N2Africa Podcaster no. 32

July and August 2015

Introduction

We devote this Podcaster to updates from all of the N2Africa PhDs. Some of the candidates started their studies during the first phase of N2Africa and are close to completion. Others just started their research last September and are still setting up their initial experiments and finalizing their proposals. The topics range from molecular studies with rhizobium to field agronomy, adoption studies and nutrition. We felt it was time for the next generation of nitrogen fixation researchers to share some of the excitement of their findings with you. I think you'll agree that George Mwenda

wins first prize for the incredibly beautiful photos of nodules shown below on this page!

We close with a couple of news items including a new collaboration within the EU with other research institutions. The next issue of the Podcaster will focus on public-private partnerships within N2Africa - so please send in your stories straight away.

Ken Giller

Evaluation of competitiveness for nodulation of Phaseolus vulgaris L. in Kenyan rhizobial strains

Beans nodulate promiscuously with rhizobia. The lack of specificity may be advantageous in certain circumstances but it generally poses a challenge to attempts to successfully inoculate beans. The inoculant strain, when applied. has to compete with indigenous strains for root infection sites and outcomes are dependent on numerous variables. In addition to identifying rhizobia nodulating beans in Kenya and assessing their nitrogen fixing potential (update appeared in Podcaster 25), my research has used a dual marker system to study the competitiveness of indigenous strains against the major commercial inoculant strain. Rhizobium tropici CIAT 899.



marked chromosomally with (blue) and NAK 104-pGM1 (magenta). (**D**) Nodule from a double infection event. nodules after inoculation with pGM1 strain. (C) Double stained nodules after inoculation with CIAT 899-gusA gusA gene while Kenyan

strains were marked with *celB* gene borne on a stable broad-host-range plasmid, pGM1. gusA gene on CIAT 899 was either under a constitutive promoter (ptac) or under a symbiotically active promoter (pnifH). The gusA bearing CIAT 899 strains were tested for characteristics such as growth rate and nitrogen fixation and did not differ from the wild type. Similarly, Kenyan strains with pGM1 were not different from parents, symbiotically or phenotypically.

lation of P. vulgaris 'KK08' in glasshouse co-inoculation experiments (Figure 1A, 1B, 1C). There was no correlation between effectiveness of the strains and their competitiveness. Single and double occupancy events were observed (Figure 1D), with stressful growth conditions seen to promote dual occupancy rates. Experiments looking at impact of different inoculation ratios on competition outcomes are ongoing. Thesis is expected to be submitted for examination by the end of the year.

Using the dual marking technique, CIAT 899 was found to out-compete a majority of 38 Kenyan strains for nodu-

George Mwenda, Murdoch University, Australia



Progress Report on PhD Studies Aliyu Anchau; Topic: Exploring the Genetic Diversity of Groundnut-nodulating Rhizobia in Moist and Dry Savannas of Nigeria for increased Symbiotic Nitrogen Fixation and Productivity Published on N2Africa Facebook on July 23rd

Aliyu Anchau, Abdullahi enrolled for his PhD studies in February, 2013 on the above topic. *Arachis hypogaea* L. root nodules were initially sampled from uninoculated plants in over ninety (90) farmers' fields from the Northern Guinea and Sudan savannas of Nigeria during the 2013/2014 growing season. Thirty two (32) rhizobia strains were successfully isolated from the root nodules, authenticated and tested for effectiveness in glasshouse experiments under axenic N-free conditions. The treatments include uninoculated controls without inorganic nitrogen applied, positive N controls (10



g L⁻¹ N applied as KNO₃), *Bradyrhizobium* Figure 1: Percent dry weight accumulated by SAMNUT 24 groundnut genotype with the newly sp. strain NC 92 as industry standard isolated Nigerian indigenous rhizobia strains in relation to the positive N control (10 g L⁻¹ N applied as KNO₃)

reference strain and the newly isolated $applied as KNO_3$) strains. Top dry matter yields and N concentrations in tops were determined. The percentage dry matter yield and N accumulation of each strain were calculated relative to the positive N control. These results indicated that the strains had varying effectiveness, with some strains performing similar to NC 92 and some higher than both NC 92 and the positive N control (Figure 1).

Positive responses of groundnut genotypes SAMNUT 22 and SAMNUT 24 were observed in field experiments, testing some of the strains in the Northern Guinea and Sudan savannas of Nigeria during the 2014/2015 growing season. Two best performing strains from the field experi-

ment along with other two promising strains are now being tested under similar field conditions during the 2015/2016 growing season for confirmation of the results. Morphological characteristics and RPO1 fingerprints indicate genetic diversity among the strains. Further molecular characterization, N calibration and root nodule morphology studies of the strains are in progress. Careful selection from these indigenous strains could fill the vacancy of effective local inoculant strains for groundnut in Nigeria and similar environments. The studies shall hopefully be concluded in February, 2016.

Aliyu Anchau, Murdoch University, Australia



Glasshouse experiment, testing some of the newly isolated Nigerian strains of rhizobia, along with NC 92, +N (10 g L⁻¹ KNO₃) and –N (Uninoculated control) on SAMNUT 22 and SAMNUT 24 groundnut genotypes at 5 weeks after sowing

Field experiment, testing some of the newly isolated indigenous Nigerian rhizobia strains (SBG 234, SNN 343 and KBU 26) on SAMNUT 24 and SAMNUT 22 groundnut genotypes at the Experimental farm of the Institute for Agricultural Research (IAR), Ahmadu Bello University, Samaru, Zaria, Nigeria during the 2014/2015 growing season: (left) 02/08/2014 at 4 weeks after sowing (right) 30/9/2014 at 12 weeks after sowing

Genetic and Symbiotic Effectiveness of Indigenous Rhizobial Strains and Strategies to Maximize the Contribution of Biological Nitrogen Fixation on Soyabean in Mozambique

Amaral Machaculeha Chibeba started his N2Africa PhD project in August 2012 at Londrina State University in Brazil. In summary, he characterized rhizobia isolated from field-grown soyabean nodules brought from Mozambique,

conducted field trials for testing symbiotic effectiveness of elite *Bradyrhizobium* strains on soyabean in Brazil and Mozambique and conducted a greenhouse experiment to evaluate the effect of co-inoculation of soyabean with



Genetic and symbiotic characterization of rhizobial strains

First, soyabean nodules from 15 locations in Mozambique were brought to Brazil. Amaral obtained 105 rhizobia isolates from these nodules. The DNA extractions, genomic profile by BOX-PRC, and 16S gene sequencing of the isolates pointed towards a large genetic diversity among the isolates. In addition, groups indicative of putative new rhizobia species were identified.

To assess the symbiotic effectiveness, Amaral first evaluated all 105 isolates on their effects on the Brazilian soyabean cultivar BRS 133, a non-promiscuous commercial genotype. He compared the effectiveness of the isolates with five elite Bradyrhizobium strains. Four of them are currently used in commercial inoculants in Brazil¹ and one strain is broadly used in commercial inoculants in Africa and also in other experiments of the N2Africa project². This first trial yielded 13 best isolates, which in turn formed the basis for another greenhouse experiment; this time with two promiscuous soyabean cultivars from Africa, TGx 1963-3F and TGx 1845–10E (Photo). If all goes according to plan, the results with the strains from Mozambigue should be analysed by October 2015. As a next step, the elite strains from Mozambique that have already been identified will be tested in field experiments in Mozambique in the next crop season.

Technology transfer

In another study the objective was to verify if it is possible to transfer technology developed in Brazil with BNF straight to Mozambique. Amaral developed field trials with the same strains in both Brazil and Mozambique (2013/2014 and 2014/15 crop seasons): five in Brazil and eight in Mozambique. In Mozambique the trials were conducted with the support of Dr. Steven Boahen from IITA. The treatments consisted of the same elite *Bradyrhizobium* strains



Figure 1: Amaral Chibeba and Mariangela Hungria visiting the greenhouse experiment of evaluation of strains in soyabean promiscuous cultivars

as in the greenhouse experiments. Some of the Brazilian strains, such as the SEMIAs 5079 and 5080, had an excellent performance in Mozambique. This shows the feasibility of transferring technology from Brazil to a country in Africa with similar edaphoclimatic conditions.

Precocity of nodulation

Another interesting topic to evaluate is the effect of co-inoculating soyabean. What happens to nodulation when you mix rhizobia and plant growth promoting rhizobacteria (PGPR)?Amaral carried out a greenhouse experiment in which he inoculated soyabean with both *Bradyrhizobium* and *Azospirillum*. It turned out that co-inoculation promotes precocity of nodulation. The results of this experiment have been published in the American Journal of Plant Sciences 6:1641-1649, 2015. Co-inoculation has potential to maximize biological nitrogen fixation in soyabean.

Amaral Chibeba (and Mariangela Hungria), Londrina State University, Brazil

¹ Bradyrhizobium elkanii strains SEMIA 587 and SEMIA 5019 (=29w), *B. japonicum* SEMIA 5079 (=CPAC 15) and *B. diazoefficiens* SEMIA 5080 (=CPAC 7)

² B. diazoefficiens USDA 110

Does the interaction of the indigenous and exotic rhizobia in contrasting Zimbabwean soil conditions result in superior isolates worthy of the inoculant strain title?

Mazvita Chiduwa is pursuing her studies for a PhD at Murdoch University. She commenced in July 2012 under the supervision of Ravi Tiwari, John Howieson, Julie Ardley, Graham O'Hara and Paul Mapfumo.

Soyabean is grown throughout Zimbabwe and is recognised as a strategic crop because of the myriad of benefits accruing. Although indigenous rhizobia have been isolated, distribution is highly erratic and inoculation is critical to obtain economic yields in Zimbabwe. Currently and since the 1960's inoculation is achieved with the elite soyabean inoculant *Bradyrhizobium diazoefficiens* MAR 1491 (=USDA 110T) and MAR 1495 (=USDA 122) supplied as inoculant sachets by the Soil Productivity Research Laboratory, where Mazvita has worked since 2005. These exotic strains have been reported to exhibit limited saprophytic competence while indigenous soyabean nodulating rhizobia are hypothesised to display superior environmental adaptation.

In the present study the diversity of 138 authenticated root nodule bacteria obtained from six sites in Zimbabwe was evaluated. Three of the sites were soyabean-breeding facilities which use research led recommendations including fertilizers, their rates and breeder's seed and the other three were smallholder-farming communities that generally





Figure 1: Species distribution of rhizobia recovered from two contrasting farm management systems

practice low external input agriculture including reduced fertilizer rates and manure, to account for the dualistic farming system in Zimbabwean agriculture. Isolates were trapped with soyabean under glasshouse conditions using soils obtained from the breeding facilities while those from communal farmers were isolated from soyabean nodules obtained directly from the fields.

The phylogeny of the isolates has been evaluated. The isolates have been evaluated for their nitrogen fixation potential under glasshouse conditions. Host range analysis has been carried out for representatives of the species identified.

All 138 isolates were slow growing and formed either dry colonies (%) or wet mucoid (%) colonies. Based on multilocus gene sequence analysis (MLSA), the majority (61%) of the strains recovered belong to *Bradyrhizobium elkanii* species whilst *B. japonicum* accounts for 21%. The newly identified species recovered from soyabean in Canada, *B. ottawaense*, was also recovered although it only accounted for 5% of the isolates. Only 13% of recovered isolates identified with the previously inoculated B. diazoefficiens strains. Interestingly, none of these were recovered from research facilities, suggesting that the higher input systems promote indigenous rhizobia at the expense of the elite, exotic strains. Using host range analysis across fourteen legumes in the glasshouse, a nodulating pattern across four legumes (G. max, P. vulgaris, C. juncea and V. radiata) distinguishes the four species. A PCR-based multiplex method to distinguish the four species based on their recA gene sequence has also been established. A further 25 root nodule bacteria have been isolated from trap soyabean in soils obtained from two virgin soils and one agricultural field that has not been inoculated to understand the phylogeny of the indigenous soyabean nodulating rhizobia. The species identity of the indigenous isolates will be established using the multiplex method.

All isolates were compared for their nitrogen fixation efficiency under glasshouse conditions. The inoculant strain MAR 1491 was used as a standard against which all strains were compared. The top 27 isolates were compared in a further experiment. The most efficacious isolates are highly similar to *B. diazoefficiens*. *B. elkanii* also generated high top dry weights in the inoculated plants and only one strain from the *B. japonicum* was comparable to the inoculant strain. Finally six strains were compared for their nitrogen fixation across three Zimbabwean soyabean cultivars Mhofu, Status and Pan 1867. The results of this experiment will be used to make recommendation for inoculant strains to be used in Zimbabwe.

Mazvita Chiduwa*, Murdoch University, Australia

* Mazvita won the best translational research poster prize at last year's Poster day held in the Veterinary and Life Sciences School. To this poster we link here.

Impact of Improved Cowpea Technology on Women Farmers in Southern Borno State, Nigeria.

This research is on cowpea (*Vigna unguiculata*, (L.)) which is an annual leguminous crop indigenous to Tropical Africa. Women farmers mostly grow cowpea as a food security crop and it is an important source of protein and income for many of them. The high protein content of cowpea, its adaptability to different types of soils, drought tolerance, ability to improve soil fertility and prevention of soil erosion makes it an important economic crop. In Africa, the women farmers produce, process and sell snacks made from this nutritious legume. With a growing market for cowpeas both within and outside the country, women can be encouraged to increase their production by using improved cowpea technology.

In 2004 IITA implemented the PROSAB project (Promoting Sustainable Agriculture in Borno State) in Borno State. The project promoted the use of improved agricultural technologies, including cowpea for sustainable agricultural production. The current PhD study aims to assess the impact of the improved cowpea on women farmers' income and food security in the study area (Southern Borno State), ten years after implementation of the project. Specifically, the study will evaluate the changes in income as a result of adopting improved cowpea and the impact of the improved technology on the food security status of the respondents. The determinants of technical efficiency of the women cowpea farmers will be estimated and the constraints associated with the use of improved cowpea will be identified.

The proposal seminar has been prepared and presented. Currently, I am preparing to train the enumerators who will assist in collecting data from the respondents and then start the field survey (data collection).

Binta Ali Zongoma, University of Maiduguri, Nigeria.



Studying the benefits of intensifying common bean cultivation on smallholder farms in the Northern Highlands of Tanzania

Intensification of common bean (Phaseolus vulgaris L.) cultivation on smallholder farms in the Northern Highlands of Tanzania is the main focus of my PhD study. This study is prompted by the low yields $(0.3 - 1 \text{ t ha}^{-1})$ obtained by smallholder farmers, despite the large prospective productivity of improved varieties of common bean (3 t ha-1) in Tanzania. The study aims to unravel the contributions of genetic, management and environment related factors to common bean yield and nitrogen fixation. It also includes cultural, social and economic analyses of growing common beans. In the study I compare the ability and benefits of beans on improvement of soil fertility and bean yields through intercropping and/or rotations. The knowledge from this study will contribute to sustainable common bean production by smallholder farmers in the Northern Highlands of Tanzania. Thereby it contributes to one of the objectives of N2Africa phase II Project.

I installed experiments in the Lower, Middle and Upper Zone in Hai district, located in the Northern Highlands of the country, Kilimanjaro region. There are 11 treatments involved in the study, looking at different rotational and intercropping structures of two different bean varieties and maize. The two bean varieties are the improved Lyamungu 90 and the local Mkanamna (Plate 1).



Plate 1. (a) Improved bean seeds (*Lyamungu 90*) and (b) Local bean seeds locally known as *Mkanamna*

The experiments have four replicates of each treatment, which each plot measuring 5 m × 3.2 m. Under sole and intercrop, there were 85 maize plants/plot equivalent to 41,666 maize plants/ha. Under sole and intercrop there were 4 and 9 rows with 459 and 204 bean plants/ plot, respectively, and this planting spacing constitutes an equivalent population of 250,000 bean plants/ha. The intercropping of maize/bean in the experimental plots was simultaneously additive (1/1), which is the common practice of farmers in the district (Plate 2).



Plate 2. Appearance of (a) Intercropped bean and (b) Sole bean as on 19th April 2015 (At 23 days of age) in the Lower Zone

The following describes the experiment in the Lower Zone. Two months after sowing, the plants were well established and covered most of the land with their canopy (Plate 3). The data collected were emergence (%), weekly ground coverage (%) and plant height, nodulation (number and colour), total harvest weight, stover weight, number of pods per plant, number of grains per pod and weight of 100-seeds (data not presented in this document).



Plate 3. Appearance of maize and bean plants as on 26th May 2015 (At 30 days of age) in the Lower Zone

The bean variety Lyamungu 90 flowered 30 days after sowing. The local beans flowered 15 days later and concurrently formed the pods, hence escaped the risks of flower abortion due to shortfall of rain. All bean plants were ready for harvest by 26th June 2015 (Plate 4a,b) but harvesting was done on 4th July 2015 (Plate 4c) and some of the seeds kept (Plate 4d).



Plate 4. Bean plants at their last stages of being in the field and harvest in the Lower Zone: (a) 26th June 2015: *Lyamungu 90* bean ready for harvest, (b) 26th June 2015: Local bean ready for harvest, (c) 4th July 2015: Harvesting and data collection (bean) and (d) Local and Lyamungu 90 bean seeds

Eliakira Kisetu Nassary, Wageningen University, The Netherlands



Update on PhD research – Adoption and adaptation of improved climbing bean technologies in Uganda

Esther Ronner is research assistant employed by N2Africa at Wageningen University and is conducting her PhD studies within the project.

In June, two students from Makerere University (Florence Ajio and Ezakiel Muranda) started the collection of data among farmers with climbing bean adaptation trials in eastern Uganda (Kapchorwa District) and southwestern Uganda (Kabale and Kanungu Districts). The two students are doing an internship in the context of my PhD research on the adoption and adaptation of improved climbing bean technologies in Uganda. The students use the 'field book for adaptation trials', but work with additional questions on the cultivation of climbing beans including staking methods, materials, densities, length, etc. We aim to get insight in the way that different types of farmers cultivate climbing beans, and to what extent farmers apply the management practices that are shown in the demonstration trials on their own field.

Some first findings from this season are that 124 out of the 148 farmers that were interviewed (84%) planted the adaptation trial package. A comparison between management practices applied in the demonstrations and on the adaptation trial plots showed that less than half of the farmers in Kabale and Kanungu Districts grew their beans as sole crops (Table 1), while all demonstrations were planted as sole crops. In Kapchorwa District this was even less than 20%. Most farmers intercropped their beans with banana, coffee or maize. Row planting was a common practice in Kanungu, but only applied by half of the farmers in Kapchorwa.

Table 1: Implementation of demonstrated climbing bean technologies in adaptation trials by farmers in Kabale, Kanungu and Kapchorwa Districts in Uganda, first season of 2015

Practice in	% of farmers applying practice in adaptation trials in:					
demonstration trial:	Kabale (n=14)	Kanungu (n=31)	Kapchorwa (n=75)			
Sole cropping	43%	44%	17%			
Row planting	79%	94%	50%			
Single stakes	100%	100%	97%			
Tripods*	0%	0%	3%			
Sisal/ banana fibre strings	0%	0%	0%			

* Not demonstrated in Kabale and Kanungu sub-counties based on farmers' feedback last season

Staking is often mentioned as major constraint for the cultivation of climbing beans. Therefore, we introduced alternative staking material of sisal string and banana fibre ropes. None of the farmers used these materials on their own field, however, despite the fact that more than half of the farmers had seen sisal strings in a demonstration, and despite positive comments by half of the farmers who had seen the method. Farmers mostly mentioned that sisal was

more costly and more labour intensive than single stakes. Two farmers mentioned they had seen the strings after they had staked their own field. Another method demonstrated were tripods (three stakes tied together on top), which were used by only two farmers in Kapchorwa. These farmers mentioned that tripods gave more support to the beans than single stakes.

The average plant density in the demonstration trials was much less than in adaptation trials in Kabale, but comparable to or higher than in trials in Kanungu and Kapchorwa (Table 2). The high densities in Kabale were mainly caused by sowing a relatively large number of seeds per hole at comparable row and plant spacing as in the demonstrations. Farmers often use high densities to compensate for poor seed quality or risk of failure through pests.

Table 2: Average plant density, number of stakes per ha⁻¹, plants per stake and stake length in demonstration trials and adaptation trials in Kabale, Kanungu and Kapchorwa Districts in Uganda, first season of 2015

	Demonstration	Adaptation trials				
	trial	Kabale Kanungu		Kapchorwa		
No of plants per ha	160,000	290,000	155,000	120,000		
No of stakes per ha	40,000	33,500	29,500	33,500		
No of plants per stake	4.0	3.9	3.8	4.1		
Stake length (m)	> 1.75	1.55	1.81	1.76		

The fact that staking is still often considered as a constraint is shown by the lower number of stakes per ha in the adaptation trials than in the demonstrations and points to the need to improve the availability of stakes. The number of plants per stake was however comparable to what was shown in the demonstrations. Stakes in Kanungu and Kapchorwa were of considerable length, but smaller than recommended in Kabale.

What can we, as researchers, dissemination partners, extension officers, etc. learn from these 'adapations' from the demonstrated practices? First, most of the farmers grow beans in intercropping, largely due to land constraints. This implies that even though farmers may be aware of advantages of sole cropping, they often do not have enough land. We should therefore diversify our research to also explore the technologies that are most suitable for intercropping (varieties, spacing of beans and intercrops, nutrient requirements, etc.). Second, although staking is still a constraint, farmers feel most comfortable with single, wooden stakes and often consider this to be the most cost and labour efficient technology after all. Strings are often demonstrated as a good alternative to wooden stakes by other projects or in other countries, but their popularity in Uganda remains to



be seen. Increasing the availability of stakes through planting of multi-purpose (legume) trees around field borders may therefore be a better solution. We should therefore carefully consider how the technologies demonstrated by N2Africa 'fit' within current farming systems, how we could improve their fit, and how we could consider a range of technologies for farmers to pick their most preferred option from.

The two students are now back in the field to measure the harvest of the adaptation trial packages that farmers planted, and of the farmers' own climbing beans which were planted next to the N2Africa plot for comparison. This will also give us an impression of the performance of the different packages on farmers' fields, under different management practices. A third student (Collins Bugingo) visited farmers who participated in N2Africa in previous seasons, to look at their independent use of the improved climbing bean technologies offered by N2Africa.

I would like to take the opportunity here to thank Florence, Ezakiel and Collins for the interesting results that their work has yielded already!

Esther Ronner, Wageningen University, The Netherlands

Legume genotypes x rhizobium strains interaction in common bean and chickpea

Objective:

To screen effective, competitive and host broad spectrum of common bean and chickpea rhizobia for use in African soils

Main activities:

- 1. Screening effective strains from local collections using Modified Leonard Jars and/or isolating new strains using plant-trapping method and screening of the isolates for effectiveness
- 2. Factorial combination of host varieties with effective rhizobial strains to select both effective rhizobia and appropriate host variety in terms of effective symbiotic interactions. Analysing the patterns of G x G interaction in relation to geographic/genetic co-evolution and nod-C variants and compatibility with certain host genotypes.
- 3. Mix strains and test them against host varieties in greenhouse on soil containing pots and on Modified Leonard jars and genomically determine nodule occupancy (testing competitiveness)
- 4. Screened legume variety and rhizobium strain will be evaluated under field conditions to see their competitiveness and effectiveness

Method:

In order to screen effective rhizobium strains for G x G interaction study, two series of experiments were established. The first experiment included 20 common bean and 28 chickpea strains obtained from Hawassa University soil microbiology laboratory and Norwegian University of Life Sciences. Habru chickpea and Nasir common bean varieties were used for screening the strains. The experiment was conducted on modified Leonard jars in a lathhouse. 1 ml of each strain was inoculated to the base of the legume varieties' growing on the jars. Positive controls contained non-inoculated seedlings that were supplied with N-free nutrient medium and 0.5% KNO₃ and the negative controls contained non-inoculated seedlings that were only supplied with N-free nutrient solution. Commercial and type strains were included for comparison purposes. All the seedlings were completely randomized and supplemented with Jenson's N-free nutrient medium and grown in the

lath-house for forty five days. The seedlings were carefully uprooted and examined for nodulation and effectiveness. The second series of experiment was set with the same design as above but with additional number of strains and Natoli chickpea variety instead of Habru. The total number of strains that was set for common bean is 65 and 61 strains for chickpea.

Results:

The first experiment had quite astounding result for chickpea strains, i.e., all the strains were failed to elicit nodules on the roots of the Habru variety. The seedlings formed yellowish patterns on their leaves, which was unusual character. The roots of the seedlings were observed to be decaying or rotting. These symptoms were not due to nitrogen deficiency rather they might be due to high temperature of the lath-house, noticed during the period of the experiment. This phenomenon was noticed in the previous works (Wondeson and Endalkachew, personal communication). Therefore; it can be said that the high temperature of the lath-house was able to abort the inoculated strains. On the other hand, it means, the strains failed to perform under high temperature.

Common bean seedlings inoculated with the strains formed nodules on their roots. The Mexican strain (CH24-10) formed the highest number (91) of nodules (data not shown). When cut and examined, the nodules had pinkish internal colour but the leave colour of the seedlings were yellowish for all, except for the positive controls. Similarly, the shoot dry weight of the negative controls and the inoculated ones were the same. There was a significant difference between the shoot biomass of the inoculated seedlings and the positive controls. Thus, this was forced us to decide that the strains was not effective and might be affected by the high lath-house temperature for which we designed the second experiment to carry during a cool season.

The result of the second experiment was better than the first one. Out of the 65 strains, CIAT 899, CH24-10 (Mexican), HB429 (local), NAK 91, NAK 103 and NAk 104 (Kenyan



strains) produced green leave colour of common bean while the others produced yellowish leave colour (Figure 1). The shoot dry weight of those seedlings were higher than the rest, except for positive controls. The highest shoot dry weight was recorded for the positive controls (Figure 2). Nodule number (removed from graph, Figure 2) could not be an indicator for effectiveness of the strains (Figure 3). Because, they did not correlated with the shoot weight measured.



Figure 1: Response of Common bean variety to rhizobial inoculation



Figure 2: Shoot, root and nodule dry weight (on y-axis) of Nasir common bean variety inoculated with rhizobium strains (on x-axis). N- is negative control, which was not inoculated and supplemented with 0.5% KNO₃ while N+ is positive control that was not inoculated but supplemented with 0.5% KNO₃. SDW = shoot dry weight; RDW = root dry weight and NDW = nodule dry weight. Effectiveness of the strains is determined based on the following criterion:

Table 1: Criteria for describing effectiveness

Abbr.	In words	Criterion
1	Ineffective	SDWs <sdwnc< td=""></sdwnc<>
е	effective	SDWs >SDWnc < 50%SDWpc
m	moderately effective	SDWs>=50%SDWpc<=75%SDWpc
E	highly effective	SDWs>75%SDWpc

SDWs = shoot dry weight of the inoculated seedlings; SDW of negative controls; SDWpc = SDW of positive controls



Figure 3: Common bean nodulation assessment

The chickpea seedlings did not show difference in leave colour (Figure 4) but their shoot height and vigour were very variable. M7, M4b, M20a, M20, CP130, CP67 and CP65 strains produced better shoot dry weight and plant vigour than others (Figure 5). The commercial strain CP113 was not able to form nodule and the original strain might be dead or it might be mutated over time and become unable

to nodulate. Examples of nodules for effective strains are indicated in figure 6.

CIAT 899, CH24-10, HB429, NAK 91, NAK 103 and NAk 104 common bean strains and M7, M4b, M20a, M20, CP130, CP67 and CP65 chickpea strains were relatively more effective than others and selected for GL x GR interaction study.



Figure 4: Response of Chickpea variety to rhizobial inoculations



Figure 5: Chickpea strain performance on Natoli variety. ASDW = Average shoot dry weight; ARDW = average root dry weight and ANDW = Average nodule dry weight



Figure 6: Chickpea nodulation assessment

Ashenafi Hailu Gunnabo, Ethiopia, Wageningen University, The Netherlands

Nigerian PhD Research update

I am Ojo Comfort Tinuade, one of the N2Africa PhD rstudents with Wageningen University and a Research Fellow with International Institute of Tropical Agriculture, Ibadan. My research interest is to explore the potential benefits of rhizobia inoculation with cowpea. My research activities includes estimating the population density of indigenous rhizobia in soil samples collected from Northern Nigeria, isolation and identification of highly competitive and efficient rhizobia strains from Nigerian soils. For the first six months at the start of the PhD programme, I developed my research proposal with supervisors in Wageningen University, took some courses and a qualifying exams. I am presently in my home country for the research activities in International Institute of Tropical Agriculture, Ibadan. The list of activities I am working on presently include; soil sample collection in Kano, Kaduna and Niger states. These areas were chosen because they are predominantly cowpea cultivated region in Nigeria, 54 soil samples were collected), physicochemical analysis, estimating the population of the indigenous rhizobia in soils using the most probable number techniques (ten-fold dilution series in six consecutive steps). Figure 1 shows growth of cowpea after



Figure 1: Growth of cowpea one week after inoculation with dilutions of the soil samples

one week of inoculation with dilutions of the soil samples. The next activity will be a glasshouse experiment that will involve a competition experiment between known elite rhizobia strain and indigenous rhizobia strains with cowpea.



Figure 2: One week after inoculation with soil

Comfort Ojo, Wageningen University, The Netherlands

Exploring options to enhance biological nitrogen fixation and yield of soyabean and common bean in smallholder farming systems of Rwanda

In Rwanda like in many sub-Saharan Africa countries, increasing population hence reduction of farm size, declining soil fertility and high fertilizer cost have been reported as major production constraints for smallholder farmers. The use of organic inputs and biological nitrogen fixation are alternative to cut down fertilizer costs. Rwandan government has recently set a policy of one cow per poor family in one hand to deal with soil fertility issues. In this study we explored the benefits of using manure and inoculation for increased nitrogen fixation and yields of bean and soyabean, and manure showed the ability to sustain productivity without repeated inoculation.

Among the activities done under the present PhD research, here we present some results on the last field experiment on the need- to- reinoculate in previously inoculated fields showing how grain and dry matter of bush bean responded to manure and inoculation at two different sites in Rwanda. The variety of bush bean used responded to inoculation when manure was added. Re-inoculation effect was more significant in Kamonyi site than Bugesera site. Low grain and dry matter yields were observed in uninoculated plots with no manure application at each site and season but consistently higher in Kamonyi than Bugesera. However, an increase in grain yield in uninoculated plots was observed



Figures (a, b, c & d). Grain (a & b) and dry matter (c & d) yield (t/ha) response P+lno in s1 of bush bean variety RWR 2245 to three manure doses in Bugesera (a & c) and Kamonyi (b & d) sites. Where: P+ino in S1: with P fertilizer addition and inoculation in season1; P+reino in S3: Plots received P in s1 with re-inoculation -P-ino in season 3; P-ino: With P addition but without inoculation.

with manure addition. Higher grain and dry matter yields were observed in inoculated and manured plots. The Bugesera site suggest the use of more manure rates compared to Kamonyi site which showed a peak at 5t/ha for both inoculated and uninoculated plots. There was no significant differences between re- inoculated plots and inoculated plots in the first season receiving 5t and 10t/ha for both grain and dry matter yields in Bugesera site (figures a & c). The high yields observed in manured treatments without inoculation shows the benefits of manure to plant growth in the case of lack of inoculants to farmers.

Currently, I am back in Wageningen for laboratory work on one component of my research on rhizobia identification and thesis write up.

Edouard Rurangwa, Wageningen University, The Netherlands

Putting nitrogen fixation to work for smallholder farmers in Africa

P+reino in S3

Effect of intercropping patterns on crop productivity across fields of different soil fertility in northern Guinea and Sudan savannah agroecological zones of Ghana

Two on-farm trials were conducted in the 2013 and 2014 cropping seasons in the northern Guinea savannah agroecological zone (AEZ 1) and the Sudan savannah agroecological zone (AEZ 2) of Ghana to evaluate the effect of intercropping patterns and the effect of cowpea-maize relay intercropping on crop productivity. The intercropping patters were within-row intercropping and 1:1 and 2:2 distinct rows of maize- legume (Figure 1a, b). The legumes tested included cowpea, soyabean and groundnut. The trials were conducted across fields of different soil fertility levels. In this article, radiation interception and land equivalent ratios of the different intercropping systems are discussed.

In the 2014 intercropping trials, radiation interception (Photosynthetically Active Radiation, PAR) of the different intercropping systems tested was measured with the Table 1 Land equivalent ratios (LER) for cowpea, soyabean and groundnut intercropped with maize in different planting patterns, at different fertility levels and agroecological zones

	AEZ 1			AEZ 2					
	HF	MF	LF	Mean	HF	MF	LF	Mean	Total AEZs
MZ-CP within row	1.3	1.6	1.4	1.4	1.4	1.4	1.8	1.5	1.5
MZ-CP 1:1 rows	1.2	1.4	1.3	1.3	1.1	1.4	1.8	1.4	1.4
MZ-CP 2:2 rows	1.2	1.3	1.2	1.2	1.2	1.3	1.4	1.3	1.3
MZ-SB within row	1.3	1.6	1.3	1.4	1.5	1.5	1.7	1.6	1.5
MZ-SB 1:1 rows	1.1	1.3	1.2	1.2	1.2	1.4	1.7	1.4	1.3
MZ-SB 2:2 rows	1.1	1.2	1.2	1.2	1.2	1.4	1.6	1.4	1.3
MZ-GN within row	1.5	1.4	1.5	1.5	1.4	1.5	1.7	1.5	1.5
MZ-GN 1:1 rows	1.4	1.2	1.4	1.3	1.3	1.3	1.5	1.3	1.3
MZ-GN 2:2 rows	1.3	1.3	1.3	1.3	1.2	1.3	1.6	1.4	1.3



Figure 1. Visual appraisal of **a**) within-row maize-legume and **b**) distinct 1:1 planting rows of maize-legume intercropping systems as well as **c**) and **d**), how radiation interception was measured. In the picture Michael Kermah is taking some radiation measurements.



medium and low fertility field types are represented by the letters a, b and c respectively. MZ-CP within-row means maize-cowpea intercropped within the same row, MZ-CP 1:1 and MZ-CP 2:2 rows indicates maize-cowpea intercropped in distinct 1:1 and 2:2 planting rows respectively. AccuPAR LP-80 ceptometer¹ (Figure 1c,d). In most cases, the IPAR was higher for the withinrow maize-cowpea system than the other two intercrop systems, particularly at 55 to 67 DAS which corresponds to early to mid-pod filling stage of the cowpea (Figure 2). The differences were significant with P<0.001 for 28 and 55 DAS (days after sowing) and P=0.009 and 0.010 respectively for 40 and 67 DAS. Only at 77 DAS the differences in intercepted PAR were not significant.

The land equivalent ratios (LER) results indicate that all the intercropping systems tested were efficient in using the available environmental resources and more productive than the sole crops of legumes and maize. These were observed in all three soil fertility levels and in both AEZs. However, in most cases, the within-row intercropping system gave higher LER and intercrop productivity. The 1:1 maize-legume distinct planting rows also seems to give better LER and productivity than the 2:2 system. The LERs were higher in the low than the high fertility fields in both AEZs.

Michael Kermah, Wageningen University

¹ Decagon Devices Inc. Pullman, Washington



Common bean and response to rhizobial strains on different soil types in Uganda

Common bean (Phaseolus vulgaris L.) is one of the most important grain legumes in Uganda. It forms an important part of the traditional diet and provides a good source of income for the rural communities. Experiments with rhizobial inoculation on beans in East Africa have met with varying success. One possible explanation is that common bean can establish effective symbiosis with broad range of native rhizobia. The role of this so-called promiscuity in the irregular response to inoculation remains to be established. It is currently not known how the composition of the rhizobial population in soil affects nitrogen fixation in common bean.

In my PhD project, I will explore the abundance and diversity of rhizobia and determine inoculation response of common bean in different soils and the effects of environmental and cropping practices. From the study, effective and competitive indigenous rhizobium strains will be identified and will be assessed for their potential as commercial inoculants with the aim of increased and sustainable bean production within the region. At the start, rhizobia will be isolated from 80 soil samples within the common bean (climbing bean variety) growing areas in the South-Western and Eastern highlands in Uganda where N2Africa is actively working with smallholder farmers. My current focus is using ISRIC maps to characterize the different soil types in the bean growing areas and to identify the land use practices in rela-



Figure 1: ISRIC soil maps for Kapchorwa (top), Kanungu, Kabale and Kisoro (bottom) at a scale of 1:10km

tion to the cropping history. This data will be used to select sample soils for physico-chemical analysis, most-probable numbers (MPNs) and rhizobial trapping.

Allan Ochieng, Wageningen University, The Netherlands

Grain legume residues as a livestock feed resource for smallholders in northern Ghana

Agriculture in Ghana is dominated by small-scale crop-livestock farming based on cereal and legume production and livestock rearing. Livestock rearing is an important component of crop-livestock farming system because it serves as an insurance against crop failure, a source of manure, a source of cash and makes good use of crop residues. The main objective of this study is to understand the roles and functions of grain legume residues in livestock farming systems and identify options to improve utilisation by smallholders in northern Ghana.

A rapid diagnostic survey has been conducted in eight communities to assess the existing and potential feed

resources, their uses and seasonal gaps with respect to ruminant production in northern Ghana. The survey was carried out in May 2015, using the Feed Assessment Tool (FEAST) developed by International Livestock Research Institute (ILRI). FEAST is an analytical tool for identification of site-specific livestock feed resources and other related production constraints. This tool comprises Participatory Rural Appraisal (PRA) and structured questionnaire for



Figure 1: Focus group discussion with farmers at Figure 2: Individual interview with a female Tusani community, Northern region

individual interviews. The survey indicated that the major problems faced by livestock keepers in northern Ghana are poor housing, disease incidence with high mortality rates, inadequate feed in the dry season, theft and high cost of veterinary drugs.

Preliminary findings on the use of grain legume residues indicated that farmers in Upper East region store about 70 -80% of their grain legume residues to feed livestock later in the year. This is followed by farmers in Northern region who store about 10 – 30% of grain legume residues for animal feed while farmers in Upper West region store none of their crop residues but allow animals to graze them on the field.



farmer at Sandu Community, Northern region



Other feed resources used in the area include agro-industrial by-products like corn mill waste flour, brewers' spent grain, maize bran, and rice bran, cassava and yam peels.

On-farm trials are currently being conducted to determine the influence of grain legume genotype, environment and management practices on fodder yield and quality of soyabean, groundnut and cowpea. Future activities under the project will include: i) Characterization of diversity of crop-

Crowdsourcing for agricultural data collection

Data availability often limits detailed assessment and explanation of yield gaps at farm level. Bottom-up data collection approaches (e.g. crowdsourcing) can be used to overcome lack of data and improve yield gap analysis. A case study with 50 farmers selected from 5 districts was carried out in Ethiopia. The main objective of the case study was to explore the opportunities and applicability of innovative bottom-up data collection approaches like crowdsourcing (using SMS) and digital devices (tablets) to collect factors that could explain yield gap. Detailed socio-economic and agronomic data were collected using tablets. Besides, short message system (SMS) using farmers owned basic phones was tested to assess its applicability to collect cropping and event calendar information (e.g. sowing date, weeding date) and incidence of pest and diseases directly from farmers.

Description of the technologies tested

Open Data Kit (ODK): Open data kit is a free and opensource set of tools which was designed to help organisations build information services. ODK provides solution to build a data collection form, to collect data on a tablet device and send it to a central server and aggregate the collected data on a server and retrieve it in a useful format. The ODK collect app was used to run the digital version of the complete field book on Nexus 7 tablets and collect both socio-economic and agronomic data and send to an ODK aggregate server (Fig. 1)

FrontlineSMS and Ushahidi: connected together

FrontlineSMS and Ushahidi are both free and open source software tools, commonly used by social change projects. FrontlineSMS enable users to send, receive and manage large numbers of incoming and outgoing SMS (http://www.frontlinesms.com/). FrontlineSMS does not require the internet to work, but does need to be connected to a mobile network. Ushahidi is a platform for collecting, visual-

livestock farming systems and the roles and functions of grain legume residues in livestock feeding; ii) Identification best storage methods of grain legume residues; iii) Assessment of farmers' perceptions on the use of grain legume residues as livestock feed. This will lead onto farm and farming system level analyses to explore options for intensification of livestock production.

Daniel Brain Akakpo, Wageningen University, The Netherlands

ising and mapping information (http://www.ushahidi.com/). Using these tools together can produce good results, with FrontlineSMS being used as a tool which can manage incoming SMS data which can then be visually represented using Ushahidi. The cloud-based version of Ushahidi (Crowdmap) was used in this pilot study to receive an automatically forwarded SMS message from the FrontlineSMS application. FrontlineSMS application uses a local SIM card; data sent to the application can only be accessed by people who have access to the local computer where the FrontlineSMS application is installed. To overcome this limitation, we linked the FrontlineSMS application with the Crowdmap platform so that SMS data received by FrontlineSMS is automatically forwarded to the Crowdmap platform and project partners from the implementation area and have access to internet can also access the SMS data sent.

Over the growing season, farmers in the five districts send around 165 SMS messages, 35 farmers were active and provided the information about the activities they performed in the trial plots via SMS. At the end of the growing season, farmers were interviewed about the SMS data collection and their motivation to continue participation. Famers were asked to rank on a Likert scale (1-5, where 1 is strongly disagree and 5 is strongly agree) statements that reflected their motivation regarding continue participation in SMS data collection. Open questions were also included in the interview to give farmers more option to explain their motivation for participation. The findings indicated that farmers are motivated both intrinsically (e.g. found it interesting) and extrinsically (e.g. expect something in return from the expert) to participate in SMS data collection. In addition, farmers also have an altruistic motivation (doing something for the benefit of others e.g. to help the researcher get his/her work done). Understanding the motivation of the participants to participate and providing the right incentive

> in crowdsourcing initiatives can help to improve both the quantity and quality of data. Technical challenges (not capable of operating the phone), unavailability of network coverage in a specific date and lack of support from the local implementing partners (extension workers) were mentioned as the major challenges



Figure 1: Overview of the data collection flow using tablets running the ODK collect app





Figure 2: Overview of the information flow between the farmer and N2Africa project

farmers faced during the SMS data collection. Loss of contact with the extension workers/local implementing partners made some farmers to withdraw from the pilot study. Lack of continuous support from local implementing partners was also one of the main challenge mentioned by extension workers who used the tablets for detailed socio-economic and agronomic information. Acknowledging the farmers with an automatic reply after receiving SMS message would have been an alternative option to keep the farmers engaged and motivated in the data provision process. The lessons learnt from this pilot are used in the implementation of other digital data collection pilot studies.

Eskender Beza*, Wageningen University, The Netherlands

* Eskender is co-financed by N2Africa

What about N2Africa and the diet of this 2 year old Ghanaian girl Talata*? An update of my PhD research

How are N2Africa activities linked with the dietary intake of infants and young children like this Ghanaian girl Talata? And how do the activities trickle down to the nutritional status of these infants and children and therefore their productivity later in life? The majority of the undernourished population in the world live in rural households in developing countries. Farming is their



mainstay and often the only means to access of food and income. Evidence for theoretically assumed linkages between agriculture and nutrition is, however, weak. In the context of the N2Africa project, we will research the linkages between agricuture and nutrition and their drivers in rural households.

My main objectives and their progress:

1. To assess the role of legumes in the sub-Saharan African diet: their nutritional values, and their current and potential role in the diet of young children

My literature research shows that grain legumes are better sources of protein and contain a larger variety and concentrations of micronutrients compared with sub-Saharan African staples like maize (in Podcaster 15). Dietary intake data is collected in Northern Ghana and in Western Kenya which I will use to analyze the current and potential role in the diet of young children with a linear modelling programme. 2. To assess the association between legume production and legume consumption, dietary diversity and nutritional status of children and its drivers

I conducted two case studies, one in Ghana and one in Kenya. In Ghana, a correlation was found between N2Africa and a higher nutrient adequacy of the diet of young children. Focus group discussions show female N2Africa farmers contribute directly to increase in food availability at home while male farmers sell most of their produce. It is unclear how and if improved sales may trickle down to improved diets (links to Podcaster 22, 29 and N2Africa TV)

3. To assess the association between crop diversity with dietary diversity and nutrient gap

The research is a preliminary phase. Analyses will be done both at detailed level in Northern Ghana, and at broader scale with existing data sets.

4. To re-design and test farm systems based on nutrient gap analysis at food level Detailed nutrition data is collected in Northern Ghana

and analyses are in progress, for preliminary results see previous podcaster: (Podcaster 31).

More in depth insight in the linkages between agriculture and nutrition will offer guidance to shape N2Africa activities or activities of other agriculture projects in such a way that increases in yield or increases in diversity of foods produce trickle down to improved dietary intake of Talata.

Ilse de Jager, Wageningen University, The Netherlands

* Talata is a real person but a fictitious name



Introducing Mr Shakiru Quadri, Youth Agri-preneur Facilitator for N2Africa-Borno State

We have the pleasure of introducing Mr Shakiru Quadri, the Youth Agri-preneur Facilitator for N2Africa-Borno State, working under the supervision of Dr. Nkeki Kamai. Mr. Quadri is a graduate of General Agriculture of the Institute of Agricultural Research and Training (IAR&T), University of Ife (1982) with a first research work on "the effect of four rhizobium strains



[local and foreign] on nodulation, N-fixation, and yield of cowpea [Ife brown]". He obtained a Master's Degree in Business Administration (MBA) from the University of Ilorin (2005), and a Master of Science (MSc) Food Security and Development of the University of Reading, United Kingdom (2014).

Mr. Quadri's job experience over the years has revolved around Farm management/Food production, Participatory Practices for Sustainable Development (PPSD), Youth mobilization/Empowerment, Leadership and Enterprise Development. He joined the N2Africa Borno team shortly after returning from the Graduate Institute of International Development and Applied Economics (GIIDAE), University of Reading; having previously worked for the Development and Leadership Institute (DLI) as Executive Director (2009 – 2013), Youth Empowerment Scheme (Project YES) as Coordinating Director (2006 – 2009), and the National Youth Service Corps (NYSC) as Farm manager (1983 – 2003). He worked on a personal initiative Global Youth and Community Development Network (GYCDN) (2003 – 2005) after undergoing training in PPSD at the International Institute for Sustainable Development (IISD), Colorado State University, Fort Collins, USA.

His latest research (2014) is on "the trend of food security parameters in Nigeria: implications for household vulnerability"; and a (2002) publication on "Community Development in Nigeria: a case study of the NYSC IRD programme in Kwara State". Journal of Nigerian Affairs vol. 10 (1), 30-35.

Fred Kanampiu

N2Africa features as case study within the PROIntensAfrica project

N2Africa was recently selected as case study within a project called PROIntensAfrica. PROIntensAfrica aims to build a long-term research and innovation partnership between Africa and the European Union, focusing on the improvement of food and nutrition security. The project has a large number of partners in Europe (including Wageningen University and CIRAD), and in Africa (including FARA, CORAF, CCARDESA, ASARECA).

As part of this project, different approaches for sustainable intensification will be studied through a literature review and a number of case studies. With N2Africa's selection as case study in this project we have the opportunity to take a closer look at N2Africa's impact on different sustainability indicators (environmental, social and economic), and on a range of different stakeholders (farmers, research, NGO partners, government, etc.)

The case study will be coordinated from Wageningen and will run in the period between October 2015 and March

2016. Next to some desk research on potential indicators for sustainable intensification and a review of existing project documents on how N2Africa performs on such indicators, there will be room for additional data collection on indicators that have not been measured yet. The case study will conclude with a workshop where a wide selection of stakeholders will be invited to reflect on the outcomes of the study and the perceived impact of the project. A selection of countries and/or specific topics for the case study will still have to be made. There are interesting examples from all corners of the project, but at least the case study should show a minimum of 2-3 years 'on the ground effects'.

We are sure that this will be a great opportunity to profile N2Africa among a wider group of stakeholders, and to dive deeper into the impact created by the project. We will keep you updated!

Esther Ronner, Ken Giller

Other newsletters

We received the Soybean Innovation Lab (SIL) July Newsletter

The Podcaster is published six to eight times per year – we look forward to receiving news and contributions – particularly from partners. Please send in contributions well in time. Contact address for this newsletter is: N2Africa.office@wur.nl

Please feel free to forward this email to colleagues you believe would be interested. This email has been sent to you by N2Africa, funded by The Bill & Melinda Gates Foundation.

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