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Australian technology reaches the field: supporting and monitoring the release of Pod-Borer Resistant Cowpea

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prepared by Jose Barrero Sanchez and TJ Higgins

*co-authors/
contributors/
collaborators* Dr. Iliyasu Utono, Dr. Aliyu Anchau, Mrs Rebecca Mewase, Prof. Hajara Shuaibu, Prof. E. Ikani, Dr. Bernard Ehirim and Dr. Francis Onyekachi

approved by Eric Huttner, Research Program Manager CROPS

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1 Acknowledgments

The Commonwealth Scientific and Industrial Research Organisation (CSIRO) wish to acknowledge the Australian Department of Foreign Affairs and Trade for funding through the Australian Centre for International Agricultural Research (ACIAR) this Small Research Activity (SRA) to support and monitor the release of the Pod-Borer Resistant (PBR) cowpea in West Africa. We also acknowledge the African Agricultural Technology Foundation (AATF) for accepting the challenge of leading this project on the field. Their contribution in preparing the experimental protocols and in conducting and coordinating the research has been outstanding.

We recognise the contributions of all the project partners who played specific roles during the duration of this project, particularly the Institute of Agricultural Research (IAR)/Ahmadu Bello University (ABU), National Agricultural Extension Research and Liaison Services (NAERLS), National Agricultural Seed Council (NASC), the Council for Scientific and Industrial Research -Savanna Agricultural Research Institute (CSIR-SARI) Ghana and Institute of Environment and Agricultural Research (INERA) Burkina Faso

We are very grateful to all the respondents and Agricultural Development Program (ADP) Staff who helped us identify and sample all the farmers.

2 Executive summary

The production of cowpea in West Africa is hampered by many factors, including insect pests that often destroy up to 80% of the crop. One of the most devastating insects is the legume pod borer, *Maruca vitrata*, a lepidopteran whose caterpillar burrows into the cowpea pods and eats the grains. To solve this problem, a genetically modified (GM) Pod Borer Resistant (PBR) cowpea variety was developed by an international team and released in Nigeria in 2019. This PBR cowpea (commercialised with the name Sampea 20-T) carries the gene encoding the insecticidal protein Cry1Ab (from *Bacillus thuringiensis* or Bt) and provides total protection against *Maruca*.

This Small Research Activity (SRA) was aimed at assisting the dissemination and monitoring the effects of the released PBR cowpea in Nigeria. Being the first introduction of a GM food crop in West Africa, it was thought necessary to oversee the post-release stewardship of the product and to monitor its effects at the farmers' level. The goal was to produce an early assessment of the effectiveness of the PBR cowpea and to evaluate its impact on the land and farmers growing this new crop. This SRA performed several studies in the 2022/2023 season of a sentinel sample of 80 farmer adopters of the PBR cowpea and compared them to 80 farmers growing conventional cowpea varieties. The farmers selected covered the 4 agroecological zones of Nigeria (Sahel, Sudan, Northern Guinea, and Southern Guinea savannah zones) where cowpea is traditionally grown and where the PBR cowpea is being commercialized.

The results from this project show that there was a clear comparative advantage of PBR-cowpea over non-PBR cowpea not only in terms of yield, but also on factors related to environment and human health. This study has also found strong support for the product and good adoption of the recommended stewardship practices. In short, the results for the performance of the PBR cowpea in farmers' fields on its second year of cultivation are very encouraging, supporting a positive impact of the PBR cowpea in many areas and providing a strong argument for the full-scale deployment of this biotechnology product. More broadly the results presented here demonstrate how biotechnology, carefully targeted to address

farmer's needs, can assist in solving some of the big agricultural problems in Africa to improve Food Security for its people.

3 Background

The African continent is still facing serious problems of food security, due to a number of complex variables including technological deficiencies, political instability or climatic challenges (Ayinde et al., 2020). In many parts of Africa farmers are only able to harvest a small fraction of the potential yields that their crops could offer. The improvement of agriculture to close those yield gaps is an important key to solving the current hunger and poverty challenges. In West Africa, one important crop is cowpea, also known as black-eye peas in other parts of the world, which is eaten daily by over 200 million people and provides a main source of protein for people who cannot afford meat. The production of cowpea in West Africa is hampered by many factors, including insect pests that often destroy up to 80% of the crop. One of the most devastating insects is the legume pod borer, *Maruca vitrata*, a lepidopteran pest whose caterpillar burrows into the cowpea pods and eats the grains. After decades of intensive research, scientists could not identify any source of natural resistance against this pest. So, to control the pest, farmers have relied on the use of chemical insecticides, which are expensive and are also impacting the environment and people's health.

To solve this problem, about 20 years ago, a public private partnership led by the African Agricultural Technology Foundation (AATF) and the CSIRO was formed to develop a biotechnological solution against Maruca: The Pod-Borer Resistant (PBR) cowpea. The project, with the support of both the Rockefeller Foundation and USAID, generated the world's first genetically modified cowpea carrying the insecticidal Bt gene Cry1Ab, which confers in-built resistance to the pest (Addae *et al.*, 2020). After years of demonstration trials, the PBR cowpea was granted environmental release approval by the National Biosafety Management Agency of Nigeria in January 2019. By December the same year, the first PBR cowpea variety was registered under the name Sampea 20-T and released for cultivation following successful varietal certification trials across the country. The PBR cowpea is the first genetically modified food crop bred and released in Nigeria. Initially three Nigerian seed companies were licensed to produce and distribute this new variety. The demand has been so great that currently ten other seed companies have been licensed to produce certified seed at scale. Also, to guarantee the best outcome of the new PBR cowpea, AATF in collaboration with the Institute of Agricultural Research (IAR) and the National Agricultural Extension Research and Liaison Services (NAERLS) developed a full stewardship protocol with recommended agronomic practices to use in combination with the PBR cowpea variety. The success in Nigeria has accelerated the acceptance of the PBR cowpea in Ghana, where it was approved for environmental release in June 2022. In Burkina Faso, approval has been given for controlled release which allowed to conduct variety certification trials in 2023. It is expected that the PBR cowpea will be approved for commercial release in Burkina Faso in 2024.

This Small Research Activity (SRA) was conceived to provide an early evaluation of the performance in farmers' fields of the PBR cowpea in several representative agro-ecological zones. The environmental and health impacts of this cowpea variety were to be investigated together with an evaluation of the genetic purity of the seeds provided to, and produced by, farmers as well as the acceptance of the PBR variety and the adoption of the recommended cultivation practices. CSIRO and AATF have successfully collaborated over the last 2 decades to develop the PBR cowpea product, so AATF was seen as the perfect partner to lead the research activities in the field and to deliver the outcomes needed. AATF, through

its office in Abuja Nigeria, and headquarters in Nairobi Kenya, coordinated the work closely with partners in Nigeria: the Institute of Agricultural Research (IAR)/Ahmadu Bello University (ABU), the National Agricultural Extension Research and Liaison Services (NAERLS) and the National Agricultural Seed Council (NASC) to execute various components of the project. CSIRO worked closely with AATF to design the project and to provide technical advice and recommendations.

4 Objectives

The Objectives were to:

1. Determine the field-based biological consequences (entomological effects and soil effects) of PBR cowpea compared to conventional cowpea.
2. Assess the genetic purity of the PBR cowpea seed in adopter farmers' fields.
3. Assess practice change, in particular compliance to stewardship program (yield and residues of chemical insecticides).
4. Conduct an overall comparison of adopters of PBR cowpea versus non-adopters.
5. Assess the interest for PBR cowpea in Ghana and Burkina Faso.

These lead researchers on the various components were coordinated by Dr. Francis Onyekachi and Dr. Bernard Ehirim from AATF:

Research question	Person responsible	Institution
What are the field-based biological consequences of PBR cowpea compared to conventional cowpeas in Nigeria?	Dr. Iliyasu Utono (non-target organisms) and Dr. Aliyu Anchau (Soil Microbial Activities).	Institute for Agricultural Research, Ahmadu Bello University, Samaru, Zaria.
Is adequate genetically pure PBR cowpea seed being used by farmers in Nigeria?	Mrs Rebecca Mewase.	National Agricultural Seed Council.
Are farmers adhering to the recommended field practices when introducing PBR cowpea?	Dr. Iliyasu Utono.	Institute for Agricultural Research, Ahmadu Bello University, Samaru, Zaria.
How do adopters compare to non-adopters after introduction, with respect to practices and results?	Prof. Hajara Shuaibu.	Institute for Agricultural Research, Ahmadu Bello University, Samaru, Zaria.
What are the expectations from farmers in Ghana and Burkina Faso about PBR cowpea future deployment?	Prof. E. Ikani.	National Agricultural Extension Research and Liaison services (NAERLS).

5 Methodology

A list of PBR cowpea adopter farmers (seed buyers) was obtained from the licensed seed companies, and from those a random sample of 80 farmers was selected for this study (Appendix 1), comprising 20 farmers from each of the 4 agro-ecologies in Nigeria (Figure 1): Sahel savannah zone (Katsina state), Sudan savannah zone (Kano state), Northern Guinea savannah zone (Kano and Kaduna states) and the Southern Guinea savannah zone (Kuje, Abuja). A total of 80 PBR cowpea adopter farmers and 80 non-adopters were used in this project. For each adopter farmer, matching non-adopter farmers who planted conventional cowpeas were identified in the neighbourhood (within approx. 20 meters) of the PBR cowpea farmers and used as controls.

For the objectives 1, 2 and 3, which required a very precise and laborious experimental approach, a smaller subset of farmers was randomly chosen in each agroecological zone from the initial pool of 20. For objectives 1 and 3, a subset of 5 adopter farmers and 5 non-adopter controls were used. For objective 2, a different subset of 5 farmers were selected per agroecological zone. For objective 4, the whole set of 80 adopters plus 80 non-adopters were surveyed.

For objective 5, 180 farmer interviews were conducted in Ghana, comprising individuals representing different regions and districts. A similar approach was used in Burkina Faso.

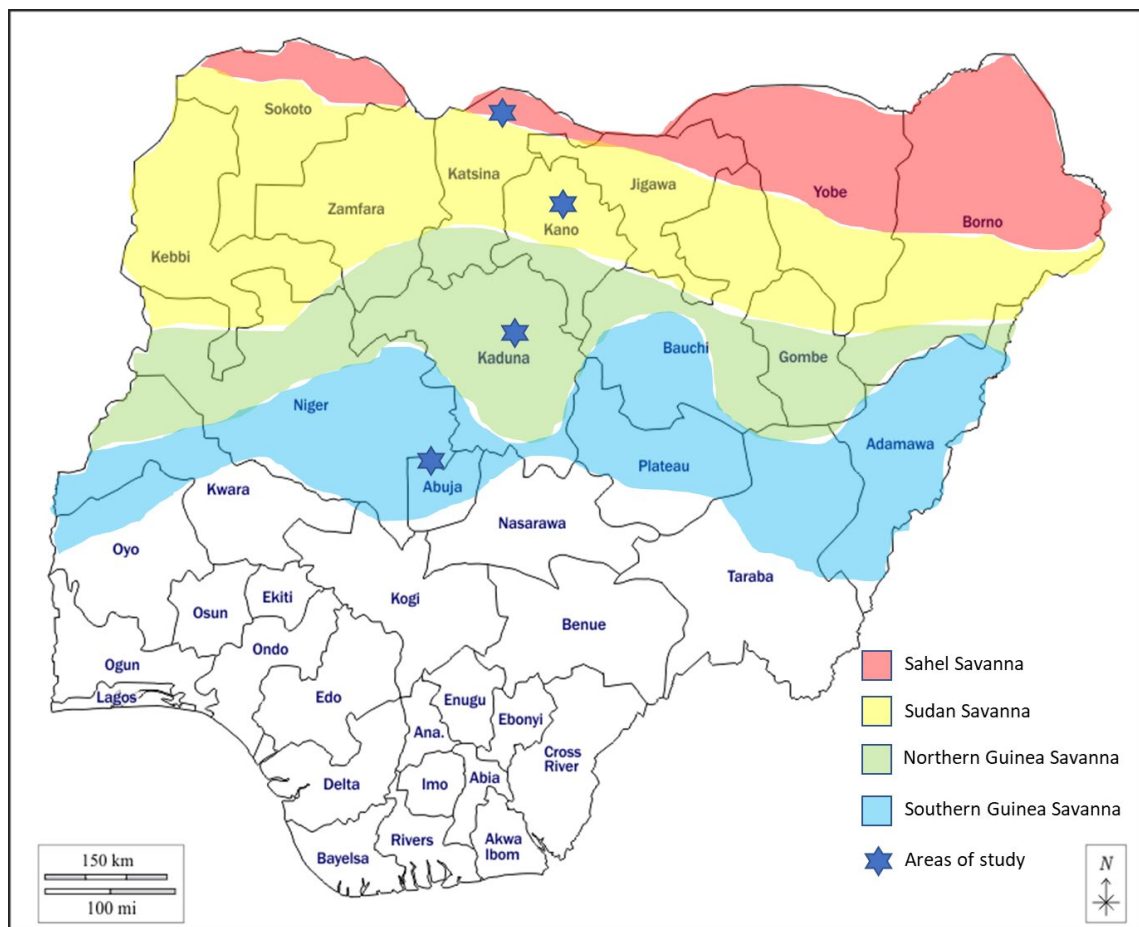


Figure 1. Map of Nigeria indicating the four agroecological zones where cowpeas are mostly grown as well as the areas of study.

Objective 1: Determine the field-based biological consequences (entomological and soil effects) of PBR cowpea compared to conventional cowpea.

This objective comprises two components relating to entomological consequences or effects (Objective 1a) and soil biological effects (Objective 1b).

Objective 1a: Determine the field-based biological consequences (entomological effects) of PBR cowpea compared to conventional cowpea.

In each of the PBR cowpea and non-PBR cowpea farmers' fields, a 5m x 5m area was demarcated at the centre of each farm. Each field was sampled for target (Maruca) and non-target organisms (NTO) such as thrips, pod sucking (PSB), aphids, bugs, ladybird beetles, ants, syrphids, spiders and other insects present during the survey. Two methods of insect sampling were used: visual observation and sticky traps.

- **Visual observation:** Forty (40) flowers were randomly sampled. Each flower was opened and the presence of thrips and Maruca recorded. Pod sucking bugs and aphids occurring in each field were visually counted and recorded. Other NTO and other insects were visually counted and recorded.

- **Sampling with sticky traps:** At the time of visual observation, 2 sticky traps were placed for two weeks at the centre of each field, 3m apart. Then each trap was retrieved for identification and recording of the insects caught. The insect samples were placed in bottles containing 75% ethanol solution and taken to the Department of Crop Protection, Institute for Agricultural Research, Ahmadu Bello University Zaria insect museum, where they were identified and counted.

Objective 1b: Determine the field-based biological consequences (use of pesticides on soil biological properties and soil fertility) of PBR cowpea compared to conventional cowpea.

Of the 5 adopters and 5 non-adopters used for the insect studies, 3 of each were sampled for the soil study. The adopters were those who adopted the PBR cowpea and its recommended agronomic practices, including only a two-pesticide spray regime. The non-adopter farmers were those who grew other cowpea varieties using their traditional and conventional practices, including four or more pesticide applications. The treatments were replicated three times in each of the agroecological zones in a randomized complete block design. A total of 24 samples were taken from the rhizosphere soils of the plants from the four agroecological zones after all the pesticide sprays were applied. These samples were used for the determination of bacterial and fungal populations as well as microbial biomass carbon, nitrogen, and phosphorus. Another 24 soil samples were taken at 0-20 cm after the harvest of the crops, targeted at determination of the influence of the treatments on the soil chemical and physical properties, as related to soil fertility.

The first set of soil samples were collected from the rhizosphere of the plants, soils attached to the roots of the plants after the application of all the pesticide sprays have been applied by the farmers. Five randomly selected plants were marked in each plot, uprooted using a hand trowel and the soils attached to the root deposited in a clean bucket then a subsample was packaged in a polyethene plastic bag and labelled. The samples were then stored at 4°C to preserve live microbes present and microbial biomass in the samples, prior to laboratory analysis for microbial populations and microbial biomass carbon (C), nitrogen (N) and phosphorus (P). The second set of soil samples were collected after harvest of the plants at 0-20 cm depth from each of the plots using an auger. Five randomly selected soil samples

were collected from each plot and deposited in a bucket, then a sub-sample was taken and labelled as above. These samples were air dried, crushed using stainless steel pestle and mortar and sieved through a 2mm sieve mesh. They were then stored safely at room temperature prior to analyses for routine soil physical and chemical properties.

The soil samples collected from the rhizosphere and stored at 4°C were subjected to viable count for enumeration of pure cultures of bacteria and fungi. The activity was conducted using Miles and Misra drop method (Surface Viable count) (Miles and Misra, 1938), as described by Anasuya *et al* (2016). The microbial Biomass C, N and P was calculated as described in Okalebo *et al.*, 2002 and Anderson and Ingram (1993). To determine the soil physical and chemical properties related to soil fertility, the samples collected after harvest were subjected to determination of routine soil properties: particle size distribution, pH, electrical conductivity, organic carbon, total nitrogen, available phosphorus, exchangeable Ca, Mg, K and Na, exchangeable acidity, cation exchange capacity and selected micronutrients (Zn, Mn, Cu and Fe) (Agbenin, 1995). All relevant data collected were subjected to statistical analyses.

Objective 2: Assess the genetic purity of the supplied PBR cowpea seed from licensed sources, as compared to sourced PBR cowpea from seed savings and conventional cowpea seed.

For the seed purity analysis, in addition to the adopters (farmers who bought the PBR cowpea seed from the seed companies) and non-adopters, a third category was included, namely, seed-savers (farmers who saved PBR cowpea seeds from the previous year or who were gifted seeds). For each one of these 3 categories, 5 farmers were selected per agro-ecological zone. Ten representative samples (pools of 20 seeds) were collected from each farmer's field for analysis. DNA was extracted from each pool of seeds and used for PCR analysis using a zygosity test developed for the PBR cowpea transgenic line released in Nigeria (approved transgenic event 709A). When visualised in an agarose gel, this PCR zygosity test produces a single band of 730bp if the sample is transgenic, a single band of 520bp if the sample is non-transgenic, or both bands if the sample was mixed (Figure 2). Each pool of seeds tested was scored as pure or mixed. The number of pools scored as mixed were entered as deviant pools for purity calculations using the “SeedCalc” method according to the ISTA standard (<https://www.seedtest.org/en/services-header/tools/statistics-committee/statistical-tools-seed-testing.html>).

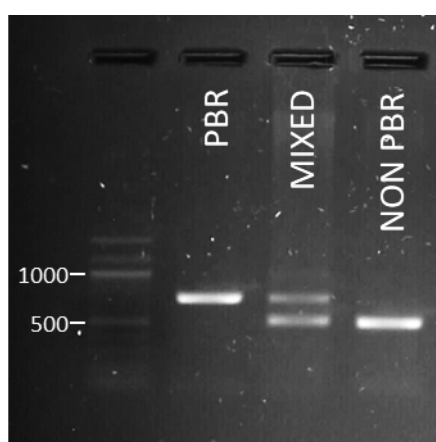


Figure 2. Gel analysis of the PCR fragments resulting from the zygosity test designed for the transgenic PBR cowpea (event 709A). When DNA from a transgenic PBR cowpea sample is used the test results in a single band of 730bp. If DNA from a non-transgenic sample is used

the test produces a band of 520bp. If the sample tested is mixed, then the two bands are produced.

Objective 3: Assess practice change, in particular due to compliance to stewardship programs.

For determination of yield in the PBR and non-PBR cowpea fields, all dried pods within each of the 5m x 5m demarcated area (same areas used for the insect studies) were harvested at plant maturity, threshed and then the grain weight measured. The yield for each field was expressed in kilograms per hectare.

For analysis of insecticide residues in dry grains, in each field 100g of seeds were randomly sampled and bulked based on whether it was PBR cowpea and number of sprays (2, 4, or 5) for non-PBR cowpea. From the bulked samples, a 600g sub-sample per category was taken and transported to the Department of Crop Protection, ABU Zaria. To evaluate the impact of sample storage on the presence of chemical residues, each 600g sample was divided into two portions of 300g. One portion was used for immediate analysis after harvest (0 storage) and the other was subjected to 4 weeks of storage (in a clean cheese cloth bag and kept on a laboratory bench). The extraction of insecticide residues from the grain samples was performed as follows. One hundred grams (100g) each of the PBR 2 sprays, non-PBR 2, 4 and 5 sprays for the 0- and 4-weeks storage were taken and milled to powder form at particle size of 0.1mm, using an MFC-90D micro-hammer mill (Culatti, Zürich, Switzerland). The pesticides were extracted from the samples using a modified QuEChERS method combined with a dispersive liquid-liquid microextraction (DLLME) procedure. Extracts were transferred to vials and 1µl of each was injected into the GC-MS system.

Certified reference mixed analytical insecticide standards containing Lambda-cyhalothrin, Cypermethrin, Dichlorvos, Dimethoate, Chlorpyrifos were obtained from analytical company (Accustandard, USA). The standards were prepared separately in acetonitrile (MeCN) at a concentration of 1000 mg/L and stored at -20°C until use. A mixed standard solution of 50 mg/L in MeCN containing all the mentioned pesticides was prepared. Working standard solutions of 0.1, 0.2, 0.5, 1.0, 2.0, 3.0, and 5.0 mg/L in MeCN were prepared and kept in a freezer at -20°C until use. The matrix-matched standard for the preparation of the calibration curve was made by adding multiple standards working solutions in the blank extracts of both matrices separately to reach the desired concentrations (0.01, 0.02, 0.05, 0.1, 0.2, 0.3, and 0.5 mg/kg) and stored at -20°C.

Gas Chromatography linked to mass spectrometry (GC-MS) was used to determine the insecticide residues in the PBR and non-PBR cowpea samples collected from farmers' fields. The sample extracts were analyzed using the Agilent Technologies network GC-MS system coupled with a universal detector. The model number of the column used was Agilent19091- 433UI capillary column with specification: 30 m x 0.25 mm id with 0.25µm film thickness (5% diphenyl, 95% dimethyl polysitoxane). The carrier gas was helium at a flow rate of 1ml/min. The oven temperature was initially programmed at 50°C for 2min and then increased by 8c/min to 300°C. A 25µl Glass Hamilton syringe was used to inject 1 µl of each sample into the GC machine. Ion count was used for compound identification and quantification. The spectrum of the separated compounds was compared with the database of known compounds saved in the reference spectra library. The records of many peaks were printed with retention times and quantification of the compounds. All the extractions and GC-MS analyses were performed at the Department of Chemistry, Ahmadu Bello University (Zaria).

Objective 4: Conduct an overall comparison of adopters of PBR cowpea versus non-adopters.

In each of the four agroecological zones, 20 PBR adopter and 20 non-adopter cowpea farmers respectively were randomly selected for the survey. A Team consisting of two socio-economics experts (Supervisors) were responsible for the survey with the assistance of well-trained enumerators. The team adopted a digital questionnaire as a data collection tool (designed using Open Data Kit-ODK Collect) to ensure good quality data. Enumerators were trained on how to use the Application before the inception of the data collection phase in the field. A pre-test survey of the questionnaire was carried out with 10 selected PBR adopters and 10 non-adopters. The pre-test was helpful in knowing which of the responses provided the required reliable information and identified faulty questions and statements in the draft. Thus, necessary additions, deletions, modifications, and adjustments were made to the questionnaire leveraging on the experience gained from the pre-test. The final data was collected from the zones from late October to late December 2022. Data was subjected to statistical analysis.

Objective 5. Assess the interest in PBR cowpea in Ghana and Burkina Faso.

In Ghana the study was conducted in three of the sixteen regions in which the country is divided: Northern region, Savannah region and Upper-East region. These three regions represent different agroecological zones where cowpea is majorly grown. Districts within these regions were selected for the study based on farmers exposure to or awareness of PBR cowpea through participation in the PBR cowpea on-farm trials and farmers field days. Tolon, Kumbungu and Nanton districts were selected from Northern region, West Gonja and Central Gonja districts from the Savannah region and West Mamprusi district from Upper-East region. From each of these districts, farmers were randomly selected to be interviewed for the study, while considering the population of farmers producing cowpea in each region and district. In total, 180 cowpea farmers were reached. The instrument used for the surveys was designed by the National Agricultural Extension and Research Liaison Services (NAERLS), Nigeria and later vetted by consultants from Ghana and other experts including the sponsors of the exercise. The instrument went through pre-test in Ghana and was later refined based on the comments observed during the pre-test exercise. The final survey instrument was designed using kobo toolbox to ensure ease of monitoring of field activities and accuracy with respect to coordinates capture. The data collection process started with training of field enumerators with diverse experience in agricultural research. The actual data collection lasted for seven days. Data was curated and processed and analysed statistically.

After the Ghana study was performed, and taking advantage of the experience, a similar study was conducted by a consultant hired by NAERLS in Burkina Faso with funding from AATF, in the Haut-Bassin (Wet zone) region. In this region, based on being major cowpea production areas, the Noumoudara, Peni, Taga, Darsalamy and Mes villages were visited, and 304 cowpea farmers were randomly selected and interviewed.

6 Achievements against activities and outputs/milestones

6.1 Objective 1: To Determine the field-based biological consequences (entomological effects, agronomic practices, yield) of PBR cowpea compared to conventional cowpea.

no.	activity	outputs/ milestones	completion date	comments
1.1	Entomological studies on target and non-target insects	<ul style="list-style-type: none"> - The result of the survey found the presence of 21 insect species of which 12 were pest, 7 predators and 2 pollinators. - No Maruca larvae were found in the PBR cowpea fields sprayed twice, however, Maruca larvae were found in the non-BPR cowpea fields sprayed 2,4, or 5 times in all zones - Other NTO were more abundant in the PBR (16 different insect species on average) and non-PBR (13 insect species) cowpea fields sprayed 2 times than the non-PBR cowpea fields sprayed 4 times (11 species) or 5 times (5 species). - The PBR cowpea field sprayed twice with insecticide had the highest cowpea grain yield compared to the non-PBR cowpea field sprayed up to and above 4 times. 	May 2023	Late commencement of the study in some locations resulted in some missing data as farmers had already harvested their crops at the time of the survey.
1.2	Soil biology studies	<ul style="list-style-type: none"> - The study revealed significant advantage of the PBR cowpea in harbouring higher population of bacteria and fungi in the rhizosphere soil relative to the non-PBR and the control. - The PBR cowpea with only two sprays showed significantly higher ability to retain microbial biomass carbon, nitrogen and phosphorus than the other two treatments. - Likewise, it showed significantly higher ability of the PBR cowpea to influence important chemical soil properties favourable to soil fertility, such as lowering of pH or acidifying the soil, higher amounts of organic carbon, total nitrogen and available phosphorus, some exchangeable cations such as calcium, magnesium and potassium and cation exchange capacity. - On the other hand, the non-PBR cowpea plots had significantly higher influence in the accumulation of Fe, and Zn, with an indication of the accumulation of Mn and Cu in the soils, relative to the PBR cowpea treatment. This could be attributed to the constituents of the pesticides applied in high doses. 	May 2023	Similar studies are typically conducted in three consecutive seasons. The study was conducted towards the end of the season, and it would have been preferred to start earlier to obtain information pre-sowing.

6.2 Objective 2: To assess the genetic purity of the PBR cowpea seed in farmers' fields.

no.	activity	outputs/ milestones	completion date	comments
2.1	Verification by random sampling and PCR test of seed purity after harvest from PBR cowpea adopters	Samples collected from Adopters fields are highly pure (97.28%), although some non-transgenic seeds were detected in some regions at a low frequency.	April 2023	
2.2	Verification by random sampling and PCR test of seed purity after harvest from PBR cowpea seed-savers	Samples collected from Seed-savers fields are highly pure, although some non-transgenic seeds were detected in some regions at a low frequency.	April 2023	
2.3	Verification by random sampling and PCR test of seed purity after harvest from non-adopters	Samples collected from non-adopters fields are were found non-transgenic, although some transgenic seeds were detected in some regions at a low frequency.	April 2023	

6.3 Objective 3: To Assess practice change, in particular, compliance to recommended field practices such as number of pesticides sprays.

no.	Activity	outputs/ milestones	completion date	comments
2.1	Evaluation of the effectiveness of only using 2 sprays, in terms of yield.	The grain yield was higher in the PBR cowpea sprayed 2 times than in the non-PBR cowpea sprayed 2 times.	May 2023	

2.2	Comparing yield in farms with different number of sprays.	The grain yield was higher in the PBR cowpea sprayed 2 times than in the non-PBR cowpea sprayed 2, 4 or 6 times.	May 2023	Late commencement of the study in some locations resulted in some missing data as farmers had already harvested their crops at the time of the survey.
3.3	Assessment of the use of common insecticides.	Percentage of usage of 6 common chemical insecticides in the 4 agroecological zones.	May 2023	
3.4	Analysis of residues of chemical insecticides in harvested grains.	In PBR cowpea seed samples there were no presence of chemical residues. In contrast, insecticide residues were detected in all non-PBR samples sprayed 2, 4 or 5 times. Residue levels in samples collected from farms sprayed 4 or 5 times were higher than the EU's MRL.	May 2023	Analysis of samples delayed due to method development efforts.

6.4 Objective 4: To Conduct an overall comparison of adopters of PBR cowpea versus non-adopters.

no.	activity	outputs/ milestones	completion date	comments
4.1	Surveying farmer impacts focussing on social perspective (changes in behaviour)	The rate of adoption (76% overall) of recommended practices by PBR cowpea farmers was fair considering the 2 years' time frame the practices has been introduced to the farmers but there is room for improvement. The adoption varied according to practice, high for some but low for others. The adoption of the practices however was higher for PBR farmers as compared to non-PBR farmers.	March 2023	
4.2	Yield assessment.	Significantly higher yields (approximately 2-fold difference) were obtained by the PBR cowpea adopters in all regions.	March 2023	

6.5 Objective 5: Assess the interest in PBR cowpea in Ghana and Burkina Faso

no.	activity	outputs/ milestones	completion date	comments
5.1	Detailed questionnaire used to gather information from farmers in Ghana.	<ul style="list-style-type: none"> - Survey carried out in 6 districts, and 180 cowpea farmers were reached. - Low (32.2% of interviewed farmers) awareness of the PBR cowpea variety was detected. - Farmers were very interested about the variety when explained to them. 	Surveys performed in January 2023	Report completed in August 2023

5.2	Detailed questionnaire used to gather information from farmers in Burkina Faso.	<ul style="list-style-type: none"> - Survey carried out in 5 villages, and 304 cowpea farmers were reached. - Low (40% of interviewed farmers) awareness of the PBR cowpea variety was detected. - Farmers were very interested about the variety when explained to them. 	Surveys performed in May 2023	Report completed in September 2023
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7 Key results and discussion

In this section we have highlighted the key results and learnings from each one of the 5 objectives in which this project was divided. The complete studies, including detailed methodologies, data, analysis, and conclusions can be found in the Appendixes included at the end of the report.

7.1 Determine the field-based biological consequences (entomological effects and soil effects) of PBR cowpea compared to conventional Cowpea.

1a. Insect studies

Insect pests has been a major challenge of cowpea production in sub-Saharan Africa. The legume pod borer (*Maruca vitrata*) is a major pest attacking cowpea which can cause a yield loss of about 20 to 80%. Chemical insecticides are widely used to manage insect pests on cowpea; however, there is always concerns on the effect of chemical residues on humans and environment resulting from indiscriminate use in cowpea production. The PBR cowpea variety has proven very effective in the control of the *M. vitrata* with promising grain yield. The PBR cowpea is now being cultivated by Nigerian farmers. However, we had no information on the influence of this new control practice on the non-target organisms in farmer's fields. Therefore, a survey was conducted to evaluate the impact of insecticide sprays on the insect population in the fields of adopters and nonadopters of PBR cowpea farmers in selected areas of four agroecological zones (Sahel, Sudan, Northern Guinea, and Southern Guinea) of Nigeria. The result of the survey has found the presence of 21 insect species of which 12 were pest, 7 predators of pests, and 2 pollinators. No *Maruca* larvae were found in the PBR cowpea fields sprayed twice, however, *Maruca* larvae were found in the non-PBR cowpea sprayed 2,4, or 5 times. Other non-target organisms (NTO) were significantly more abundant in the PBR and non-PBR cowpea sprayed 2 times than in the non-PBR cowpea sprayed multiple times (4 or 5).

As example, Table 1 shows the average number of different insect species found in PBR cowpea fields sprayed 2 times and non-PBR cowpea fields sprayed 2, 4 or 5 times in the northern guinea savannah of Nigeria. A total of 17 different insect species belonging to 11 different insect orders were found. Ten of the insect species were pest and 7 species were either predators or pollinators. There was no *Maruca* larvae found in the PBR cowpea field samples sprayed 2 times with insecticides, however between 3-5 *Maruca* larvae were found in the non-PBR cowpea fields sprayed 2, 4 or 5 times with insecticides, with 4 and 5 sprays having the least. All insect species were present in the PBR cowpea field sprayed 2 times. However, in the non-PBR cowpea field, some species were absent or occurred in a small number, particularly 4 and 5 sprays. This could be the result of frequent spraying, which can affect the abundance of biodiversity, and then of beneficial insects such as predators and pollinators. It is also worth noticing that the recommended agronomical management practices used by the PBR cowpea farmers can bring beneficial effects in the control on non-targeted pests (for example, for aphids the sowing window is a key factor). The results from the other agroecological zones show similar trends and can be found in the complete study in the Appendixes.

Table 1. Taxonomic order, status and number of insect species found in PBR and non-PBR cowpea farmers fields in the northern Guinea savannah of Nigeria.

Common Name	Order	Species name	Status	Average number of insect species/ 5m ²			
				BPR- 2 sprays	Non-PBR 2 sprays	Non-PBR 4 sprays	Non-PBR 5 sprays
Cowpea pod borer	Lepidoptera	<i>Maruca vitrata</i>	Pest	0 (0.0)	5 (1.2)	3 (1.3)	3 (1.4)
Cowpea Flower Thrips	Thysanoptera	<i>Megalurothrips sjostedti</i>	Pest	2 (0.7)	6 (1.4)	4 (1.7)	3 (0.4)
Cowpea Aphids	Hemiptera	<i>Aphis craccivora</i>	Pest	10 (4.0)	37 (9.0)	33 (5.7)	1 (0.8)
Brown pod-sucking bug	Hemiptera	<i>Clavigralla tomentosicollis</i>	Pest	4 (1.2)	5 (1.6)	4 (1.2)	4 (2.8)
Pod-sucking bug	Hemiptera	<i>Riptortus dentipes</i>	Pest	1 (0.4)	2 (0.5)	0 (0.0)	0 (0.0)
Black ant	Hymenoptera	<i>Componotus perris</i>	Predator	9 (1.5)	4 (2.0)	0 (0.0)	0 (0.0)
Surgar ant	Hymenoptera	<i>Componotus terebrans</i>	Predator	4 (1.5)	6 (3.7)	3 (1.5)	0 (0.0)
Honeybee	Hymenoptera	<i>Apis mellifera</i>	Pollinator	1 (0.3)	1 (0.2)	0 (0.0)	0 (0.0)
Housefly	Diptera	<i>Musca domestica</i>	Pollinator	1 (0.2)	1 (0.2)	1 (0.1)	0 (0.0)
Blister beetle	Coleoptera	<i>Mylabris fimbriatus</i>	Pest	2 (0.8)	1 (0.4)	2 (0.4)	2 (0.6)
Blister beetle	Coleoptera	<i>Mylabris phalerata</i>	Pest	1 (0.4)	1 (0.2)	1 (1.2)	0 (0.0)
Spittle bug	Homoptera	<i>Locris rubens</i>	Pest	3 (0.7)	1 (0.2)	0 (0.0)	0 (0.0)
Ladybirds beetle	Coleoptera	<i>Cheilomenes sulphurea</i>	Predator	1 (0.4)	0 (0.0)	0 (0.0)	0 (0.0)
Huntsman spider	Araneae	<i>Palystes Castaneus</i>	Predator	1 (0.4)	0 (0.0)	0 (0.0)	0 (0.0)
Leafhopper	Hemiptera	<i>Empoasca dolichi</i>	Pest	1 (0.8)	2 (1.2)	2 (0.8)	12 (2.7)
Earwig	Dermaptera	<i>Forficula senegalensis</i>	Predator	2 (0.8)	1 (0.2)	1 (0.5)	0 (0.0)
Bushcricket	Orthoptera	<i>Phaneroptera nana</i>	Pest	1 (0.6)	0 (0.0)	0 (0.0)	0 (0.0)

The figures in bracket are standard deviations.

1b. Soil studies

The soil microbial study revealed significant advantage of the PBR cowpea to harbour higher population of bacteria and fungi in the rhizosphere soil than the non-PBR (Figure 3), allowing for more nitrogen fixation, nitrification, decomposition of organic matter and solubilization of soil nutrients.

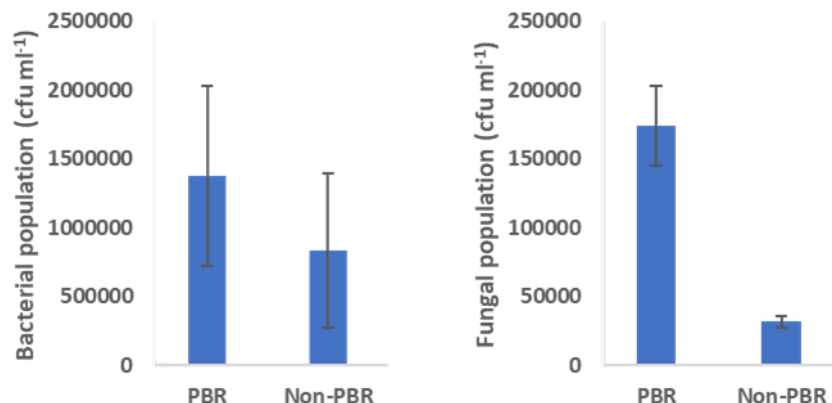


Figure 3. Effect of the PBR cowpea on bacterial and fungal populations relative to non-PBR plots. Pooled results from the four agroecological zones of Nigeria are shown.

The study also showed that PBR cowpea had significantly higher ability to retain microbial biomass carbon, nitrogen, and phosphorus than the non-PBR. The use of PBR cowpea in the study showed significantly higher influence for important chemical soil properties favourable to soil fertility, such as higher amounts of organic carbon, total nitrogen and available phosphorus, some exchangeable cations such as calcium, magnesium and potassium and cation exchange capacity (Figure 4). These advantages of the PBR cowpea could be attributed to the reduced number sprays of pesticides and improved production practices not obtainable in the non-PBR soils.

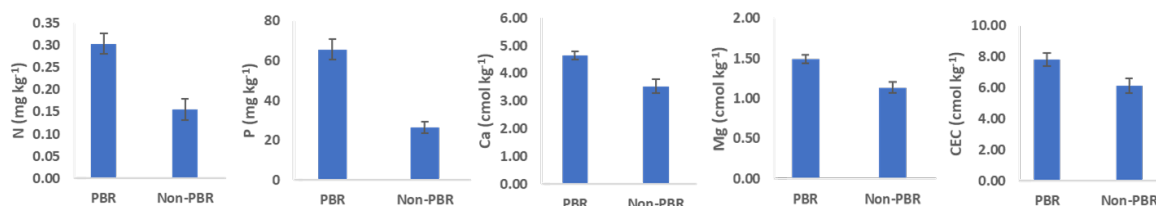


Figure 4. Effect of the PBR cowpea on nitrogen, phosphorus, calcium, magnesium and cation exchange capacity relative to non-PBR plots. Pooled results from the four agroecological zones of Nigeria are shown.

On the other hand, in the non-PBR cowpea plots, a minor increase on the accumulation of Fe, and Zn was detected (Figure 5), together with a non-significant indication of the accumulation of Mn and Cu in the soils, relative to the PBR cowpea soils. This could be attributed to the constituents of the pesticides applied in high doses, of these elements are part, hence the need for caution to avoid excess accumulation to toxic levels with time.

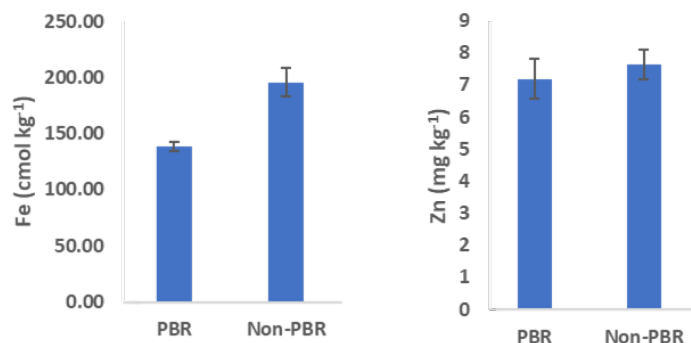


Figure 5. Effect of the PBR cowpea on the accumulation of iron and zinc in the soil relative to non-PBR plots. Pooled results from the four agroecological zones of Nigeria are shown.

7.2 Assess the genetic purity of the PBR cowpea seed in farmer's fields.

Three groups of farmers were used for the purposes of this genetic purity test. It became necessary to add a third class of farmers which was not in the original protocol because, history showed that some of them saved their seeds and didn't buy from licenced seed companies. The three classes of farmers were thus: The adopters, the non-adopters, and the seed savers group.

Results of the genetic purity tests (Table 2) showed that samples collected from the farmer group classified as Adopters (those who bought directly from licenced seed companies) had a 97.28% purity overall, which is quite high. However, deviant samples with some contaminated seeds were found in 3 of the 4 agroecological regions. Similarly, results from the farmer group classified as Seed savers (those who bought retained seed from the previous season) had a 97.24% purity overall. Again, deviant samples with some contaminated seeds were found in other 3 of the 4 agroecological regions. Finally, results from the non-adopters group indicated the 93.51% of the samples are non-transgenic. However, we were able to identify the presence of some transgenic seed in 3 of the 4 agroecological regions. In one case (Non-Adopters Site 4), most of the seeds were PBR cowpea seeds, which most likely is explained by an error in labelling the site samples.

These results can be interpreted in several ways. The high and similar purity% of the PBR seed found in the adopters and seed savers fields could imply that during the two years in which this PBR cowpea product has been with farmers, there has not been a significant breakdown of purity levels. We think the presence of some non-PBR cowpea seeds in some of the samples is likely due to postharvest handling, which could have introduced some contamination. Regarding the analysis of the seed samples from non-adopters' farms, we have been able to detect the presence of some transgenic seeds. These results are not surprising giving that different seed lots from different farmers or fields can come to proximity during the harvesting, threshing, or packing activities. For instance, it was observed that some sampled PBR cowpea (Sampea 20T) farmers also had other fields where they planted non-PBR cowpea (Sampea 14 or 15). In this case, that can explain that some of the non-PBR cowpea fields had stands of PBR cowpea which could be due to post harvest handling or even volunteers from fields previously planted with PBR cowpea.

Table 2. Summary of seed purity tests performed on samples collected from Adopters, Seed Savers and Non-Adopters fields from all agroecological zones.

	Sahel Savanna	Northern Guinea Savanna	Sudan Savanna	Southern Guinea Savana	ALL
Adopters Site 1	98.51	98.51	98.51	98.51	
Adopters Site 2	98.51	98.51	98.51	98.51	
Adopters Site 3	98.51	98.51	98.51	98.51	
Adopters Site 4	92.76*	98.51	95.44*	98.51	
Adopters Site 5	92.76*	98.51	92.76*	94.21*	
Adopters ALL	96.21	98.51	96.746	97.65	97.28
Seed Savers Site 1	98.51	98.51	98.51	98.51	
Seed Savers Site 2	98.51	97.53*	98.51	95.44*	
Seed Savers Site 3	98.51	95.44*	98.51	96.53*	
Seed Savers Site 4	98.51	95.44*	98.51	94.21*	
Seed Savers Site 5	98.51	95.44*	98.51	92.76*	
Seed Savers ALL	98.51	96.472	98.51	95.49	97.24
Non-Adopters Site 1	98.51	98.51	97.53*	98.51	
Non-Adopters Site 2	98.51	98.51	98.51	98.51	
Non-Adopters Site 3	97.53*	98.51	98.51	98.51	
Non-Adopters Site 4	2.47*	97.53*	98.51	98.51	
Non-Adopters Site 5	98.51	97.53*	98.51	98.51	
Non-Adopters ALL	79.106	98.118	98.314	98.51	93.51

*Indicates sites were deviant pools with contaminated/mixed seeds were found.

7.3 Assess practice change, in particular compliance to stewardship program (yield and residues of chemical insecticides).

Using the same study fields selected for the insect studies, a survey was conducted to evaluate the impact of insecticide sprays on cowpea yield and the presence of insecticide residue in grains harvested from the fields of PBR cowpea adopter and non-adopter farmers.

3a. Yield study in relation to the use of chemical insecticides

The PBR cowpea field sprayed twice with insecticide had the highest cowpea grain yield compared to the non-PBR cowpea field sprayed up to 5 times. Table 3 shows the cowpea grain yield of PBR cowpea 2 sprays and non-PBR cowpea 2, 4 or 5 sprays fields in the four Agro-ecological zones of Nigeria. The PBR cowpea fields, sprayed twice, had the highest gran yield compared to the 2, 4 or 5 sprays of non-PBR cowpea fields in all the Agro-ecological zones.

Table 3. PBR and Non-PBR cowpea grain yield after harvest at four Agro-ecological zones of Nigeria.

Cowpea grain yield Kg/ha

Location	PBR- 2 sprays	Non-PBR-2 sprays	Non-PBR 4 sprays	Non-PBR 5 sprays	SE±
Northern Guinea	1476 ^a	796 ^b	1248 ^a	1533 ^a	195
Southern Guinea	1720 ^a	696 ^c	1252 ^b	1557 ^a	105
Sahel Savannah	1226 ^a	*	919 ^b	*	122
Sudan Savannah	1521 ^a	*	1291 ^a	*	187

*Data not available: The survey commenced at the late season when most farmers harvested their cowpea; farmers under this category in the Sahel and Sudan savannah were missed out. Superscript letters indicate statistical comparisons. Means with the same letter along the row are not statistically significant at $p < 0.05$.

3b. Presence of residues of chemical insecticides in harvested grain samples

The result of the chemical residue analysis using GC-MS detected 6 common insecticide compounds (Chlorpyrifos, Cypermethrin, Methyl parathion, Lamdacyhalothrin, Dimethoate, Fenpropathrin) in the samples (Table 4). Only Lamdacyhalothrin and Fenpropathrin were detected in the PBR cowpea 2 sprays, with a residue concentration below the EU's MRL. The residue level of Cypermethrin sprayed 5 times (0.06mg/kg), Methyl parathion sprayed 4 and 5 times (0.05mg/kg), Dimethoate sprayed 4 (0.09mg/kg) and 5 (0.06mg/kg) times and Fenpropidin sprayed 4 (0.02mg/kg) and 5(0.03mg/kg) times in the non-PBR cowpea were higher than the EU's Maximum Residue Limits. The concentration of some of the compounds was affected by the storage period (0 or 4 weeks) of the samples. Farmers need to be made aware of the significant health benefits of adopting the PBR cowpea technology. Multiple spray regimes that result in such high levels of pesticide residues are harmful to humans, animals and the environment and should be discouraged. This study also reveals part of the reasons why cowpeas from Nigeria have been previously rejected in the international market due to high pesticide residue levels.

7.4 Conduct an overall comparison of adopters of PBR cowpea versus non-adopters.

The results of the survey of the adoption of the PBR cowpea recommended management practices (Figure 6) by PBR cowpea farmers in the four agroecological zones is presented in Table 5. We would like to highlight here that the results related to the adoption of recommended spray regimes for pests and diseases (2 sprays, at vegetative and flowering stage) was high amongst the farmers (76% overall), and when compared across zones, it was higher in Northern Guinea (85%) and Southern Guinea (80%) and slightly lower in Sahel (65%) and Sudan (75%). This is in our view one of the most important recommendations and one that can bring significant economic, health and environmental benefits.

PBR Cowpea management recommended practices:

- Seed treatment: Use either Apron star or Allstar (Fungicides) at the rate of 10 g Sachet/ 4 kg of seeds. Seed treatment protects against fungal infections and ensures good germination.
- Land preparations: Ploughing (new land), harrowing (twice, depending on soil), ridging (tractor or animal-drawn implements).
- Seed rate: 20-25 kg/ha.
- Sowing time: Sahel (3rd week of June), Sudan Savannah (1st to 2nd week of July), Northern Guinea Savannah (3rd week of July to 1st week of August), Southern Guinea Savannah (1st to 3rd week of August).
- Spacing Between rows: 75 cm between rows, 20 cm within rows.
- Thinning: May be required at 2 weeks after sowing if manually planted with more than 2 plants per hill
- Fertilizer application: 50kg/ha, 2 bags at land preparation or sowing, 3 – 4 bags at sowing.
- Weed Management: Land preparation, spray: Glyphosate at 4 litres/ha, use green reflector nozzle. Manual weeding: 1st weeding (2 to 3 weeks after planting). 2nd weeding (5 to 6 weeks after planting). Chemical weed control: Use selective herbicide, such as pendimethalin, at the rate of 4L/ ha immediately after planting (apply pre-emergence).
- Pests and Diseases Management: Fungal diseases are the most prevalent. Spray fungicides, such as Mancozeb, preferably a systemic fungicide like Carbendazim at the rate of 2L/ha. For insects spray with: Lambda-cyhalothrin (100 -150 ml/20L of water) or Cypermethrin + Dimethoate at same rate above. At flowering and pod development: 1st at 45 days after Sowing (DAS) and 2nd at 55 DAS. If more pod sucking bugs observed and pods are still forming, a 3rd spray is required at 7 days interval.
- Maturity: Early maturing (70 – 75 days).
- Harvesting/Threshing: Harvest when the cowpea pods are fully mature and dry (pods with brown colour). The pods should be picked properly and sun-dried. Thresh either manually or use a threshing machine and ensure grains are not mixed during threshing and throughout the post-harvest operations.
- Bagging/Storage: Harvested pods should be threshed, winnowed, sorted, and cleaned properly. Use Purdue Improve Crop Storage (PICS) bags for storage after drying seeds to a moisture content of 10 – 12%.

Figure 6. PBR Cowpea management recommended practices. These recommendations are part of the Technology Use Guide (TUG) for PBR Cowpea farmers, developed by AATF.

The survey reveals other practices that were adopted relatively highly. For example, 40% of PBR cowpea farmers adopted the recommended seed treatment management practices in the Sahel region, 55% in Sudan, 80% in Northern Guinea, and 70% in Southern Guinea region. From the pooled results, 61% of PBR cowpea farmers adopted the recommended seed treatment in the sampled zones. The majority of PBR cowpea farmers adopted recommended land preparations practices with a pooled result of 86%.

On the other hand, the survey indicated that some other recommended practices are less adopted, with less than half of the farmers adopting them. The rate of adoption of the practices with low scores for the pooled farmers were 48%, 43%, 28%, 20% and 19% for row spacing, weed management, triple bagging, fertilizer application and thinning respectively. Results were very similar in all the agro-ecologies, and this could suggest the

need for an increase in awareness or for a review of those practices to decide if they are or are not critical for cowpea production.

Table 4. Concentration (mg/kg) of the insecticide residues in the PBR and Non-PBR cowpea grain sample in four Agro-ecological zones of Nigeria.

Cowpea Sample	Chlorpyrifos	Cypermethrin	Methyl parathion	Lamdacyhalothrin	Dimethoate	Fenpropathrin
PBR 2 sprays 0 storage	ND	ND	ND	0.01	ND	0.001
PBR 2 sprays 4 weeks storage	ND	ND	ND	ND	ND	ND
Non-PBR 2 sprays 0 storage	0.02	0.03	0.01	ND	0.03	0.01
Non-PBR 2 sprays 4 weeks storage	0.01	0.02	0.01	ND	0.03	0.01
Non-PBR 4 sprays 0 storage	0.01	0.04	0.05	0.02	0.09	0.02
Non-PBR 4 sprays 4 weeks storage	0.01	0.03	0.03	0.02	0.09	0.02
Non-PBR 5 sprays 0 storage	0.02	0.06	0.05	0.01	0.06	0.03
Non-PBR 5 sprays 4 weeks storage	0.02	0.06	0.04	0.01	0.05	0.03
EU's MRL*	0.05	0.05	0.01	0.01	0.02	0.01

MRL: Maximum Residue Limits; ND: not detected.

Table 5. Adoption of PBR cowpea recommended practices by farmers.

Recommended crop management practices	Sahel		Sudan		Northern Guinea		Southern Guinea		Pooled	
	Freq.	%	Freq.	%	Freq.	%	Freq.	%	Freq.	%
Seed treatment	8	40	11	55	16	80	14	70	49	61
Land preparations	18	90	16	80	17	85	18	90	69	86
Seed rate (20-25 kg/ha)	15	75	17	85	19	95	18	90	69	86
Sowing time	17	85	15	75	19	95	19	95	70	88
Spacing Between rows	9	45	11	55	8	40	10	50	38	48
Thinning	2	10	4	20	3	15	6	30	15	19
Fertilizer application (50kg/ha)	3	15	2	10	4	20	7	35	16	20
Weed Management	8	40	10	50	7	35	9	45	34	43
Pests and Diseases Management	13	65	15	75	17	85	16	80	61	76
Maturity	17	85	15	75	19	95	19	95	70	88
Harvesting/Threshing	20	100	20	100	20	100	20	100	80	100
Bagging/Storage	3	15	5	25	8	40	6	30	22	28

The results for the comparative analysis of PBR farmers and non-PBR farmers with respect to practices is presented in Table 6. The analysis revealed that PBR Farmers from all the agroecological zones used seed treatment practice (61%) more than the non-PBR farmers

who represent only 5%. This implies that the majority of farmers who used PBR seed adopted treatment technology in relation to the farmers that used conventional seeds. Likewise, while 86% of PBR farmers applied the recommended seed rate, only 20% non-PBR farmers used the recommended practice. Regarding pest and disease management the adoption rate in PBR farmers was significantly higher (76%) than for the non PBR farmers (30%)

From the results, it was observed that there was a high compliance to sowing time for both PBR and non-PBR farmers across all zones though slightly higher for PBR farmers (88%) than non-PBR farmers (78%). In the same way there was a high percentage of both groups in the use of recommended land preparations practice (86% PBR farmers and 68% non-PBR farmers). There was also high adherence to use of recommended maturity date and harvesting and threshing technique by the two categories of farmers across the zones. About 88% and 64% of PBR and non-PBR farmers respectively allowed their cowpea to reach the recommended maturity period of 70 – 90 days before harvest. On further enquiry, PBR farmers said that they harvest earlier as the crop matures earlier than the recommended period, while non-PBR farmers harvest later than recommended period as conventional cowpea takes longer to mature. Compliance to recommended harvesting and threshing technique for PBR farmers was 100% and 80% for non-PBR farmers.

Table 6. Comparative analysis of PBR cowpea adopters and non-adopters with respect to Practices.

Recommended crop management practices	PBR Farmers		Non-PBR Farmers	
	Freq.	%	Freq.	%
Seed treatment:	49	61	4	5
Land preparations	69	86	54	68
Seed rate (kg/ha): 20 – 25 kg/ha	69	86	16	20
Sowing time	70	88	62	78
Spacing Between rows:	38	48	2	3
Thinning	15	19	26	33
Fertilizer application (50kg Bag/ha)	16	20	0	0
Weed Management:	34	43	5	6
Pests and Diseases Management:	61	76	24	30
Maturity:	70	88	51	64
Harvesting/Threshing:	80	100	64	80
Bagging/Storage:	22	28	6	8

Conversely, the compliance with the use of some recommended practices (spacing, thinning, fertilizer rate, weed management, and bagging/storage) was low for both groups. But even in those, the rates were lower for non-PBR farmers when compared to PBR farmers for the recommended practices listed in the parenthesis above except for the thinning process probably because they did not adhere to recommended seed rate. Specifically, farmers who used PBR applied the recommended spacing between rows method (48%) more than the conventional cowpea farmers (3%) in the four ecological zones. Similarly, 43% of PBR adopters applied the recommended weed management practice, while only 6% non-PBR farmers followed the method. For recommended practice

of bagging and storage, 28% of the PBR farmers adopted bagging and storage recommended practice, while only 8% non-PBR farmers considered the practice, indicating that PBR farmers were more involved in the practice. For thinning practice, 19% of PBR farmers applied the recommended practice as against a higher value of 33% for non-PBR farmers.

In terms of crop performance measured as grain yield, PBR cowpea farmers did it significantly better. Figure 7 shows the yield obtained by PBR and non-PBR farmers from the four zones and the pooled total. There was a general higher yield obtained by PBR farmers in comparison to non-PBR farmers in all the zones. When all the zones were pooled together, the was about 2 tons for PBR cowpea. For non-PBR cowpea however, the yields for the zones were far lower. Only Sudan zone had yield slightly above 1 ton per hectare as the others were either at 1 ton or below. When all the zones were pooled the average yield stood at about 0.9 ton per hectare.

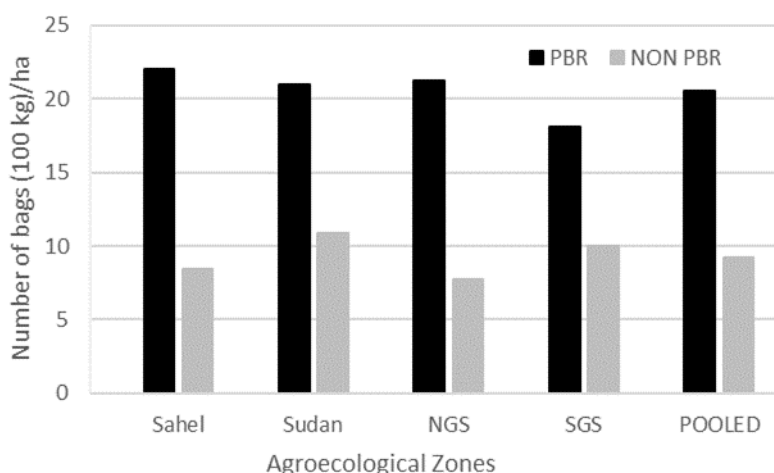


Figure 7. Yield obtained by PBR adopters and non-PBR farmers in the study areas.

In conclusion, the rate of adoption of recommended practices by PBR farmers was fair considering the time frame the practices were introduced to the farmers but there is room for improvement. The adoption varied according to practice, high for some but low for others. The adoption of the practices however was higher for PBR farmers as compared to non-PBR farmers. All this resulted in a two-fold increase in yield obtained by the PBR farmers.

7.5 Assess the interest in PBR cowpea in Ghana and Burkina Faso

Farmers perceptions of the PBR cowpea in Ghana

Structured checklists were designed, digitized into ODK research instrument and distributed among 180 farmers that cut-across the northern, savannah and north-east regions of Ghana. Majority (72.2%) of farmers reached were from the Northern region because it's the region with largest cowpea farmers and also the site where the PBR cowpea demonstration plots were located.

Cowpea farmers within the age bracket of 36-59 years formed the majority of 61.1% in Ghana and about 62% of the entire farmers reached are the heads of households. Just like in most developing nations, only few (13.3%) of the farmers reached were females. Use of

reading materials for extension delivery services can be regarded as almost ineffective as over 65.6% of interviewed farmers had no formal education and could possibly be a challenge when using print extension publications. Crop production had a response of 96.1% among farmers implying the major livelihood means therein. An average of 11 household members was recorded and 44.4% of the farmers had between 16-30 years of experience in crop production. As for experience in cowpea production, 90% of the farmers had experience of less than 15 years. Cowpea farmers in the country are small scale in nature and had 88.3% of farmers reached operating between landholdings of 1.0-2.5 acres having an average of 1.7 acres.

Our study shows that awareness level of the PBR cowpea amongst farmers in Ghana is quite low (32.2%) and the information source is majorly from extension agents (57.8%), Mass media (55.6%) and the least (7.2%) from NGOs. Once the farmers were informed about PBR cowpea, over 71% of farmers reported PBR cowpea as an important topic for discussion and 96.7% of cowpea farmers interviewed indicated intentions to produce PBR cowpea in the next 5 years. In terms of willingness to pay for PBR cowpea seeds, 63.9% of farmers indicate intentions to pay. This result implies that majority of the respondents are willing to commit their personal resources for them to enable them to grow this variety in regions visited in Ghana. Figure 8 shows the acceptability of PBR cowpea among the respondents. 62.22% of the respondents indicated that it will likely be accepted, and 33.89% affirming that it will very likely be accepted, with only 2.22% of the respondents who are undecided. This result implies that the acceptability rate/potentials of the PBR cowpea is very high.

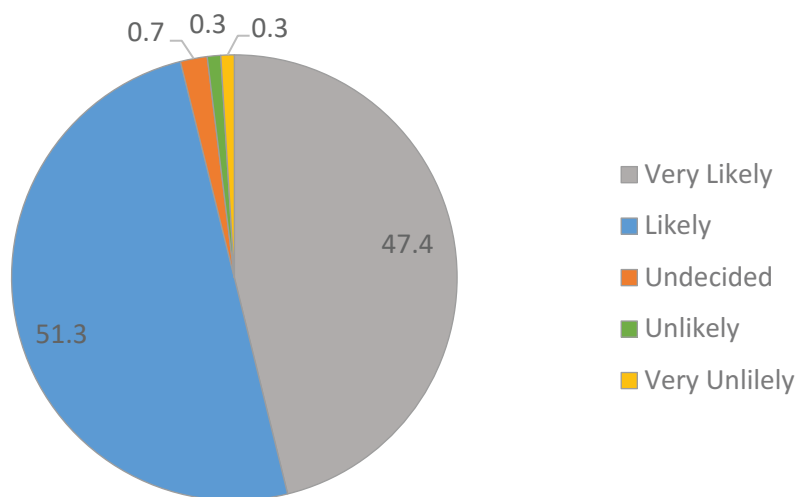


Figure 8. Acceptability of the PBR cowpea variety. The graph indicates the percentage of respondents that would likely to accept this product.

The result in Table 7 provide insight on the key performance indicators for Pod-borer resistant cowpea. From the table, 61.67% of the respondents agreed that cultivating PBR cowpea will give a positive advantage to cowpea farm as non-use of chemicals will aide soil microbes development thus improves soil fertility and structure, and that 56.67% also agreed that PBR cowpea is expected to save cost of applying chemicals thus brings about diversification, while about 46.67% of the respondents are indifference to the fact that this variety is expected to improve farmers access to credit since the fear of pod borer infestation is been taken care of. The PBR cowpea is therefore expected to remediate a major threat faced in cowpea production which is crop loss due to pest infestation and the problem of food poisoning that has caused lots of problems in recent times to consumers.

Table 7. Pod-Borer Resistant cowpea Performance Indicators.

Performance Indicators (%)	SA	A	I	D	SD
Cultivating PBR cowpea will give a positive advantage to cowpea farm (Non use of Chemicals will aide soil microbes development thus improves soil fertility and structure)	7.78	61.67	24.44	6.11	-
PBR cowpea will reduce the chances of having food poisoning and environmental pollution	16.11	53.89	27.22	2.78	-
PBR cowpea will fetch more market price than conventional cowpea	8.89	50	36.67	4.44	-
Will Ensure sustainable crop cultivation over other cowpea seeds	4.44	49.44	39.44	6.67	-
PBR cowpea is expected to save cost of applying chemicals thus brings about diversification	14.44	56.67	26.11	2.78	-
PBR cowpea will generate higher yield, income which translate to enhanced living standard	11.11	50.56	33.33	4.44	0.56
Expected to improve farmers access to credit since the fear of pod borer infestation is been taken care of	3.33	37.22	46.67	12.78	-

SA= Strongly agree, A= Agree, I= Indifference, D= Disagree, SD= Strongly disagree

In summary, the general conclusion from the field survey in Ghana is that farmers were not generally aware of the PBR cowpea variety. Since it is not yet a released variety, this is not totally surprising. However, once informed, farmers were very excited about the variety when explained to them. Many of the farmers who took part in the last field demonstrations expressed that the project should release the PBR cowpea seed as soon as possible. Since farmers are however not so exposed to the seeds some of the perception indicators were not particularly in favour of the variety.

Farmers perceptions of the PBR cowpea in Burkina Faso

Like in the study performed in Ghana, structured checklists were designed, digitized into ODK research instrument and distributed among 304 farmers that cut-across the Noumoudara, Peni, Taga, Darsalamy and Mes villages of Haut-Bassin (Wet Zone) of Burkina Faso. Majority (74.7%) of farmers reached were males. Cowpea farmers within the age bracket of 35-44 years formed the majority (30.9%) and about 91% of the farmers reached were married. A significant proportion (39.5%) of farmers had no formal education and the majority (92.8%) had crop production as their major source of livelihood while 34.1% had animal husbandry as secondary occupation. The major information source for agricultural topics used by farmers is through family which accounted for 60.5%.

Awareness level of PBR cowpea was relatively low as only 40% of farmers reported been aware of the variety, and for those, agricultural-related meetings (84.4%) was the main source of the information. In Burkina, the most important challenge in cowpea production identified were pest control (99.3%), quality of seeds (62.8%) and seed storage (58.6%). Ninety-six percent (96%) of cowpea farmers indicated willingness to accept an alternative over the seed variety that they currently use and 48.0% indicated PBR cowpea discussion to be very important, hence perceiving the PBR cowpea as a better alternative. Farmers further proved the importance of the PBR cowpea by indicating their willingness to pay for this variety, with the majority (45.4%) indicating they would pay between 500-1000 FCFA for a kg of the PBR cowpea seed.

Figure 9 shows the acceptability of PBR cowpea among the respondents. 51.3% of the respondents indicated that they will likely accept it, and 47.4% responded that they will very likely accept it. The respondents that indicated that were unlikely to accept it were only 0.3% and another 0.3% were very unlikely. 0.7% of the respondents were undecided. This result implies that the acceptability rate/potentials of the PBR cowpea is very high.

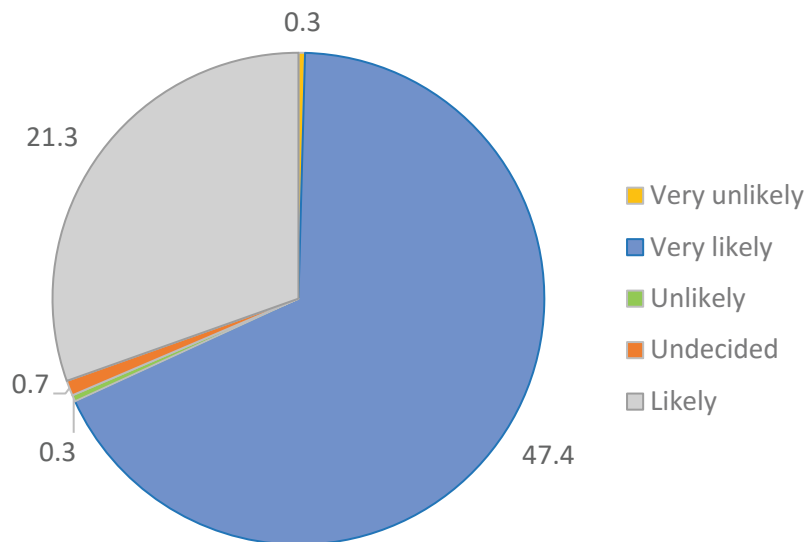


Figure 9. Acceptability of the PBR cowpea variety in Burkina Faso. The graph indicates the percentage of respondents that would likely accept this product.

The survey indicated that all the PBR cowpea performance indicators were positively evaluated by the cowpea farmers, which supports the positive attributes of this variety. About 63.2% of the respondents agreed that PBR cowpea will ensure sustainable crop cultivation over other cowpea seeds, 61.5% also agreed that the PBR cowpea is expected to save costs of applying chemicals. About sixty percent (60%) agreed that PBR cowpea will generate higher yield, income which translates to enhanced living standards. The PBR cowpea is therefore expected to provide solutions to major threats faced in cowpea production which are crop waste and/or loss due to pest infestation and the case of food poisoning that has caused lots of health issues in recent times to consumers.

In summary, and similarly to the results found in Gnana, the general conclusion from the field survey in Burkina Faso is that farmers were not generally aware of the PBR cowpea variety. Probably because it is not yet a released variety. Again, in Burkina Faso, farmers were very excited about the variety when explained to them.

8 Impacts

8.1 Scientific impacts – now and in 5 years

The immediate scientific impacts of this SRA are:

- 1- Demonstration of the positive effect of PBR cowpea on non-targeted organisms. The entomological survey conducted has shown that the pest management practices associated with the PBR cowpea variety result in an increased presence of beneficial insects (pollinators and predators).
- 2- Demonstration of the positive effect of PBR cowpea on the population of bacteria and fungi in the rhizosphere. This could be translated into better soil characteristics associated with functions such as nitrogen fixation, decomposition of organic matter and solubilization of soil nutrients.
- 3- Demonstration of the positive effect of PBR cowpea on the physical and chemical characteristics of the soil. Our soil studies indicate an increase in nitrogen, phosphorus, and carbon as well as in exchangeable cations such as calcium, magnesium, and potassium. We have also detected a trend of reduced levels of iron, zinc, manganese and copper, which could be associated the reduction of chemical insecticides sprays.
- 4- Demonstration of the beneficial impact of PBR cowpea on grain chemical contamination. Our analysis of PBR cowpea grains and non-PBR cowpea grains show that the presence of residues of six common chemical insecticides was non detected in PBR cowpea samples, while they were found in non-PBR cowpea grains, sometimes at levels higher than those recommended by the European Union.
- 5- Indication of no critical breakdown on farmers' fields of the genetic purity of the PBR cowpea grains after two cropping seasons. Our PCR analysis have found very low levels of contamination between the PBR and non-PBR fields.
- 6- Demonstration of a highly significant increase in yield associated with the use of PBR cowpea. Our survey has detected a two-fold increase in yield across the four agro-ecologies of Nigeria.
- 7- Demonstration of the implementation of the PBR cowpea recommended crop management practices. Our survey demonstrates a positive trend in adoption of the recommended practices by adopter farmers. This results in better agronomical management, diligence, and outcomes by the PBR cowpea adopters when compared to non-adopter farmers.

Following on this, in the next 5 years we will expect these impacts:

- 1- Confirmation of the positive effects of the PBR cowpea by performing multiyear and multilocation studies.
- 2- Deployment of the PBR cowpea in Ghana and Burkina Faso.

8.2 Capacity impacts – now and in 5 years

In this SRA we have established some methods and identified key qualified researchers to monitor the PBR cowpea performance. This effort has resulted in several capacity impacts:

- 1- Establishment of a network of sentinel PBR cowpea farmers that can be used for future follow-up studies.
- 2- Establishment of protocols and methods to measure the impact of PBR cowpea in several scientific fields.

- 3- Establishment of the methodology for measuring chemical residues in grain samples.
- 4- Development of a network of extension officers that have been trained to survey farms in Nigeria, Ghana and Burkina Faso.

In 5 years, we expect the consolidation of a set of standard protocols developed and tested for the study and assessment of the PBR cowpea product. We also expect the strengthening of the network of trained personnel in Nigeria, Ghana and Burkina Faso able to evaluate biotechnology products.

8.3 Community impacts – now and in 5 years

It is expected that many of the scientific and capacity impacts describe above will be translated into significant community impacts if the PBR cowpea was deployed at scale.

8.3.1 Economic impacts

- 1- Significant increase in yield associated with the use of PBR cowpea. Our survey has detected a two-fold increase in yield across the four agro-ecologies of Nigeria.

In 5 years, we could expect about 10 million people adopting the PBR cowpea, which will be translated in unparalleled economic benefits for farmers because of yields gains and savings in chemical sprays. If the success of the PBR cowpea in Nigeria continues and extends to Ghana and Burkina Faso, we may see a wave effect in other African countries who may embrace other biotech crops.

8.3.2 Social impacts

- 1- Beneficial impact of PBR cowpea on grain chemical contamination. Our analysis of PBR cowpea grains and non-PBR cowpea grains show that the presence of residues of six common chemical insecticides was non detected in PBR cowpea samples, while they were found in non-PBR cowpea grains, sometimes at levels higher than those recommended by the European Union.
- 2- Positive acceptance of the PBR cowpea variety in Ghana and Burkina Faso. While our extensive survey has shown relatively low levels of awareness about the PBR cowpea, it has clearly indicated that its attributes and benefits are highly desirable, and that the farmer community will very likely embrace its deployment in both countries.

In 5 years, with adequate deployment and adoption at scale of the PBR cowpea, we will see a massive decrease in the use of chemical insecticides, which will have a direct impact in improving food safety and human health, by reducing the current number of cases of food poisonings for consuming grains contaminated with chemical residues.

8.3.3 Environmental impacts

- 1- Positive effect of PBR cowpea on non-targeted organisms. The entomological survey conducted has shown that the pest management practices associated with the PBR cowpea variety result in an increased presence of beneficial insects (pollinators and predators).
- 2- Positive impact in soil biological, physical, and chemical properties.

In 5 years, the adoption of PBR cowpea will significantly reduce the damage on the populations of beneficial insects produced by indiscriminate application of insecticides. This has been the case for other Bt crops such as cotton, where its use has improved the numbers of beneficial insects. In the same way, the decrease in the use of chemicals will positively impact the soil health, thus its fertility.

8.4 Communication and dissemination activities

It is expected that the results from this SRA will be shared in relevant publications reaching farmers and consumers, although a plan for communication and dissemination activities have not yet been prepared. Such a plan should be prepared with high priority, and project partners must engage with the relevant bodies to publicise some of the key findings of this SRA. These activities should be focussed on how the use of the PBR cowpea could bring significant benefits to farmers, consumers, and the environment:

- Better yields.
- Reduction in pesticide residues in grains.
- Protection of beneficial insects.
- Improvement of soil characteristics.

9 Conclusions and recommendations

This SRA was designed to provide an early and quick evaluation of the Pod-Borer Resistant cowpea with the aim of monitoring its release in Nigeria. This activity has provided a first attempt to measure several key aspects of this new biotech product in farmers' fields. We have established the relevant methods and protocols, identified key local expertise to develop the project and found significant positive impacts in numerous critical areas. However the study did not compare properly matched samples of adopting and non-adopting farmers. Therefore the yield impact of the biotechnological innovation cannot be ascertained since it is confounded with the adoption of other production practices (improved varieties and improved agronomy). Furthermore, the study did not provide socio-economic information about adopting farmers compared to non-adopters. Understanding adoption better would inform future dissemination of the technology. The compliance of adopting farmers with the stewardship requirements of the biotechnological innovation (for example the use of refugia crop) was not explored. This study sets the basis for future larger and longer-term studies, with a focus on the socio-economic drivers of adoption, stewardship, and impacts.

9.1 Conclusions

These are the conclusion for each of the research topics:

1. Field-based biological consequences (entomological and soil effects) of PBR cowpea compared to conventional cowpea.

The adoption of the PBR cowpea and its recommended pest-management practices, shows great efficacy against the target pest, the legume pod-borer *Maruca*, while it has beneficial effects on the populations of non-targeted insects, i.e., pollinators and predators. Similarly, the farm soils where PBR cowpea is cultivated show healthier biological properties and better physical and chemical properties. These effects can be attributed to the reduced number of chemical insecticides sprays required by the PBR cowpea (only two).

2. Genetic purity of the PBR cowpea seed in adopter farmers' fields.

The purity of the seed harvested from PBR cowpea adopters' fields, both seed buyers or seed savers, is adequate, near 97% overall agro-ecologies.

3. Assessment of yield and residues of chemical insecticides in grains.

The PBR cowpea fields, sprayed only twice, had the highest grain yield compared to the 2, 4 or 5 sprays of non-PBR cowpea fields in all the agroecological zones. At the same time, harvested PBR cowpea grains did not show traces of chemical insecticides while in non-PBR cowpea grains six common chemical insecticides were detected, sometimes at levels higher than those recommended by the European Union.

4. Overall comparison of adopters of PBR cowpea versus non-adopters.

We have identified high levels of implementation of the PBR cowpea recommended crop management practices by the adopter farmers. This results in better agronomical management, diligence, and outcomes by the PBR cowpea adopters compared to non-adopter farmers.

5. Assessment of the interest in PBR cowpea in Ghana and Burkina Faso.

In Ghana, relatively low levels of awareness about the PBR cowpea were found across farmers, although they were highly receptive to the PBR product and would very likely adopt it if it were available. Almost identical results were found in Burkina Faso.

9.2 Recommendations

This SRA have identified numerous significant results supporting positive economic, environmental, and health-related impacts of the PBR cowpea. We recommend, in first place, sharing this report across project-partners and stakeholders to disseminate the conclusions as soon as possible, with the aim of consolidating support for the commercialization at scale of the PBR variety. In second place, we recommended to prepare a communication plan to make sure the positive benefits of the PBR cowpea are expose to the communities in West Africa, with the objective of increasing the demand for the product and gaining supporters not only in Nigeria, but also in Ghana and Burkina Faso.

This SRA was designed to provide a quick snapshot of the PBR cowpea performance on a single cropping season, with the aim of having an early evaluation of this biotech product. For achieving a more robust and statistically significant conclusions we also recommend further funding to perform multiyear and multilocation studies, extending beyond biophysical to capture social and economic understanding of the technology and its performance.

10 References

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10.2 List of publications from the project

From all the different studies completed during this SRA (see the Appendixes) it is expected that several scientific and outreach publications will come out in the next 6 months:

- Publication regarding the entomological studies.
- Publication regarding the soil studies.
- Publication on the topic of chemical residues in seeds.
- Publication on the evaluation of the stewardship practices.

We need to notice that this SRA is a small activity restricted to one single cropping season and that has used a relatively low number of samples. The project team will meet to discuss how the results should be combined and if follow-up studies are needed to warrant the scientific soundness of the current results.

11 List of Appendixes (available at ACIAR)

11.1 List of PBR cowpea adopter farmers selected for the study.

11.2 Determination of the field-based biological consequences of insecticide sprays on insect population, cowpea grain, and yield in adopters of PBR and non-PBR cowpea in four agroecological zones of Nigeria.

11.3 Determining the Effect of PBR cowpea in reducing the Adverse Effects of Pesticides on Soil Biological Properties and Improving Soil Fertility

11.4 Socio-economic assessment of agronomic practices amongst adopters and non-adopters of Pod-Borer Resistant (PBR) cowpea farmers in some selected agroecological zones of Nigeria.

11.5 Trait purity report.

11.6 Survey report of farmers perceptions of Sampea 20-T in Ghana.

11.7 Survey report of farmers perceptions of Sampea 20-T in Burkina Faso.
