that the use of *A. pinnata* increases the yield of rice grain in Senegal. These results are also consistent with those of (DJOGBEDE *et al.,* 2012). Who concludes that the use of *A. pinnata* in rice cultivation in Benin, allows the acceleration of the growth, the foliation, the setting up of the thalli and panicles then the increase of the yield of the rice. Indeed, according to (DJOGBEDE *et al.,* 2012). Azolla can therefore be a valid substitute for chemical fertilizers that are

very expensive and not accessible to poor farmers. The results of this study also show that under the organic fertilization of compost the number of apples obtained is high and the yield of cabbage apples increases by 8% or 4.57 t / ha compared to the control. The positive role of organic matter through compost on yield, demonstrated by the results obtained in this test, are confirmed by other authors as (N'Dayegamiye *et al.,* 2004). Which recorded significant increases in barley yields compared to the control, after application of mixed sludge and manure. This situation is justified by the incorporation of hen dung, recognized rich in phosphorus during composting. Indeed, phosphorus is an important element for fruit production (FAO, 2000). This corroborates the results of (Useni *et al.,* 2014). On the cultivation of Chinese cabbage after application of chicken manure composts.

Conclusion

Evaluation of the effects of Azolla and compost on cabbage crop yields. Agronomic traits such as picking density, leaf length, width and spread, leaf number, and apple yield per hectare were measured. All the results obtained highlighted the effect of Azolla and compost on the development and growth of cabbage. Morphological parameters were monitored and studied to elucidate the action of different biofertilizers on cabbage plants. The results obtained illustrate the beneficial effects of Azolla produced on the cabbage crop, as well as on the morphology of the plant. The analysis of the results of the various treatments carried out on the parameters studied showed that Azolla recorded appreciable increases compared to the compost and to the control. Thus, these applied treatments have a very highly significant influence on the growth parameters studied. The best results were recorded at Azolla filiculoides and Azolla caroliniana. The permanence of assimilable nitrogen around the plants and the improvement of the physicochemical and biological properties of the soil have influenced the nutrient supply of the plants. Our work asserts that Azolla is able to fertilize the soil by its richness in essential element for the plant and able to influence the vegetative growth and to increase the yield of the cabbage. So in order to achieve a sustainable and conservative agriculture of biodiversity, we plan to know the effects of Azolla on varieties of cabbage grown in Côte d'Ivoire, the behavior of other crops such as the cultivation of chilli, salad against the fertilizing quality of Azolla, extend the study to other agro-ecological zones.

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