Student profile

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Bio data

October 2018- Present **Visiting Msc Student** for Msc mobility program in the Department of Crop Production and Landscape management of Ebonyi State University (Nigeria)

2016: **Bachelor Degree** in Crops production. School of Crops Sciences, Faculty of Agronomic Sciences, University of Abomey-Calavi.

RESEARCH INTERESTS: Conservation and utilization of plant genetic resources, Bioinformatics, Genetics and Plant Breeding, Ethnobotany Plant physiology, Horticulture

WORK EXPERIENCE

September 2018 - October 2018 **Assistant:** Bioversity international, West and central Africa Office Cotonou, Benin

Tasks: Interviewing farmers and organizing the workshops

November 2017-Present **Technician**: Laboratory of Genetics, Horticulture and Seed Sciences (GBioS), University of Abomey-Calavi

Responsibilities: Assistant of PhD and MSc researchers

November 2016- December 2016 **Technician**: Laboratory of plant physiology and study of Environmental stresses, Faculty of sciences and technology

Activities: Assessment of salinity stress tolerance in *Amaranthus cruentus* and *Solanum Lycopersicum* (Tomato) cultivars from Benin.

Training and Workshop

April 2017-May 2017:

Oil palm breeding: Traits of interest, basics concepts for breeding, breeding methods, interspecific breeding and Marker Assistant Selection (MAS)''. Agricultural Research Centre of Perennial Plants (CRAPP), Faculty of Agronomic Sciences, University of Abomey-Calavi, Benin.

June, 2019:

Workshop in Grant Proposal Writing. Facilitated by West Africa Farmer-to-Farmer (F2F) Program of Winrock International Volunteer Technical Assistance (VTA), Ebonyi state university (EBSU), Nigeria.

GRANTS AND AWARDS

2011 Partial Scholarship of Benin Government for Bachelor studies at the University of Abomey-Calavi

2018 Msc scholarship Mobility at the University of Ebonyi State (Nigeria), Mobreed (Mobility for breeders in Africa) founded by the European Commission to enhance training and research mobility for novel crops breeding in Africa.

Crops of interest:

Amaranthus cruentus commonly called Amaranth or green.

Summary of proposal

Background

The ability to conduct crosses at a large scale with little effort is essential for the development and production of hybrid crop varieties (Veerappan et al., 2014). Sometimes crosses between two different plants fail due to morphological or physiological barriers (Van de Wiel et al., 2010). Such difficulties are identified in *Amaranthus cruentus*, a species in which crosses are more difficult than in other crops (Stetter et al., 2016b). According to Stetter et al. (2016b), Further

studies may be carry out to the figure out the adequate temperature and treatment duration for high success rate which are not yet identified. All this will facilitate future hybridizations planned in breeding programs for intra-species hybridization within *A. cruentus* and inter-species hybridization between *Amaranthus cruentus* and *Amaranthus dubius* to evolve a new population upon which selection can be made (Adeniji and Aloyce, 2013) for the development of varieties with high leaf yield (Dinssa et al., 2016). Besides, attention is progressively focus on increasing its nutrient content such as iron and zinc leaves content to reach high source thresholds per nutrient trait by Codex Alimentarius (1997) definitions: 4.2 mg/100 g Fe, 90 mg/100 g Mg, 300 mg/100 g Ca, and 4.5 mg/100 g Zn, by fresh weight basis (Byrnes et al., 2017). According to Achigan-Dako et al. (2014), interspecific and intraspecific variations in the genus could be exploited to improve macro and micro-nutrients in *Amaranthus cruentus* mostly for vitamin A, iron and zinc contents. Although, intraspecific variation for nutrient content was assessed, the former ones were focus just on very few number in the germplasm available.

Breeding objective

The main objective of this study is to develop relevant crossing methods and high nutrient content cultivars for *A. cruentus* in order to mitigate food and nutritional insecurity in West Africa.

The specific objective of this research is twofold:

1. Establish a protocol to enhance success in crossing methods in *Amaranthus cruentus*; and
2. Assess the genetic variability and the performances of *A. cruentus* accessions for leaf carotenoids, iron and zinc concentration and leaf yield related-traits.

Establish a protocol to enhance success in crossing methods in *Amaranthus cruentus*

Plant material

Four accessions (parents) will be selected based on variability for stem colour and earlier flowering initiation. Stem colour is a qualitative trait in *Amaranthus cruentus* with homozygous recessive (rr) for green stem and heterozygous dominant for red stem (RR or Rr) (Kulakow and Jain, 1985).

Mating design and Experimental design

The trial will consist in crossing four phenotypes without reciprocal mating. Two Red stem genotypes (Rg) and three two stem phenotypes (Gg) crossing will result in 4 couples [C= (Rg x Gg)]. The Green stem phenotype will be considered as female parents during the crossings.

Table 1: Mating design

	Rg1
Gg 1	C1
Gg 2	C2
Gg 3	C3

For this purpose, a completely randomized design will be used in which couples under several treatments (Table 2) randomly assigned with 3 replications. These treatments are resumed in the next table as follow:

Table 2: Factors and modalities arrangement

Factors	**Water temperature**	**Duration**	**Crosses**
Modalities	t_1 = 45°C	d_1 = 10 min	C_1, C_2, C_3
	t_2 =47°C	d_2 = 15 min	
	t_3 = no water (control)		

According to García-Yzaguirre and Carreres (2008), warm water emasculation is done at flowering initiation in dipping inflorescence in the warm water buckets. Each emasculation treatment will be repeated at 3 days intervals during trial period on the same experimental unit. All the treatments are arranged in a CRD (Completely randomized design). Thus the total number of experimental unit is 39 in a replication giving 195 for all the experiment. Each experimental unit contains two genotypes (which will be crossed) which will be put in bag filled with sterilized soil (soil: sand: poultry manure) at 3: 2: 1 (v/v/v), respectively. The experiment Design has been

generated with R software indicating the number of experimental unit, replication and the random arrangement of all the treatments in the experiment.

Seeds of female parents will be harvested three weeks after crossing. Basal inflorescence seeds will be selected and for each cross 50 seeds will be randomly selected for germination. Seedlings will be observed for their stem colour and the number of hybrids will be scored (Hybrid progeny = red stem, selfed progeny= green stem). The differences between the crosses in terms of proportion of hybrids will be assessed using log linear model analysis through GLM function (generalized linear models) to reveal the best treatment or combination of treatments to achieve the highest success rate viz 100% of F1 hybrid progeny.

Estimate, heritability and genetic correlation for leaf carotenoids, iron and zinc concentration and leaf yield related-traits in *A. cruentus*.

Plant materials 1

In this study, a total of 25 *A. cruentus* accessions, of which 12 from the collection of the Laboratory of Genetic Horticulture and Seed Science (GBioS) and 13 genotypes from EBSU will be used

Experimental design 1

All genotypes of amaranth will be assessed in simple lattice design with three replications in two different seasons April-June and August-September. Each replication will contain 25 plots grouped in one block giving a total of two blocks and 50 plots/site. Each plot will consist of two rows of 30 plants spaced at 0.25 m within and between rows. Traits measurements will be done on 5 plants/ accession.

Figure 2: Experimental design (Triple lattice design)

Rep 1

Acc18	Acc4	Acc19	Acc10	Acc22
Acc5	Acc23	Acc25	Acc3	Acc16
Acc6	Acc1	Acc8	Acc24	Acc9

Rep 2

Acc8	Acc19	Acc21	Acc25	Acc2
Acc24	Acc10	Acc12	Acc3	Acc11
Acc1	Acc4	Acc14	Acc23	Acc13

Acc20	Acc13	Acc2	Acc11	Acc7
Acc15	Acc14	Acc21	Acc12	Acc17

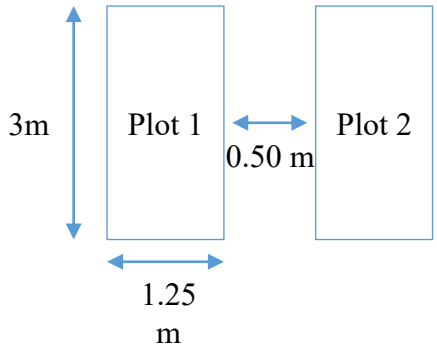
Acc9	Acc22	Acc17	Acc16	Acc7
Acc6	Acc18	Acc15	Acc5	Acc20

ABAKALIKI Local 1 (Acc7) is the check in the experimental design.

Manure application, pests control and others will be performed with respect to cultural practices used by (Shukla et al., 2006; Sogbohossou, 2014).

Experimental unit

Spacing of 0.5m, each plot will be divided in three rows each one. Plot size for each accession is sized 3.75 m^2 with 0.25 m as row to row distance and 0.25 m as plant-to-plant. The replication will be spaced of 1m. In considering the interval between the plots, the total area required is 500 m^2 (20.5 m x 24.39 m).



Collected bulked soil samples will be analyzed for chemical soil health indicators in a Laboratory of Soil Sciences. The following soil health indicators and samples will be measured: pH, electrical conductivity, nitrate nitrogen (NO_3), organic carbon, available potassium, Iron, and Zinc.

Morphological related measurements as plant height, number of leaves, leaf area, and number of branches, stem diameter and flowering date are collected. At harvest (3 weeks after transplanting) a pruning is made on some plants to determine the yield parameters by measuring the total yield weight and the marketable weight edible biomass.

At this stage, a quantitative variation for leaf related-traits, carotenoids, iron and zinc content will be assessed; therefore, the average and the coefficient of variation for each trait (nutrient content, leaf related-yield traits) will be estimated in using the descriptive statistics with respect to the

number of plants measured. The data will also be subjected to an analysis of variance (ANOVA). Afterwards, the mean of data will be normalized and subjected to multivariate analysis using the procedure of principal component analysis (PCA) to assess the patterns of variation. Afterwards, the broad sense heritability will be estimated for each trait of interest with this formula:

$$Heritability = \frac{Genotypic\ variance}{Phenotypic\ variance}$$

with $Phenotypic\ Variance = Genotypic\ variance + Error\ MS$

and $Genotypic\ variance = \frac{Genotype\ MS - Error\ MS}{Replicate}$

The expected genetic advance (GA) and percentage of GA will be calculated with this Formula: (GA) = i. σ*p*. h^2 (with i = selection intensity)

h^2= *Heritability,* σ*p= Phenotypic Standard deviation*

GA (%) = GA x 100/X

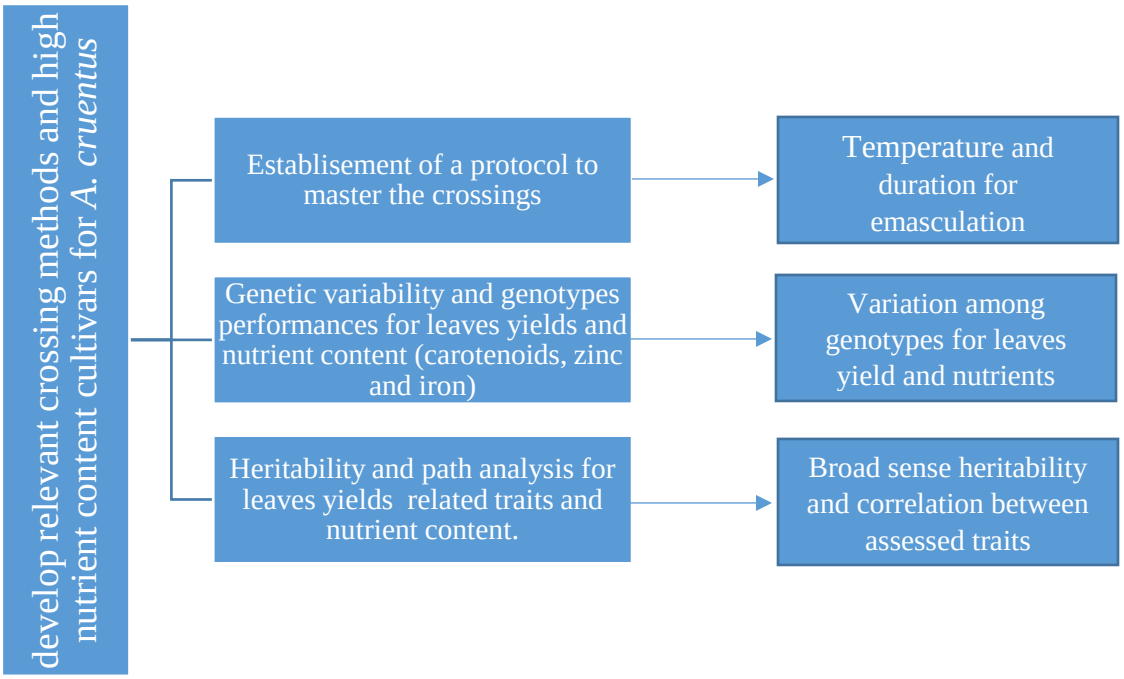
X= Grand mean of the traits

The estimates of covariance between pairs of traits will be derived from a multi-trait analysis of covariance and subsequently the corresponding correlation also called genotypic correlation r_g Bernardo (2002) will be calculated:

$$r_{gxy} = \frac{\sigma_{xy}}{\sigma_x \sigma_y}$$

Where r_{gxy}= genetic correlation coefficient between trait x and trait y; σ_{xy}= genotypic covariance between trait x and trait y; σ_x σ_y= genotypic standard deviations of trait x and trait y, respectively.

A graphic abstract of the research proposal



Useful pictures

Photo 2

Photo 1: Plants one week after transplanting

Photo 2: Marketable weight of 5 plants/ bed

Photo 3: two months old plants grown in pots for crossing purpose

Personal comment about the Mobreed opportunity

This mobility is a very nice opportunity for postgraduate students who are endeavoring themselves so that they might be successful in the beginning of their research careers.

Progress report

From February to July, two activities have been conducted regarding the achievement of the two aforementioned research objectives.

Trial location

The first one was to deal with the assessment of 25 accessions of *Amaranthus cruentus* in the experimental farm of the college of Agricultural Sciences (CAS) of Ebonyi State University (EBSU) in the South-eastern region of Nigeria at 6°151N latitude and 8°100E longitude. This first seasonal trial was carried out from (April-June) which was covered with an average temperature of 27.66°C with a mean daily maximum of 33.33°C and daily minimum of 22.67°C. Abakaliki city has a bimodal rainfall with the main rainfall pattern occurring from April to October with its first peak in July and the second occurs in September (Diagi and Nwagbara, 2018). The total amount of rainfall received during this season was 152.86 mm.

A soil analysis was performed, showing that the site has a sandy loam soil with others parameters as follows: pH= 5.8, ECEC= 7.828, %N= 0.14, %P= 25.5, %K= 0.164.

Agronomic practices

The evaluation of 25 accessions from Nigeria, Benin, USA, Mexico and India was made in a Simple lattice (5*5). At first, all accessions including the check one were germinated in a nursery made on the soil and the seedlings sprung up, were trans-planted at 4 weeks old. Each replication contained 25 plots grouped in five blocks of five accessions. The replications were spaced by 1 m and 0.5m between plots with each plot consisted of three rows of 10 plants spaced at 0.25 m within and between rows. Poultry manure was applied just after beds diggings at a rate of 10t/ha, 3 days before transplanting. Lambda cyhalothrin 2.5 EC(Lambdacal) was applied to control caterpillars and Mancozebe combined with metalaxyl to control fungi.

Data collection

As for data collection, 15 traits were measured on five plants randomly chosen in the middle of each plot which resulted in 10 plants measured per accession. These traits include plant height (PH), Number of leaves (NL), width of 3 leaves randomly selected (WL), Length of 3 leaves randomly selected (smallest, medium, largest): (LL), Number of branches per plant (NB), days to 50% flowering: Number of days after transplanting when 50% of the plants in a plot have opened flowers, marketable fresh vegetative yield (Kg/plot): total marketable vegetative yield harvested/ plot (MFVY), total yield (TY), spread canopy (SC), Number of leaves (2 weeks after cutting: NL1), Number of branches (2 weeks after cutting: NB1). Besides, an elemental micronutrient analysis for carotenoids, iron and zinc content is ongoing at Soil Science and Environmental

Management Laboratory, EBSU, on foliar subsamples of three fresh leaves randomly selected on five plants from each line.

As for the second trial (crossings), the nursery has been made at the beginning of this month and 195 pots have been filled with top soil and arranged in a complete randomised factorial design.

Table: Summary of the half-yearly activities

Activities	February				March			
	1st week	2nd week	3rd week	4 th week	1st week	2nd week	3rd week	4 th week
Proposal Validation (minor comments and modification, adjustments and some clarifications)								
Permission to start my project								
	April				May			
	1st week	2nd week	3rd week	4 th week	1st week	2nd week	3rd week	4 th week
Field preparation (cleaning, digging beds)								
Implementation of the Nursery								
Resowing (unviable seeds and low								

germination)								
Soil analysis								
Usual practices for nursery								
Filling of 195 plastic bags with top soil for the second experiment								
Poultry Manure applied and Transplantation in the field								
Usual practices (watering, weeding and hoeing)								
	June				July			
	1st week	2nd week	3rd week	4 th week	1st week	2nd week	3rd week	4 th week
Data collection of quantitative traits (leaves yields and relatives traits)								
Leaves sampling for								

carotenoids , iron and zinc analysis								
Data collection after cuttings								
50%Flowering data collection								
Nursery of the 2nd trial (crossings)								