

Exploration of Leguminous Crops and Their Wild Relatives in Western Regions of Cambodia, 2014

Yu TAKAHASHI ^{1), 2)}, Seang LAY HENG ³⁾, Ty CHANNA ³⁾, Ouk MAKARA ³⁾
and Norihiko TOMOOKA ¹⁾

1) Genetic Resources Center, National Institute of Agrobiological Sciences, Kannondai 2-1-2, Tsukuba, Ibaraki 305-8602, Japan

2) Research Fellow of the Japan Society for the Promotion of Science

3) Cambodian Agriculture Research and Development Institute, National Road 3, Prateahlang, Dangkor, P.O Box 01, Phnom Penh, Cambodia

Corresponding author: N. TOMOOKA (e-mail: tomooka@affrc.go.jp)

Summary

We have conducted a field survey on legume genetic resources in western regions of Cambodia from 9th to 22nd of November, 2014. A total of 42 accessions were collected, including 1 accession of *Glycine max*, 15 accessions of *Vigna minima*, 5 accessions of *V. radiata*, 3 accessions of unidentified *Vigna* (*Vigna* sp.), 6 accessions of *V. umbellata*, 10 accessions of *V. unguiculata* and 2 accessions of *V. vexillata*. The seed samples were conserved in the gene bank of Cambodian Agriculture Research and Development Institute (CARDI), Cambodia, and the subsets were transferred to the Genebank of National Institute of Agrobiological Sciences (NIAS), Japan using sMTA. NIAS Genebank plan to multiply the seeds collected in this survey, and conserve them for research and breeding purposes.

KEY WORDS: Cambodia, Crop wild relatives, Legume, *Vigna*, *Glycine*

Introduction

Conservation of crop wild relatives can be conducted by two strategies, i.e., *in situ* and *ex situ* conservation. *In situ* conservation was defined as “conservation of ecosystems and natural habitats and the maintenance and recovery of viable populations of species in their natural surroundings and, in the case of domesticates or cultivated species, in the surroundings where they have developed their distinctive properties” (Maxted *et al.* 2015). Because this strategy does not target the conservation of specific genotypes, there are cases in which specific genotypes might become extinct in a habitat. In contrast, *ex situ* conservation was defined as “conservation of components of biological diversity outside their natural habitats” (Maxted *et al.* 2015). Currently, *ex situ* conservation plays an important role, as there is progressive destruction of habitat such as desertification or urbanization.

The National Institute of Agrobiological Sciences (NIAS) Genebank, Japan has been conducting

explorations for conserving genetic resources of crops and their wild relatives. In the present exploration, we have mainly focused on the collection of the wild plants belonging to the genus *Vigna*. The genus *Vigna* belongs to the legume family (Leguminosae), and includes several crops such as cowpea (*Vigna unguiculata* (L.) Walp.), mung bean (*Vigna radiata* (L.) Wilcz.), rice bean (*Vigna umbellata* (Thunb.) Ohwi & Ohashi) and azuki bean (*V. angularis* (Willd.) Ohwi & Ohashi). Their wild relatives inhabit diverse environments such as arid lands, coastlines or lime stone mountains (Tomooka *et al.* 2014). In addition, some of the wild relatives can cross with the *Vigna* crops. For example, *Vigna minima* (Roxb.) Ohwi & Ohashi can cross with rice bean and azuki bean, while *Vigna radiata* (L.) Wilcz. var. *sublobata* (Roxb.) Verdc. can cross with mung bean (Tomooka *et al.* 2002). Therefore, it is expected that these wild relatives could be used directly as breeding materials to develop stress tolerant crops (Tomooka *et al.* 2014).

This is a report of the fourth field survey on the collection of legume genetic resources in Cambodia under the Joint Research Agreement of Plant Genetic Resources between NIAS and the Cambodian Agricultural Research and Development Institute (CARDI). So far, we have conducted three field surveys in Cambodia; (1) southern regions (Kampong Speu and Kep provinces) and eastern regions (Kratie and Mondol Kiri provinces) in 2011 (Tomooka *et al.* 2012), (2) southwestern regions (Sihanoukville and Koh Kong provinces) and northern regions (Siem Reap, Anlong Veng, Tbaeng Meanchay and Kampong Thom provinces) in 2012 (Tomooka *et al.* 2013), and (3) northeastern regions (Stung Treng and Ratanakiri provinces) in 2013 (Takahashi *et al.* 2014). Therefore, in this survey, we targeted western regions of Cambodia where we did not visit in the former explorations.

Materials and Methods

A field survey was conducted in Cambodia from 9th to 22nd of November, 2014 (Table 1). We started from Phnom Penh, and visited Kampong Chhnang, Pursat, Battambang, Pailin, Banteay Meanchey and Oddar Meanchey Provinces by car, then returned to Phnom Penh (Fig. 1). When we found wild *Vigna* plants from the car or came across a habitat where wild *Vigna* plants seemed to grow, we stopped our car and searched the area. In addition, we sometimes conducted interviews to farmers about the cultivation of legume crops and/or the utilization of wild *Vigna* plants.

In general, a bulk seed sample was collected from a natural population in one site. However, if a population contained plants with different morphological traits, several seed samples were collected separately as different accessions according to their traits. Naturally growing *Vigna* plants were classified either as a wild form or an intermediate form between wild and domesticated based on their morphology. Wild form shows typical characteristics of wild *Vigna* plants, such as black mottled small seeds, shattering pods and twining slender stems. Intermediate form shows some of the characteristics of the domesticated form, generally having light-colored larger seeds, non-shattering pods and less-twining thicker stems. The domesticated form (crop landrace) was collected from a farmer's seeds stocks, from a farmer's cultivation field, or from a local market.

Identification of wild *Vigna* species was conducted based on a taxonomic key (Tomooka *et al.* 2002). As a passport data, collection locality, latitude, longitude, altitude and special characteristics of the plants and/or habitat recognized during collection were recorded. Latitude, longitude and altitude were measured by WGS84 world geodetic system using Garmin GPSmap 60CSx.

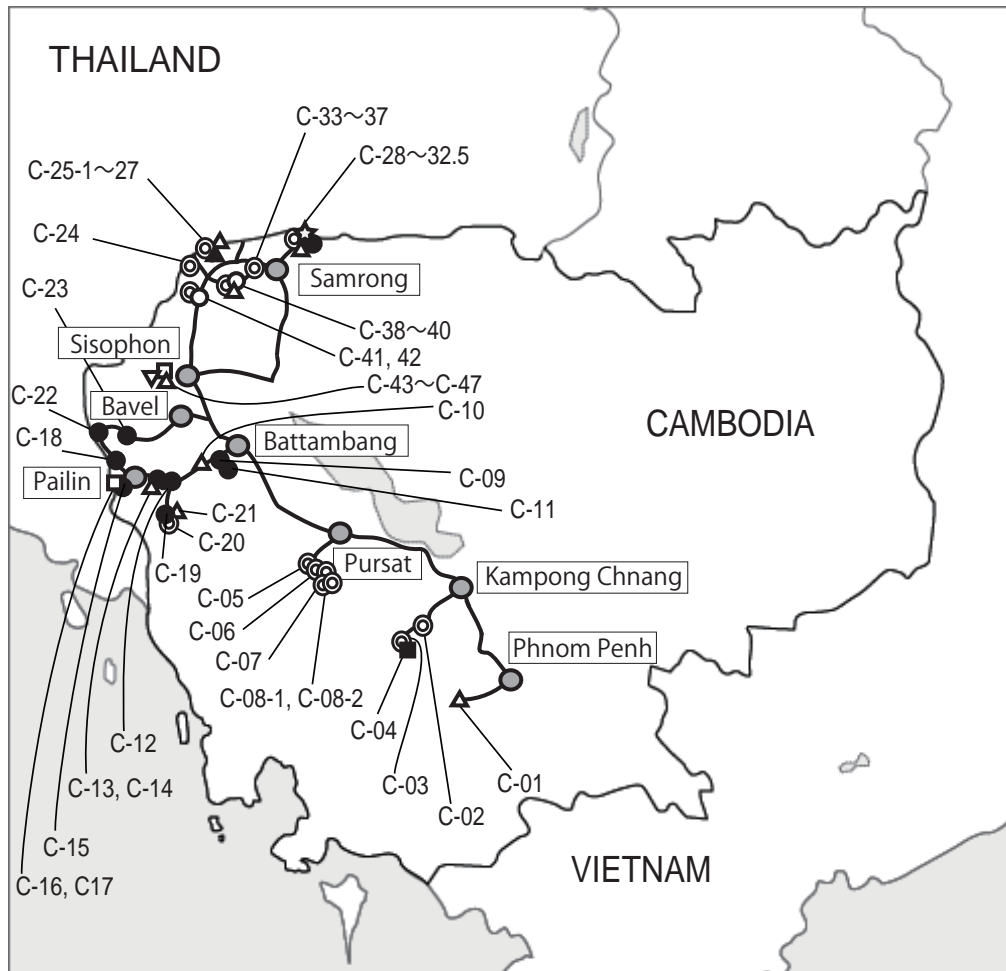


Fig. 1. A map showing the collection sites of the collected materials.

- ▼ *Glycine max* (soybean)
- ⊙ *Vigna minima*
- *Vigna radaita* (mungbean)
- *Vigna radaita* var. *sublobata*
- ★ *Vigna* sp.
- *Vigna umbellata* (rice bean)
- ▲ *Vigna unguiculata* (cowpea)
- ▲ *Vigna unguiculata* (yard long bean)
- *Vigna vexillata* (tuber cowpea)

Table 1. Itinerary of the field survey in western regions of Cambodia, 2014.

Date	Itinerary	Stay	
		City / Town	Province / Municipality
09-Nov	Narita - Bangkok - Phnom Penh	Phnom Penh	Phnom Penh
10-Nov	Phnom Penh (visit CARDI)	Phnom Penh	Phnom Penh
11-Nov	Phnom Penh - Kampong Chhnang	Kampong Chhnang	Kampong Chhnang
12-Nov	Kampong Chhnang - Pursat	Pursat	Pursat
13-Nov	Pursat - Battambang	Battambang	Battambang
14-Nov	Battambang - Pailin	Pailin	Pailin
15-Nov	Pailin - Battambang	Battambang	Battambang
16-Nov	Battambang - Sisophon	Sisophon	Banteay Meanchey
17-Nov	Sisophon - Samrong	Samrong	Oddar Meanchey
18-Nov	Samrong - Sisophon	Sisophon	Banteay Meanchey
19-Nov	Sisophon - Phnom Penh	Phnom Penh	Phnom Penh
20-Nov	Phnom Penh (visit CARDI)	Phnom Penh	Phnom Penh
21-Nov	Phnom Penh - Bangkok	On flight	-
22-Nov	Bangkok - Narita	-	-

Results and discussion

A total of 42 accessions consist of 6 species in 2 genera of leguminous plants were collected (Table 2, Table 3). These collections include 1 accession of *Glycine max* Merr., 15 accessions of *Vigna minima*, 5 accessions of *V. radiata*, 3 accessions of unidentified *Vigna* (*Vigna* sp.), 6 accessions of *V. umbellata*, 10 accessions of *V. unguiculata* and 2 accessions of *V. vexillata* (L.) A. Rich. The feature of each collection is described below.

***Glycine max* Merr.**

An accession of soybean was obtained from Sisophon market in Oddar Meanchey Province (Table 3). Soybean was called “Sandaek Sien”. It has whitish yellow seed coat with brown hilum. The vendor said the seeds were produced in Battambang Province.

***Vigna minima* (Roxb.) Ohwi & Ohashi**

It was reported in the previous exploration reports that *V. minima* plants in Cambodia have diverse morphological variations (Takahashi *et al.*, 2014). The present survey was conducted about 10 days earlier than last year’s survey. Probably because of the timing or the raining pattern of this year, even when we found many *V. minima* plants growing, mature pods could not be found in many collection sites. However, in those localities, it was possible to observe the growth condition of *V. minima* plants in their own habitat. The taxonomic key characteristics of this species are small stipule, sparse hairs on stem, short inflorescence with few buds and yellow flower with bracteole shorter than calyx (Fig. 2) (Tomooka *et al.* 2002).

V. minima plants with whitish light colored seeds were found in Kampong Chhnang and Pursat Provinces. In Kampong Chhnang Province, an accession ‘C-03’ with whitish colored seeds almost lacking black mottles grew in a shrub forest floor covered by fine white sand (Photo 1, 2). White soil layer and backside hills consisting of white rocks were observed only in this locality around the area (Photo 3). In Pursat Province, ‘C-08-1’ with slightly lighter gray seeds grew at the foot of hills that had white sands and boulders (Photo 4, 5). The fields in this area are recently opened from forests to cultivate cassava, and farmers who work in these cassava fields lived in temporary huts at the foot of the hills (Photo 6). Since *V. minima* with light colored seeds were found growing only on white soil area, we considered that this trait might have some advantages in natural selection such as contributing to feeding avoidance from animals. However, since seed coat color is sometimes associated with seed coat dormancy, further field studies and laboratory experiments using multiplied seeds will be required to clarify the reason why whitish seed coat color evolved.

In the northern areas of Oddar Meanchey Province facing to the north border to Thailand, *V. minima* plants with diverse leaflet morphology were found (Photo 7-13). ‘C-24’ with shallowly lobed terminal leaflet was found growing in a rice paddy field (Photo 7, 8). ‘C-25-1’ grew in an open forest and its lower leaflets were round (ovate). However, leaflets on upper stem nodes gradually became slender to linear within an individual (Photo 9). In a paddy field near ‘C-25-1’, a *V. minima* plant with white patched round leaflets was found (Photo 10). ‘C-33’ had slender linear leaflets (Photo 11), and a *V. minima* plant having five leaflets was found at the same site (Photo 12). In addition, a *V. minima* plant with lobed lateral leaflets was found at a site where ‘C-42’ was collected (Photo 13).

Some of the *V. minima* plants found in the northern areas of Oddar Meanchey Province seemed to have some morphological characteristics similar to those of *V. umbellata*. For example, although they were identified as *V. minima* based on the overall morphological traits, the lobed leaflets of ‘C-24’ (Photo 8),

Table 2. A summary of collected materials in Cambodia.

Species	Domesticated form	Naturally growing plants		Total
		Intermediate form	Wild form	
<i>Glycine max</i>	1	-	-	1
<i>Vigna minima</i>	-	-	15	15
<i>Vigna radiata</i>	4	-	1	5
<i>Vigna</i> sp.	-	3	-	3
<i>Vigna umbellata</i>	1	3	2	6
<i>Vigna unguiculata</i>	6	4	-	10
<i>Vigna vexillata</i>	-	-	2	2
Total	12	10	20	42

the long pods of ‘C-35’ (Photo 14), the vigorous growth (Photo 15) and the long inflorescence with many flower buds of ‘C-42’ (Photo 16) are the typical characteristics of *V. umbellata*. Although *V. minima* and *V. umbellata* have retained some level of cross compatibility (Tomooka *et al.* 2002), natural hybrid populations have not been reported. It would be worth studying the possibility that these accessions were derived from natural hybridization between *V. umbellata* and *V. minima*.

***Vigna umbellata* (Thunb.) Ohwi & Ohashi**

Wild form of *V. umbellata* is distributed from north India, Southeast Asia to south China (Tomooka *et al.* 2006). Domesticated form of *V. umbellata* has been cultivated in the regions where the wild form was found including Cambodia. The domesticated forms have diverse seed coat color such as red, yellow, green and black. The wild form can cross with domesticated form (Tomooka *et al.* 2006). The taxonomic key characteristics of this species are long stipule, dense fine long hairs on stem, long inflorescence with many flower buds and bright yellow flowers (Fig.2) (Tomooka *et al.* 2002).

We found a total of 6 naturally growing wild *V. umbellata* populations. Distribution of wild *V. umbellata* population found in the present survey was restricted to Battambang and Pailin Provinces. All the populations were in flowering and pod maturing stages and thus we could not collect mature seeds during the survey. As was experienced from the previous surveys in Cambodia, wild *V. umbellata* populations always showed later maturity time compared with other wild *Vigna* species. Although we could not collect mature seeds, we found wild *V. umbellata* populations ‘C-09’, ‘C-11’, ‘C-12’, ‘C-13’, ‘C-15’ and ‘C-19’ in Battambang and Pailin Provinces (Photos 17-20). ‘C-13’ was a vigorous large population growing at a wet swampy lowland habitat in Pailin Province (Photo 17). Since the habitat of ‘C-09’ (Photo 18) and ‘C-11’ were not far from the city of Battambang (Fig. 1), we asked a researcher of Agricultural Office in Battambang Province, who guided us during the exploration in Battambang Province, to collect seeds when the plants in those populations reached mature stage.

In Battambang Province, 3 populations of intermediate form between wild and domesticated *V. umbellata* were found. ‘C-18’ was a large population growing from the side ditch of a main road to a garden of a farmer’s house (Photo 20), and the plants had large vegetative organs and large yellow seeds. The farmers living near this site said, “we do not cultivate them at present and they grow naturally, but we use mature seeds for making Khao Lan (a glutinous rice cooked with beans in a bamboo shoot)” (Photo 21). Although the seed color of collected ‘C-18’ was yellow, a farmer said she bought a black seeded rice bean (*V. umbellata*) in Rattanakiri Province (northeast of Cambodia) in 1993 and brought it to cultivate here. There are vast cassava

fields recently opened in this area. Leaves of cassavas and also of road side trees have become yellow (Photo 22). This may be because of the Fe deficiency caused by the alkaline soil, since many lime stone rock hills are seen in this area. However, leaves of 'C-18' were still green and the plants were growing vigorously, suggesting that *V. umbellata* might be resistant to high pH soil. 'C-22' having large red seeds was collected nearby place to the site of 'C-18'. The plants were growing in a cassava field where plenty of strange grey rocks with white patches were scattered (Photo 23). These rocks might be derived from limestone rock hills. A farmer who owned this field said, "We use the seeds of *V. umbellata* in the same way as mung beans."

***Vigna* sp. (unidentified *Vigna* accessions)**

Three unidentified *Vigna* accessions were collected at a northernmost locality in Oddar Meanchey Province ('C-30', 'C-31', 'C-32', Fig. 1, Table 2). We found these plants in an edge of a farmers' paddy field and several plants of *V. minima* ('C-29') were also found growing nearby but could not collect mature seeds (Photo 24). At this site (Photo 25), unidentified *Vigna* plants were found under piled up harvested rice straw ('C-30', Photo 26), beside a cassava plant ('C-31', Photo 27) and in grassland at the edge of paddy field ('C-32', Photo 28) within a distance of 10 m. Since the plants were diverse in their morphology, they were collected separately using different accession numbers ('C-30' 'C-31' 'C-32'). Among the plants of unidentified *Vigna*, 'C-32' had smaller leaves and 'C-30' had seeds with black mottle, while 'C-31' had larger leaves and seeds without black mottle. However, in general, they possessed slender leaves, short pods and skewed seed hilum which were similar to *V. minima* and large leaves, dense fine long hairs on stem and yellowish seeds which were similar to the domesticated form of *V. umbellata* (Fig. 2). In addition, we obtained rice bean 'C-32.5' (*V. umbellata*) with red seed coat from an old lady who owns this land. She said "These rice bean seeds were cultivated and harvested from a field far away from here, but the original seeds were harvested here." Since unidentified *Vigna* accessions possessed characteristics of both *V. minima* and *V. umbellata*, we considered them as hybrid derivatives between both species. In addition, since their traits seemed to be still segregating, it is considered that these plants might be in a young generation after hybridization and could be a hybrid swarm. We are planning to confirm the hybrid nature of these accessions using SSR markers, DNA sequences of chloroplasts and nuclear genomes.

In this survey, *V. minima* was found on arid sandy soil habitat (Kampong Chnang, Pursat, mountain area of Battambang and Oddar Meanchey Provinces), and wild *V. umbellata* was found only on lowland wet habitat (plain area of Battambang and Pailin Provinces). Difference of the habitat between *V. minima* and *V. umbellata* was also observed in the previous explorations in Cambodia (Tomooka *et al.*, 2012, 2013, Takahashi *et al.*, 2014). Although these two species have different habitat, some of the collected *V. minima* plants showed some morphological characteristics similar to those of *V. umbellata* as mentioned in the section of *V. minima*. Therefore, possibility of some gene flow between the two species should be investigated.

***Vigna radiata* (L.) Wilczek**

One accession of wild form of *V. radiata* was collected in the present exploration (Table 2). The wild form of *V. radiata* is distributed in Africa, Asia and Oceania, and the domesticated form (mung bean) is cultivated in the same regions (Tateishi 1996). This species has ovate stipule, long peduncle and pale greenish yellow flower (Tomooka *et al.* 2002).

In Kampong Chnang Province, a wild form of *V. radiata* 'C-04' was found crawling on the ground (Photo 29) and also climbed to grass plants (Photo 30). Although the wild form of *V. radiata* usually inhabits

relatively arid land, habitat of 'C-04' was wet grassland beside rice paddy field (Photo 31).

In Pailin Province, we collected 2 accessions of domesticated form, mung bean 'C-16' and 'C-17' cultivated in the same field. The farmers who cultivated these mung bean accessions were eating immature mung bean seeds in boiled green pods when we visited (Photo 32). The mature pod colors were black or brown (Photo 33) and the flowers were pale green or pale yellow (Photo 34, 35). However, pod and flower colors did not appear to be associated.

***Vigna vexillata* (L.) A. Rich.**

Wild form of *V. vexillata* is distributed throughout Africa, Asia, Oceania and the Central and South Americas (Damayanti *et al.* 2010). Domesticated form of *V. vexillata* was recently found in Bali and proposed to call "tuber cowpea" (Karuniawan *et al.* 2006). This species belongs to the subgenus *Plectrotropis* and has pink, purple or yellowish flowers, keel curves to the left with a pocket, and hairy long pods (Verdcourt 1970, Maxted 2004).

Two accessions ('C-39' and 'C-41') of wild *V. vexillata* were collected in northern regions of Oddar Meanchey Province (Fig. 1, Photo 36 - 41). Both accessions were growing sympatrically with *V. minima* plants in rice paddy fields. Although collection sites of these 2 accessions were not far, 'C-39' had brown seed coat and 'C-41' had black seed coat color (see Seed photos). 'C-39' had slender lanceolate leaflets (Photo 36) and growing together with *V. minima* accession 'C-38' having very similar slender lanceolate leaflets (Photo 37, 38). 'C-41' had slightly larger lanceolate leaflets and produced numerous long mature pods (Photo 40, 41).

***Vigna unguiculata* (L.) Walp.**

Wild form of *V. unguiculata* is distributed throughout Africa, and the domesticated form is cultivated from temperate to tropical zones of Africa, Asia and the Americas (Maxted *et al.* 2004). This species belongs to the subgenus *Vigna* and has symmetrical flowers colored white, greenish, yellow or lilac-purple and keel without pocket (Verdcourt 1970, Maxted 2004). Verdcourt (1970) classified the domesticated form into 3 subspecies, but Maréchal *et al.* (1978) proposed that they should be classified as cultivar-group (cv-gr.). Among them, cv-gr. *Unguiculata* (cowpea) and cv-gr. *Sesquipedalis* (yard long bean) are found cultivated in Asia. Ten accessions of *V. unguiculata* were collected in the present exploration (Table 2).

In northern areas of Oddar Meanchey Province, we found that vegetable crops were often cultivated on mounds in rice paddy fields. At one site, yard long bean 'C-26' and cowpea 'C-27' were cultivated with eggplants and chili peppers on a mound (Photo 42, 43, 44). At a site about 40 km east from the previous site, black seeded cowpea 'C-28' was cultivated amongst various shrubs on a mound. In addition, chili peppers were cultivated on a mound where *V. vexillata* plants 'C-41' and *V. minima* plants 'C-42' grew (Photo 45). This type of cultivation practice using mounds for vegetables production seems to be common in this area. These mounds should be surveyed in case vegetable crops are the target genetic resources.

Collected materials

We have explored and collected legume genetic resources for four years in Cambodia in 2011, 2012, 2013 and 2014 (this survey), and 22, 38, 74 and 42 accessions were collected respectively resulting in a total of 176 accessions. These 176 collections have been conserved in the CARDI genebank and the subsets were transferred to the NIAS genebank using a standard material transfer agreement. Most of the accessions

collected in the past three years have already been grown, evaluated and the multiplied seeds were conserved in the NIAS genebank for distribution. We plan to grow, evaluate and multiply the seeds of accessions collected in 2014 (this survey) in 2015. The collected materials are expected to be used in the breeding program.

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カンボジアにおけるマメ科遺伝資源多様性の保全 2014 年

高橋 有^{1),2)}, Seang LAY HENG³⁾, Ty CHANNA³⁾, Ouk MAKARA³⁾, 友岡 憲彦¹⁾

- 1) 農業生物資源研究所・遺伝資源センター
- 2) 日本学術振興会・特別研究員 PD
- 3) カンボジア農業研究機構

摘要

本稿は農業生物資源研究所とカンボジア農業研究機構の間で締結された共同研究協定に基づくカンボジアにおける第4回目のマメ科植物の調査報告である。調査は2014年11月9日から22日に行われた。その結果、ホソバツルアズキ (*Vigna minima* (Roxb.) Ohwi & Ohashi), ツルアズキ (*Vigna umbellata* (Thunb.) Ohwi & Ohashi), リョクトウ (*Vigna radiata* (L.) Wilczek), アカササゲ (*Vigna vexillata* (L.) A. Rich.), ササゲ (*Vigna unguiculata* (L.) Walp.), *Vigna* 属種未同定植物 (*Vigna* sp.), ダイズ (*Glycine max* (L.) Merr.) を含む計42点の遺伝資源が収集された。これらはカンボジア農業研究機構において保存され、その一部が標準材料移転契約を用いて農業生物資源研究所に移転された。今後我々はこれらの種子増殖と特性評価を行う計画である。

Table 3. A passport data of collected materials 収集品のパスポートデータ

JP No.	Coll. No.	Map ID	Coll. Date	Scientific name	Status	Coll. Site	Latitude	Longitude	Altitude (m)	Soil	Type of seed sample	Remarks
252340	2014C-01	C-01	2014/11/10	<i>Vigna unguiculata</i>	intermediate	CARDI campus, ca. 15 km SW of Phnom Penh	N11-28-37.59	E104-48-22.61	16	organic soil	bulk	growing in CARDI campus, soft pod with weak-shattering habit
-	2014C-02	C-02	2014/11/11	<i>Vigna minima</i>	wild	ca. 50 km SW of Kampong Chhnang	N11-55-15.3	E104-19-32.6	110	sand	no	growing in an arid sandy habitat, a few plants found, only young seedlings
252341	2014C-03	C-03	2014/11/11	<i>Vigna minima</i>	wild	ca. 55 km SW of Kampong Chhnang	N11-54-31.9	E104-19-25.0	130	fine white sand	bulk	growing in an open <i>Dipterocarpus</i> forest on fine white sand, only two mature pods and one immature pod found, whitish seeds
252342	2014C-04	C-04	2014/11/11	<i>Vigna radiata</i> var. <i>sublobata</i>	wild	ca. 55 km SW of Kampong Chhnang	N1-53-51.3	E104-19-12.4	120	fine gray sand	bulk	growing in a grassland beside rice paddy, reddish vein in the leaves
252343	2014C-05	C-05	2014/11/12	<i>Vigna minima</i>	wild	Seborai, Korowan Dist., ca. 40 km SSW of Pursat	N12-15-03.3	E103-50-01.3	65	clay	bulk	growing beside a road next to a bamboo grove and in an open space where <i>Mimosa</i> plants grew, mature small seeds with a prominent hilum found in the open space only.
-	2014C-06	C-06	2014/11/12	<i>Vigna minima</i>	wild	ca. 50k m S of Pursat	N12-11-58.7	E103-52-56.3	70	fine sand	no	growing in an open <i>Dipterocarpus</i> forest beside road, crawling along the forest floor, no seeds could be collected
-	2014C-07	C-07	2014/11/12	<i>Vigna minima</i>	wild	ca. 50km S of Pursat	N12-10-58.8	E103-54-45.7	80	fine sand	no	growing in an open <i>Dipterocarpus</i> forest, no seeds could be collected
252344	2014C-08-1	C-08-1	2014/11/12	<i>Vigna minima</i>	wild	ca. 50 km S of Pursat	N12-11-51.6	E103-54-45.9	120	white sand	bulk	growing at the base of a hill, crawling type, light gray seeds
252345	2014C-08-2	C-08-2	2014/11/12	<i>Vigna minima</i>	wild	ca. 50 km S of Pursat	N12-11-51.6	E103-54-45.9	120	white sand	bulk	growing at the base of a hill, climbing type, light gray seeds
252346	2014C-09	C-09	2014/11/13	<i>Vigna umbellata</i>	wild	Battanban city	N13-04-38.9	E103-11-11.3	15	gravel	bulk	growing in a grassland beside a road near the Battanban Agricultural Office, no mature seeds found, so a researcher harvested them after the survey
252347	2014C-10	C-10	2014/11/13	<i>Vigna unguiculata</i>	domesticated	farmer's house beside Kamping Puoy Reservoir ca. 25 km W of Battanban	N13-03-03.9	E102-58-33.0	20	no information available	bulk	local name: "Sandai Koua", mature pods harvested and hung above a fire place in a farmer's house, black seeds, its young pods are eaten with "Nam Prick" and the mature seeds are cooked to make "Khao Lan" (a steamed sticky rice cooked in a bamboo shoot with beans).
252348	2014C-11	C-11	2014/11/13	<i>Vigna umbellata</i>	wild	Ksai Poui, Wat Kho commune, Ban Non Dist., ca. 5 km S of Battanban	N13-02-44.8	E103-11-31.3	20	gravel	bulk	growing between a road and a rice paddy near the Battanban Agricultural Office, no mature seeds found, so a researcher harvested them after the survey
-	2014C-12	C-12	2014/11/14	<i>Vigna umbellata</i>	wild	Battanban	N12-50-28.6	E102-44-59.1	106	organic soil	no	growing between grassland and ditch beside a road, young leaves eaten as "Nam Prick" or "Som Tam", flowers visited by carpenter bees, no mature seeds
-	2014C-13	C-13	2014/11/14	<i>Vigna umbellata</i>	wild	ca. 5 km SE of Pailin	N12-49-16.5	E102-38-26.4	220	organic soil	no	growing between a road and a cassava field, large population around lowland wet place, many pods but no mature seeds
252349	2014C-14	C-14	2014/11/14	<i>Vigna unguiculata</i>	intermediate	ca. 5 km SE of Pailin	N12-49-16.5	E102-38-26.4	220	gravel	bulk	growing beside a road, small black seeds, probably escaped from cultivation
-	2014C-15	C-15	2014/11/14	<i>Vigna umbellata</i>	wild	Bo Yakha, Pailin	N12-52-52.5	E102-32-41.2	190	organic soil	no	growing between a red gravel road and a cassava field, no seeds could be collected
252350	2014C-16	C-16	2014/11/14	<i>Vigna radiata</i>	domesticated	Mrs. Chilap, ca. 10 km NW of Pailin	N12-53-28.8	E102-31-25.2	165	reddish clay	bulk	local name: "Sandai Kiu", a small mungbean field among cassava fields beside a farmer's house, cultivated since 1996, black pods, young boiled pods tasted good, they sell the mature seeds.
252351	2014C-17	C-17	2014/11/14	<i>Vigna radiata</i>	domesticated	Mrs. Chilap, ca. 10 km NW of Pailin	N12-53-28.8	E102-31-25.2	165	reddish clay	bulk	local name: "Sandai Kiu", a small mungbean field among cassava fields beside a farmer's house, cultivated since 1996, black pods, young boiled pods tasted good, they sell the mature seeds.

Table 3 (Continued).

JP No.	Coll. No.	Map ID	Coll. Date	Scientific name	Status	Coll. Site	Latitude	Longitude	Altitude (m)	Soil	Type of seed sample	Remarks
252352	2014C-18	C-18	2014/11/14	<i>Vigna umbellata</i>	intermediate	Slow LanThong, ca. 15 km N of Pailin, Battanbang	N13-01-19.8	E102-33-29.4	100	organic soil	bulk	growing along a road, bordering a garden of a farmer's house, intermediate type, yellow seeds, only one mature pod and two near mature pods collected, a farmer said, "I bought some black rice beans from Rattanakiri Province in 1993 and they were easy to grow", she uses mature seeds for making "Khao Lan" (a steamed sticky rice cooked in a bamboo shoot with beans), cassava and trees around this area showed yellow leaves but leaves of C18 were healthy green.
-	2014C-19	C-19	2014/11/15	<i>Vigna umbellata</i>	wild	Kas Sadai, Som Load, Battanbang	N12-40-27.3	E102-43-44.4	150	gravel	no	growing beside a road near a stream, no mature seeds collected
252353	2014C-20	C-20	2014/11/15	<i>Vigna minima</i>	wild	ca. 15 km S of Pailin, Battanbang	N12-39-40.8	E102-42-34.7	170	organic soil	bulk	growing beside a road, black long pods, gray seeds, leaves damaged by insects
252354	2014C-21	C-21	2014/11/15	<i>Vigna unguiculata</i>	intermediate	ca. 15 km SE of Pailin, Battanbang	N12-42-18.9	E102-44-47.0	130	organic soil	bulk	climbing on a fence between a road and a mango field, black seeds, probably escaped from cultivation
252355	2014C-22	C-22	2014/11/15	<i>Vigna umbellata</i>	intermediate	Otasok, Phom Pruk Dist., ca. 40 km N of Pailin, Battanbang	N13-13-56.0	E102-26-19.2	160	organic soil	bulk	growing between a road and a cassava field with plenty of grey rocks with white patches, a farmer said, "we use them in the same way as mung beans." two near mature pods collected, large red seeds, probably escaped from cultivation
252356	2014C-23	C-23	2014/11/15	<i>Vigna umbellata</i>	intermediate	Jamrong, Kom Rien Dist., ca. 30 km N of Pailin, Battanbang	N13-07-54.2	E102-38	70	organic soil	bulk	growing in a grassland beside a road, slender leaves, whitish pods, red seeds, probably escaped from cultivation
252357	2014C-24	C-24	2014/11/16	<i>Vigna minima</i>	wild	near Thai border, ca. 70 km NNE of Sisophon, Amphur commune, Oddar Meanchey	N14-16-40.1	E103-07-42.3	105	pale brown fine sand	bulk	growing in a rice paddy with trees, small flower, shallowly lobed terminal leaflet
252358	2014C-25-1	C-25-1	2014/11/16	<i>Vigna minima</i>	wild	ca. 70 km NE of Sisophon, in a dry rice field, Oddar Meanchey	N14-13-09.3	E103-17-27.4	50	fine gray sand	bulk	growing in a rice producing forest, lower leaves were ovate, leaves on the upper nodes progressively become lanceolate.
252359	2014C-25-2	C-25-2	2014/11/16	<i>Vigna minima</i>	wild	ca. 70 km NE of Sisophon, in a dry rice field, Oddar Meanchey	N14-13-09.3	E103-17-27.4	50	fine gray sand	bulk	growing in a rice producing forest, all leaves were ovate
252360	2014C-26	C-26	2014/11/16	<i>Vigna unguiculata</i> cv-gr. <i>Sesquipedalis</i>	domesticated	cultivated on a mound, ca. 70 km NE of Sisophon, in a dry rice field, Oddar Meanchey	N14-13-09.3	E103-17-27.4	50	organic soil	bulk	cultivated in a rice producing forest, cowpeas, yard long beans and other crops were cultivated on a small mound in a paddy field
252361	2014C-27	C-27	2014/11/16	<i>Vigna unguiculata</i>	domesticated	cultivated on a mound, ca. 70 km NE of Sisophon, in a dry rice field, Oddar Meanchey	N14-13-09.3	E103-17-27.4	50	organic soil	bulk	cultivated in a rice producing forest, cowpeas, yard long beans and other crops were cultivated on a small mound in a paddy field
252362	2014C-28	C-28	2014/11/17	<i>Vigna unguiculata</i>	domesticated	Kiriwon, Somrao Dist., ca. 10km NNE of Samrong, Oddar Meanchey	N14-18-35.3	E103-37-48.9	70	pale gray fine sand	bulk	cultivated beside a rice paddy with trees
-	2014C-29	C-29	2014/11/17	<i>Vigna minima</i>	wild	Kiriwon, Somrao Dist., ca. 10km NNE of Samrong, Oddar Meanchey	N14-18-35.3	E103-37-48.9	70	pale gray fine sand	no	growing in a rice paddy with trees, few plants and only immature pods, no seeds could be collected
252363	2014C-30	C-30	2014/11/17	<i>Vigna</i> sp.	intermediate	Kiriwon, Somrao Dist., ca. 10 km NNE of Samrong, Oddar Meanchey	N14-18-35.3	E103-37-48.9	70	pale gray fine sand	bulk	found growing under some harvested rice straw beside a farmer's house, larger leaves, immature pods collected, black mottled pale yellow seeds

Table 3 (Continued).

JP No.	Coll. No.	Map ID	Coll. Date	Scientific name	Status	Coll. Site	Latitude	Longitude	Altitude (m)	Soil	Type of seed sample	Remarks
252364	2014C-31	C-31	2014/11/17	<i>Vigna</i> sp.	intermediate	Kiriwon, Somrao Dist., ca. 10 km NNE of Samrong, Oddar Meanchey	N14-18-35.3	E103-37-48.9	70	pale gray fine sand	bulk	found growing beside a cassava plant near a farmer's house, larger leaves, immature pods collected, pale yellow seeds
252365	2014C-32	C-32	2014/11/17	<i>Vigna</i> sp.	intermediate	Kiriwon, Somrao Dist., ca. 10 km NNE of Samrong, Oddar Meanchey	N14-18-35.3	E103-37-48.9	70	pale gray fine sand	bulk	found growing in a grassland, immature pods collected, smaller leaves
252366	2014C-32.5	C-32.5	2014/11/17	<i>Vigna umbellata</i>	domesticated	Kiriwon, Somrao Dist., ca. 10 km NNE of Samrong, Oddar Meanchey	N14-18-35.3	E103-37-48.9	70	no information available	bulk	an old lady gave us seeds stored in her house and said, "these seeds were harvested in a far away field, but the original seeds were harvested in this garden", red seeds
252367	2014C-33	C-33	2014/11/18	<i>Vigna minima</i>	wild	ca. 15 km W of Samrong, Oddar Meanchey	N14-10-38.1	E103-26-19.9	46	gray fine sand	bulk	growing in a rice paddy, slender leaves and stems, immature pods collected
252368	2014C-34	C-34	2014/11/18	<i>Vigna minima</i>	wild	ca. 15 km W of Samrong, Oddar Meanchey	N14-10-38.3	E103-26-17.56	46	gray fine sand	bulk	growing in a rice paddy, broad leaves, vigorous growth, immature pods collected
252369	2014C-35	C-35	2014/11/18	<i>Vigna minima</i>	wild	ca. 15 km W of Samrong, Oddar Meanchey	N14-10-39.1	E103-26-16.4	46	gray fine sand	bulk	growing in a rice paddy, vigorous growth, long pods
252370	2014C-36	C-36	2014/11/18	<i>Vigna minima</i>	wild	ca. 15 km W of Samrong, Oddar Meanchey	N14-10-40.6	E103-26-15.8	46	gray fine sand	bulk	growing in a rice paddy, twining up to a tree, slender leaves, early-ripening, short thick pods
252371	2014C-37	C-37	2014/11/18	<i>Vigna minima</i>	wild	ca. 15 km W of Samrong, Oddar Meanchey	N14-10-39.1	E103-26-16.4	46	gray fine sand	bulk	growing in a rice paddy, near C-36, crawling type, slender leaves, early-maturing, short thick pod
252372	2014C-38	C-38	2014/11/18	<i>Vigna minima</i>	wild	Ohrundual, Kyokruna commune, ca. 30 km W of Samrong, Oddar Meanchey	N14-09-11.2	E103-21-06.0	35	gray fine sand	bulk	growing in a rice paddy with trees, slender leaves, large population, mature pods collected, a farmer said they eat mature seeds boiled and make "Khao Lan" (glutinous rice cooked in a bamboo shoot with beans). Young pods are boiled and eaten like vegetable soybean (eda mame).
252373	2014C-39	C-39	2014/11/18	<i>Vigna vexillata</i>	wild	Ohrundual, Kyokruna commune, ca. 30 km W of Samrong, Oddar Meanchey	N14-09-14.8	E103-21-03.3	40	gray fine sand	bulk	growing in a rice paddy with trees, purple flowers, slender leaves, together with <i>Vigna minima</i>
252374	2014C-40	C-40	2014/11/18	<i>Vigna unguiculata</i>	intermediate	Ohrundual, Kyokruna commune, ca. 30 km W of Samrong, Oddar Meanchey	N14-09-14.8	E103-21-03.3	40	gray fine sand	bulk	growing in a rice paddy with trees, probably escaped from cultivation
252375	2014C-41	C-41	2014/11/18	<i>Vigna vexillata</i>	wild	ca. 45km W of Samrong, Oddar Meanchey	N14-07-31.7	E103-11-04.3	50	reddish fine sand	bulk	growing on a small mound in a rice paddy with trees, slender leaves
252376	2014C-42	C-42	2014/11/18	<i>Vigna minima</i>	wild	ca. 45 km W of Samrong, Oddar Meanchey	N14-07-32.2	E103-11-03.8	50	reddish fine sand	bulk	growing on a small mound in a rice paddy with trees, slender leaves, vigorous growth
252377	2014C-43	C-43	2014/11/19	<i>Vigna radiata</i>	domesticated	from Sisophon market, Oddar Meanchey	N13-36-08.3	E102-58-08.0	24	no information available	bulk	local name "Sandaek Bai", latitude and longitude data indicate a locality of Sisophon market.
252378	2014C-44	C-44	2014/11/19	<i>Glycine max</i>	domesticated	from Sisophon market, Oddar Meanchey	N13-36-08.3	E102-58-08.0	24	no information available	bulk	local name "Sandaek Sien", produced in Battambang Province, latitude and longitude data indicate a locality of Sisophon market.
252379	2014C-45	C-45	2014/11/19	<i>Vigna unguiculata</i>	domesticated	from Sisophon market, Oddar Meanchey	N13-36-08.3	E102-58-08.0	24	no information available	bulk	white seeded cowpea, produced in Kampong Chhnang Province, latitude and longitude data indicate a locality of Sisophon market.
252380	2014C-46	C-46	2014/11/19	<i>Vigna radiata</i>	domesticated	from Sisophon market, Oddar Meanchey	N13-36-08.3	E102-58-08.0	24	no information available	bulk	yellow seeded mungbean, produced in Battambang Province, latitude and longitude data indicate a locality of Sisophon market.
252381	2014C-47	C-47	2014/11/19	<i>Vigna unguiculata</i>	domesticated	from Sisophon market, Oddar Meanchey	N13-36-08.3	E102-58-08.0	24	no information available	bulk	black seeded cowpea, latitude and longitude data indicate a locality of Sisophon market.



Photo 1. Habitat of *V. minima* (C-03) in Kampong Chnang



Photo 2. *V. minima* (C-03) in Kampong Chnang



Photo 3. White sand habitat of *V. minima* (C-03) in Kampong Chnang



Photo 4. Habitat of *V. minima* (C-08-1) in Pursat



Photo 5. *V. minima* (C-08-2) in Pursat



Photo 6. Habitat of *V. minima* (C-08-1 & 2) in Pursat



Photo 7. Habitat of *V. minima* (C-24) in Oddar Meanchey



Photo 8. Leaflets of *V. minima* (C-24) in Oddar Meanchey



Photo 9. Variation of leaflet shape within an individual plant of *V. minima* (C-25) in Oddar



Photo 10. Ovate leaflets of *V. minima* (near C-25) in Oddar Meanchey



Photo 11. Linear leaflets of *V. minima* (C-33) in Oddar Meanchey



Photo 12. Penta-foliate leaflets of *V. minima* (near C-33) in Oddar Meanchey



Photo 13. Lobed lateral leaflets of *V. minima* (C-42) in Oddar Meanchey



Photo 14. Pods of *V. minima* (C-35) in Oddar Meanchey



Photo 15. Vigorous growth *V. minima* (near C-35) in Oddar Meanchey(Ikema)



Photo 16. Inflorescence of *V. minima* (C-42) in Oddar Meanchey



Photo 17. Swampy habitat of *V. umbellata* (C-13) in Pailin



Photo 18. Roadside habitat of *V. umbellata* (C-09) in Battambang



Photo 19. Roadside habitat of *V. umbellata* (C-19) in Battambang



Photo 20. Habitat (road side ditch) of *V. umbellata* (C-18) in Battambang



Photo 21. Interview with farmers at rice bean (C-18) collection site in Battambang



Photo 22. Cassava with probable Fe deficiency symptom around *V. umbellata* (C-18) collection site in Battambang



Photo 23. *V. umbellata* (C-22) climbing on a rock in Battambang



Photo 24. *V. minima* (C-29) growing near *Vigna* sp. (C-30~32) in Oddar Meanchey



Photo 25. Habitat of *Vigna* sp. (C-30, 31, 32) in Oddar Meanchey



Photo 26. *Vigna* sp. (C-30) in Oddar Meanchey



Photo 27. *Vigna* sp. (C-31) in Oddar Meanchey



Photo 28. *Vigna* sp. (C-32) in Oddar Meanchey



Photo 29. *V. radiata* (C-04) in Kampong Chhnang



Photo 30. Pods of *V. radiata* (C-04) in Kampong Chhnang



Photo 31. Habitat (beside paddy) of *V. radiata* (C-04) in Kampong Chhnang



Photo 32. Boiled young mung bean pods (C-16, 17) for eating immature seeds



Photo 33. Brown and black pod mung bean (C-16, 17) grown sympatrically in Pailin



Photo 34. Yellowish flower of mung bean at (C-16, 17) field in Pailin



Photo 35. Greenish flower of mung bean at (C-16, 17) field in Pailin



Photo 36. Narrow lanceolate leaflets of *V. vexillata* (C-39) in Oddar Meanchey



Photo 37. *V. minima* (C-38) and *V. vexillata* (C-39) growing sympatrically in Oddar Meanchey

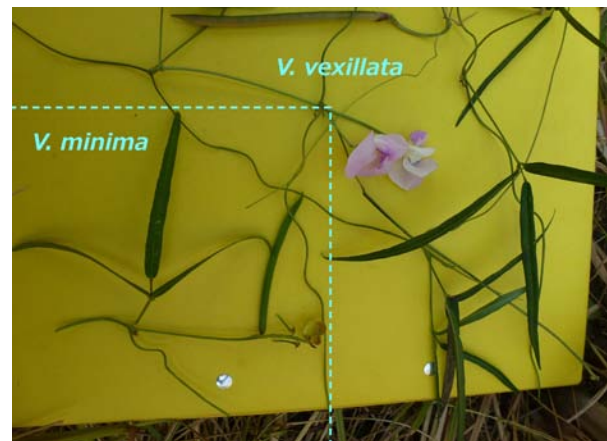


Photo 38. Very similar leaflets morphology of *V. minima* (C-38) and *V. vexillata* (C-39) in Oddar



Photo 39. Habitat (a mound in paddy field area) of *V. vexillata* (C-41) in Oddar Meanchey



Photo 40. Leaflets of *V. vexillata* (C-41) in Oddar Meanchey



Photo 41. Long slender pods of *V. vexillata* (C-41) in Oddar Meanchey



Photo 42. Pods of cowpea (C-27) in Oddar Meanchey



Photo 43. Pods of yard long bean (C-26) in Oddar Meanchey



Photo 44. Crops cultivated mound in paddy field area where C-26 and C27 were collected in Oddar Meanchey



Photo 45. Chili peppers cultivated on crops mound (with C-41) in Oddar Meanchey



C-01, JP252340,
Vigna unguiculata



C-03, JP252341,
Vigna minima



C-04, JP252342,
Vigna radiata var. *sublobata*



C-05, JP252343,
Vigna minima



C-08-1, JP252344,
Vigna minima



C-08-2, JP252345,
Vigna minima



C-09, JP252346,
Vigna umbellata



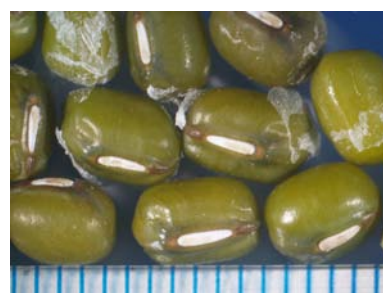
C-10, JP252347,
Vigna unguiculata



C-11, JP252348,
Vigna umbellata



C-14, JP252349,
Vigna unguiculata



C-16, JP252350,
Vigna radiata



C-17, JP252351,
Vigna radiata



C-18, JP252352,
Vigna umbellata



C-20, JP252353,
Vigna minima



C-21, JP252354,
Vigna unguiculata



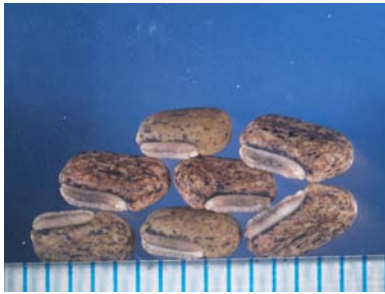
C-22, JP252355,
Vigna umbellata



C-23, JP252356,
Vigna umbellata



C-24, JP252357,
Vigna minima



C-25-1, JP252358,
Vigna minima



C-25-2, JP252359,
Vigna minima



C-26, JP252360, *Vigna unguiculata* cv-gr. *Sesquipedalis*



C-27, JP252361,
Vigna unguiculata



C-28, JP252362,
Vigna unguiculata



C-30, JP252363,
Vigna sp.



C-31, JP252364,
Vigna sp.



C-32, JP252365,
Vigna sp.



C-32.5, JP252366,
Vigna umbellata



C-33, JP252367,
Vigna minima



C-34, JP252368,
Vigna minima



C-35, JP252369,
Vigna minima



**C-36, JP252370,
*Vigna minima***



**C-37, JP252371,
*Vigna minima***



**C-38, JP252372,
*Vigna minima***



**C-39, JP252373,
*Vigna vexillata***



**C-40, JP252374,
*Vigna unguiculata***



**C-41, JP252375,
*Vigna vexillata***



**C-42, JP252376,
*Vigna minima***



**C-43, JP252377,
*Vigna radiata***



**C-44, JP252378,
*Glycine max***



**C-45, JP252379,
*Vigna unguiculata***



**C-46, JP252380,
*Vigna radiata***



**C-47, JP252381,
*Vigna unguiculata***