Internal Ex-Post Evaluation for Technical Cooperation Project (SATREPS[1])

conducted by Brazil Office: October, 2019

Country Name	Development of Genetic Engineering Technology of Crops with Stress Tolerance against Degradation of Global Environment
Federative Republic of Brazil	

I. Project Outline

Background	In year of 2006/07, Brazil was the second largest producer of soybean in the world next to the United States. The soybean production in Brazil was 58.4 million tons which accounted for approximately one-fourth of the total production in the world. However, in the country, frequent droughts caused by climate change happened and significantly damaged soybean cultivation. For example, the total production declined by 27.2 million tons for the period from 2003 to 2006. The economic loss amounted 1.8 billion dollars in 2003/04 and 2.32 billion dollars in 2004/05. Under the situation, the development of varieties of crops with tolerance against environmental stress including droughts, in particular for crops with large-scale cultivation in dry areas with relatively few rainfalls such as soybeans and maize, was an urgent issue for the country.		
Objectives of the Project	Through identification of genes with environmental stress tolerance and stress-responsive promoters, optimization of combinations between the identified promoters and useful genes, introduction of the optimized combinations into soybean, and evaluation of the stress tolerance of the genetically engineered soybean, the project aimed at developing genetic engineering technology of soybean with environmental stress tolerance, and thereby contributing to development of soybeans to be adapted to environmental stresses which contributes to the stabilization of the soybean production in Brazil. 1. Expected Overall Goal: Soybeans adapted to environmental stresses are developed, which contributes to the stabilization of the soybean production in Brazil. 2. Project Purpose: Genetic engineering technology of soybean with environmental stress tolerance is developed.		
Activities of the Project	1. Project Site: Brazilian Agricultural Research Corporation (EMBRAPA) Soybean (Londrina, Paraná) 2. Main Activities: (1) Identification of genes with environmental stress tolerance and stress-responsive promoters, (2) Optimization of combinations of the identified promoters and useful genes, (3) Introduction of the optimized combinations into soybean, (4) Evaluation of the stress tolerance of the genetically engineered soybean, etc. 3. Inputs (to carry out above activities) Japanese Side 1) Experts: 33 persons 2) Trainees Received: 13 persons 3) Equipment: vehicle, meteorological observation equipment, low temperature seed storage, etc. 4) Equipment for Japanese side: multilabel reader, artificial climate chamber, multifunctional shaking incubator, etc. 5) Local Expense: costs for project activities Brazilian Side 1. Staff Allocated: 35 persons 2. Equipment: automatic workstation, SNPs genetic analysis system, etc. 3. Land and facilities: an office space for Japanese experts and researchers, existing biotechnology buildings, etc. 4. Local expenses: scholarship for undergraduate, graduate and postdoctoral students, utility costs, communication costs, etc.		
Project Period	March 2010 – March 2015	Project Cost	(ex-ante) 360 million yen, (actual) 289 million yen
Implementing Agency	Brazilian Agricultural Research Corporation (EMBRAPA) Soybean		
Cooperation Agency in Japan	Japan International Research Center for Agricultural Sciences, The University of Tokyo, RIKEN		

II. Result of the Evaluation

1 Relevance

<Consistency with the Development Policy of Brazil at the Time of Ex-Ante Evaluation and Project Completion>

The project was consistent with Brazil's development policies of "Biosafety Law" (No.8974, January 1995) (1995-2005) promoting biotechnology including gene recombination at the time of ex-ante evaluation, and also, "Agricultural and Livestock Plan" (2012-2013) and "Multi-year Plan" (2012-2015) raising mitigation of effects by climate change as a major challenge in the agricultural sector at the time of project completion.

<Consistency with the Development Needs of Brazil at the Time of Ex-Ante Evaluation and Project Completion >

The project was consistent with Brazil's development needs of developing varieties of crops with large scale cultivation in dry areas with relatively few rainfalls, such as soybeans and maize, which can be tolerant against environmental stress including droughts in order to cope with damages by droughts brought by climate change.

<Consistency with Japan's ODA Policy at the Time of Ex-Ante Evaluation>

The project was consistent with Japan's ODA policy[2], focusing on the five priority areas including agriculture, based on the top-level agreement between Brazil and Japan at the time of the President Lula's visit in Japan in May, 2005.

[1] SATREPS: Science and Technology Research Partnership for Sustainable Development

[2] Ministry of Foreign Affairs "ODA Databook" (2007)

<Evaluation Result>
In light of the above, the relevance of the project is high.

2 Effectiveness/Impact

<Status of Achievement of the Project Purpose at the time of Project Completion>

The Project Purpose was achieved by the time of project completion. 12 useful genes related to environmental stress tolerance were identified (Indicator 1). Out of the identified 12 useful genes, 6 stress-responsive promoters were isolated and 17 combinations were optimized (Indicator 2). Among the combinations introduced in soybean, more than 3 transgenic lines were produced for 5 combinations (Indicator 3). As a result of experiments in greenhouse and field and crossbreed using the transgenic lines, 2 types of lines with environmental stress tolerance were selected (Indicator 4).

<Continuation Status of Project Effects at the time of Ex-post Evaluation>

The Project Effects have been continued since the project completion. As the 2 lines with environmental stress tolerance selected by the project have a characteristic of drought tolerance, they are considered useful nationwide in Brazil, and it has been aimed at adaptation of the genetically engineered soybean by the private sector through public and private sector collaboration. However, since there are differences in natural conditions in each area of vast land in Brazil, further research on the lines is indispensable. Meanwhile, the development cost is so large to be borne only by private companies that EMBRAPA Soybean started a new research on genetic engineering technology of lines with environmental stress tolerance of soybean under collaboration with private companies.

Also, owing to its usefulness, research related to the genetic engineering technology of lines with environmental stress tolerance of soybean and its application were initiated. For instance, EMBRAPA Genetic Resources and Biotechnology has endeavored to acquire further knowledge about genetic recombination. EMBRAPA Agroenergy has been applying the research outputs of the project to sugar cane. EMBRAPA Temperate Agriculture has conducted applied research for temperate fruits by using the research outputs of the project as countermeasures against droughts and floods. Besides, the research outputs of the project have been utilized for researches to apply to cotton, maize, and sugar cane.

The equipment procured by the project (meteorological observation equipment, low temperature seed storage, clean bench, guelph permeameter) has been continuously used by EMBRAPA Soybean based on its intended use of each equipment such as climate monitoring, seed preservation, plant management, and radioactivity management.

<Status of Achievement for Expected Overall Goal at the time of Ex-post Evaluation>

The Overall Goal has been partially achieved at the time of ex-post evaluation. At the time of survey for ex-post evaluation (June, 2019), it was confirmed that soybeans, which has a potential to adapt to environmental degradation, were developed by using the lines with environmental stress tolerance produced by the project and the developed outputs have been being established through field verification.

In addition, EMBRAPA Soybean has provided the technical guidance to private companies and delivered science and technology education to students of academic institutions as a part of efforts toward utilization of the research outcomes produced by the project. Specifically, from the period from 2015 to 2019, a course on biological and cytological identification technology of genetically engineered crops was provided to university students (undergraduate students: 3, postgraduate students: 6).

<Other Impacts at the time of Ex-post Evaluation>

No other positive or negative impact was observed at the time of ex-post evaluation.

<Evaluation Result>

Therefore, the effectiveness/impact of the project is high.

Achievement of Project Purpose and Overall Goal

<table>
<tr><th>Aim</th><th>Indicators</th><th>Results</th></tr>
<tr><td rowspan="2">(Project Purpose)
Genetic engineering technology of soybean with environmental stress tolerance is developed.</td><td>(Indicator 1)
At least 10 useful genes related to environmental stress tolerance are identified in plants such as soybean.</td><td>Status of the Achievement: achieved (continued)
(Project Completion)
• 12 useful genes related to environmental stress tolerance were identified by the researchers of JIRCAS, the University of Tokyo and RIKEN.
[Number and Name of the identified useful genes]
<table>
<tr><th>Organization</th><th>No. of the genes</th><th>Name of the genes</th></tr>
<tr><td>JIRCAS</td><td>7</td><td>AtDREB1A, AtDREB2A, AtAREB1, GmAREB1, GmAREB2, GmAREB3, GmAREB4</td></tr>
<tr><td>RIKEN</td><td>2</td><td>GmNCED3A, GmNCED3B</td></tr>
<tr><td>The University of Tokyo</td><td>3</td><td>GmDREB2A;2, GmHK1A;1, GmHK1B;1</td></tr>
</table>
(Ex-post Evaluation)
• Refer to the achievement of the indicator 4</td></tr>
<tr><td>(Indicator 2)
At least 5 stress-responsive promoters are identified, and combinations with useful genes are optimized.</td><td>Status of the Achievement: achieved (continued)
(Project Completion)
• 6 stress-responsive promoters were identified, and 17 combinations were optimized.
• Information about the isolated promoters
➢ 1 stress-responsive promoter (RD29) from Arabidopsis
➢ 5 stress-responsive promoters (Gm2, Gm3, Gm4, Gm5, Gm11) from soybean
[Information about the optimization]</td></tr>
</table>

		<table><tr><th>Organization</th><th>No. of the combinations</th><th>Combination of the optimization</th></tr><tr><td>JIRCAS</td><td>7</td><td>J1, J2, J3, J4, J5, J6, J7, J8, J9</td></tr><tr><td>RIKEN</td><td>2</td><td>R1, R2</td></tr><tr><td>The University of Tokyo</td><td>3</td><td>T1, T2, T3, T4, T5, T6, T7, T8, T9</td></tr></table> (Ex-post Evaluation) • Refer to the achievement of the indicator 4
	(Indicator 3) At least 5 combinations of useful genes and promoters are introduced in soybean, and at least three transgenic lines are produced for each combination.	Status of the Achievement: achieved (continued) (Project Completion) • 7 combinations (J1, J2, J3, J4, J5, J6, J7) between promoters and useful genes were introduced in soybean by particle gum method, and 3 combinations (J5, R1, R2) were introduced in soybean by Agrobacterium method. More than 3 transgenic lines were produced for 5 combinations (J1, J2, J5, J6, R1) out of them. (Ex-post Evaluation) • Refer to the achievement of the indicator 4
	(Indicator 4) At least 1 stress-tolerant line with environmental stress tolerance is selected.	Status of the Achievement: achieved (continued) (Project Completion) • As a result of experiments and crossbreeding in greenhouse and field and, 1a line with a characteristic of dry tolerance and 5a line with excellent characteristics of extreme drought tolerance and insect pest resistance were selected. (Ex-post Evaluation) • Being useful across Brazil, the selected lines with environmental stress tolerance (1a and 5a) have been continuously utilized in order to introduce the genetically engineered soybean by the private sector under collaboration between public and private sectors. • As there are differences in the natural conditions in each area of Brazil, EMBRAPA Soybean started a new joint research pertinent to genetic engineering of a line with environmental stress tolerance of soybean with private companies.
(Expected Overall Goal) Soybeans adapted to environmental stresses are developed, which contributes to the stabilization of the soybean production in Brazil.	(Indicator 1) Soybeans adapted to environmental stresses are developed before 2019.	(Ex-post Evaluation) partially achieved • In June 2019, soybeans, which has a potential to adapt to environmental stresses, were developed, and its developed output has been being established through field verification.

Source：Terminal Evaluation Report, Questionnaire and Interview with EMBRAPA Soybean

3 Efficiency

The project cost and period were within the plan (ratio against the plan: 80% and 100%, respectively). The outputs were produced as planned. Therefore, the efficiency of the project is high.

4 Sustainability

<Policy Aspect>

"Biosecurity Law" (No.11.105, March 2005), a successive law of "Biosafety Law" (No.8974, January 1995), promotes biotechnology including genetic recombination. The project has been endorsed by the policy, in that it aims at developing genetic engineering technology of crops with environmental stress tolerance against the global environment degradation.

<Institutional Aspect>

[Development of genetic engineering technology of soybean with environmental stress tolerance]

There have not been any major changes in the institutional arrangement for development of genetic engineering technology targeted by the project. EMBRAPA Soybean has carried out 1) research on genetic engineering technology of soybean with environmental stress tolerance, 2) research related to development of soybean by using genetic engineering technology of soybean with environmental stress tolerance, 3) research for commercialization of soybean with environmental stress tolerance. Even after the project completion, EMBRAPA Soybean has retained a relationship with the Japanese research institutes through academic conferences inviting the Japanese experts involving in the project, daily contacts via e-mail, and co-authoring of research papers.

[Utilization of the research outcomes]

EMBRAPA Soybean has introduced the research outputs produced by the project to private companies and delivered technical guidance and education on biological and cytological identification technology of genetically engineered crops to university students as a part of efforts toward utilization of the research outcomes of the project

[Maintenance of the equipment and facilities for the research]

The research equipment procured by the project has been placed in and maintained by EMBRAPA Soybean. According to them, as described above, the equipment has continuously been utilized and be in good conditions.

<Technical Aspect>

[Development of genetic engineering technology of soybean with environmental stress tolerance]

EMBRAPA Soybean has sustained the necessary knowledge and skills for development of genetic engineering technology of soybean with environmental stress tolerance. In this background, as mentioned above, there have been continuous efforts of EMBRAPA Soybean for obtaining new knowledge and improving the existing knowledge and skills through academic conferences inviting the Japanese expert involving in the project and so on. Also, EMBRAPA Soybean has provided reeducation to their researchers in collaboration with EMBRAPA Temperate Agriculture, University of Sao Paulo, and University of West Paulista though on an ad hoc basis.

[Utilization of the research outcomes]

EMBRAPA Soybean has sustained the necessary knowledge and skills for utilization of the research outcomes since they have contributed to society through the implementation of science and technology education and the provision of the technical guidance of the research outputs of the project to private companies. Specifically, EMBRAPA Soybean has provided a postgraduate course on molecule and gene in cooperation with Londrina State University and fostered 2 Ph.D holders. Furthermore, they have accepted 4 postdoctoral researchers (Sao Paulo Research Foundation: 2, Brazilian Science and Technology Committee: 2) and have made their efforts for utilization of the research outcomes related to the research outputs by the SATREPS project

[Maintenance of the equipment and facilities for the research]

EMBRAPA Soybean has sustained the necessary knowledge and skills for maintenance of the research equipment procured by the project. Staff of the unit has been strengthening their knowledge and skills for the maintenance of the research equipment by participating in internal trainings and related workshops.

<Financial Aspect>

Every year, the necessary budget for development of genetic engineering technology of soybean with environmental stress tolerance and utilization of the research outcomes as well as the maintenance of the research equipment and facilities installed by the project has been allocated by the federal government. According to EMBRAPA Soybean, being able to conduct their activities without any major problems, the budget has been sufficiently ensured.

Budget of EMBRAPA Soybean

(Unit: Real)

2015	2016	2017	2018	2019	2020 (Plan)
1,019,808	1,178,532	699,256	1,065,834	1,019,808	1,178,532

<Evaluation Result>

Therefore, the sustainability of the effects through the project is high.

5 Summary of the Evaluation

The project achieved the Project Purpose aiming at development of genetic engineering technology of soybean with environmental stress tolerance and partially achieved the Overall Goal aiming at developing soybeans adapted to environmental stresses.

Considering all of the above points, this project is evaluated to be highly satisfactory.

III. Recommendations & Lessons Learned

Recommendations for Implementing Agency:

- EMBRAPA Soybean has made efforts toward utilization of the research outcomes by conducting a multitude of activities regarding the project. However, as the information about specific activities related to each research has not been appropriately compiled and well-organized, one sometimes cannot readily refer to what kind of and in what way research outcomes of the project are linked to the current research activities and what achievements of the research activities have attained so far. In order to promote research activities even more, to ensure research funds and to cooperate with related agencies including private companies for utilization of the research outcomes in the future, it is recommended that researchers of EMBRAPA Soybean, who had been handed over the research outputs by the SATREPS project, regularly organize the data and information on the past activities (especially, seminars, workshops, agreement, and so on) and establish a system where they can smoothly disclose them to external agencies and carry out publicity activities.

Lessons Learned for JICA:

- The success of the project can be attributed to the following factors; 1) the experience of EMBRAPA as an implementing agency and an important cooperation partner of JICA in the agricultural sector since 1980s, 2) the established mutual trust between JICA and the implementing agency, 3) the smooth project formulation and implementation by EMBRAPA with deep understanding on the JICA's cooperation schemes. Moreover, the smooth project implementation, namely the implementation of the joint research by the Brazilian researchers and the Japanese researchers under the SATREPS project, was led by participation of the Japanese expert who had already established trust with EMBRAPA through the past technical cooperation projects under the partnership with EMBRAPA in addition to a great deal of trust of the Brazilian researchers to the knowledge of the Japanese researchers. Besides, since the research object of the SATREPS project met the unchanged needs in Brazil, the Brazil side has continued the research activities related to the project and made efforts toward utilization of the research outcomes. Thus, for smoothly implementing SATREPS projects and achieving the research outputs by the project, it is essential that a cooperation relationship or a trusting relationship between research institutes of developing countries' and Japanese sides to be involved in a joint research should be established before starting projects. Also, while efforts for utilization of the research outcomes require continuation of research activities, the research object should respond to high and unchangeable needs in the target countries in order to further promote the research activities. In addition, it is necessary to identify the needs of the partner countries and to plan a research reflecting the needs at the time of selection of the research plans for the SATREPS projects.

Crossbreed experiments using gene recombination at EMBRAPA Soybean

Field experiments at EMBRAPA Soybean