

## 中国雲南省およびラオス北部におけるマメ類および 共生微生物遺伝資源多様性の現地調査と保全

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## Ecological Survey and Conservation of Legume-Symbiotic Rhizobia Genetic Diversity in Southern China and Northern Laos, 2004

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### Summary

A field survey was conducted in southern China (Wenshan district of Yunnan province) from October 10 to 22, 2004 and in northern Laos (Udom Xai, Luang Nam Tha and Phongsali provinces) from November 6 to 22, 2004. In Yunnan, *Vigna umbellata*, *V. angularis*, *V. unguiculata*, *V. hirtella*, *Lablab purpureus*, *Glycine max*, and *Phaseolus vulgaris* were collected. In northern Laos, *Vigna reflexo-pilosa*, *V. unguiculata*, *V. minima*, *V. umbellata*, *V. hirtella*, *V. tenuicaulis*, *Psophocarpus tetragonolobus*, *Lablab purpureus*, and *Glycine max* were collected. In both areas, the most important traditional leguminous crop among minor ethnic groups was found to be the rice bean (*V. umbellata*). A high level of seed color variation in rice bean landraces was observed in northern Laos, while creamy tan seed color rice bean was predominant in Yunnan. Rice bean had already been harvested in mid-October in Yunnan, while

most of the rice bean plants were still in the shifting cultivation fields when we visited (mid-November) in northern Laos .

Seed samples as well as root nodules were collected when they are available and were deposited in each country of origin.

## **Introduction**

The mountainous area of northern mainland Southeast Asia and southern China is the center of “Shouyou Jurin Bunka” ( “SJ” culture) literally means “Shining-leaf forest culture” (Sasaki, 1982). This name was given because species of *Quercus*, *Cinamonum* and *Machilus* have shining leaves and are dominant canopy species in the ever green forest zone of warm temperate south, southeast and east Asia. “SJ” culture spreads from Nepal, Bhutan, Shikkim, Assam through northern mountainous Southeast Asia, southern China to Korea and southwestern Japan. “SJ” culture is characterized by several common cultural elements such as “tea drinking” , “lacquer painting” , “silk production” , fermented soybean products such as “miso” and “natto” and glutinous rice, etc. In addition, similar legends, myths and rituals are commonly found in this region.

The history of “SJ” culture is divided into 3 developmental stages, “pre-agriculture” , “millets-based shifting cultivation” and “irrigated rice cultivation” . “Shifting cultivation” is historically the most important and prevailing production system in “SJ” culture areas especially in mountainous regions. Various crops including cereals [rice (*Oryza sativa*), job's tear (*Coix lacryma-jobi*), barnyard millet (*Echinochloa* sp.)], beans [soybean (*Glycine max*), azuki bean (*Vigna angularis*), rice bean (*Vigna umbellata*), hyacinth bean (*Lablab purpureus*), sword bean (*Canavalia gladiata*), velvet beans (*Mucuna* spp.)], root crops [yams (*Dioscorea* spp.)], tea (*Camellia sinensis*) and oranges (*Citrus* spp.), etc. have been domesticated in this region.

Yunnan province of China and northern Laos are located in the central area of “SJ” culture and therefore high levels of genetic diversity can be expected for traditional crops. Various ethnic groups are living in this region keeping their own culture and this may contribute to the production and maintenance of high genetic diversity in their traditional crops. Considering the rapid urbanization of this region influenced by Chinese economic expansion, it is an urgent task to survey and conserve traditional crop germplasm and wild species diversity. In this survey, conservation of traditional legume crops, wild relatives and their symbiotic root nodules (rhizobia) are the main objectives.

## **Methods**

Field study was conducted in southern China (Wenshan district of Yunnan province) from October 10 to 22, and in Northern Laos (Udom Xai, Luang Nam Tha and Phongsali provinces) from November 6 to 22, 2004. Seeds, herbarium specimens and root nodules were collected when available. Information on collection sites including village name, altitude, latitude, longitude, habitat and other ecological data together with detailed sketch maps of the collection

sites were recorded as passport data.

## Results

### Wenshan district, Yunnan province, China (October 10 to 22, 2004)

Collected materials are listed in Table 1. A locality map of the collection sites is shown in Fig. 1. Altitude of the surveyed area ranged from 650 m to 1,600 m. Limestone-derived soil is prevailing in most of the surveyed area. Slopes of limestone hills were used for maize cultivation while flat lands were mainly used for paddy rice fields. Buckwheat (*Fagopyrum esculentum*) and edible canna (*Canna edulis*) are sporadically seen.

Among the leguminous crops, rice bean (*Vigna umbellata*) is the most popular in Wenshan district and nine accessions were collected. Zhuang ethnic group (壮族), a major ethnic group in this district, usually called this bean “Fan Dau (饭豆)” which is the same name as Han's (汉族). However, in Xiao Guang Nan (小广南) village (Hei Zhuang ethnic group 黑壮族), rice bean has an unique name “Luoli”. In another Zhuang village (Bai Mei Cun: 坝美村), a farmer told that rice bean is important on celebration days and cooked with glutinous rice or used to make “Zong Zi (粽子)”, a cake of glutinous rice filled with paste of rice bean. In a road side market near Xi Yang (西洋), rice bean was sold by Sa (Chinese letter not available) ethnic group. In a village (Cai Hua Cun: 菜花村) of the Miao ethnic group (苗族), rice bean is cultivated on a mountain located 1 to 2 hours on foot from the village under a shifting cultivation system. The population of Miao is the second largest among ethnic groups in this mountainous district. They called rice bean “Tu Tei (No Chinese letter)”. It has no relationship with celebration there. In a Yi ethnic group (彝族) village near Qiu Bei (邱北), we could not find rice bean. Seed color of collected rice bean accessions is either yellow or yellow mottled with black. Plant type of Y-15 collected in Tong Jian Cun (同剪村) is nearly erect with slightly viny shoot tip. Pod length is 10 - 12cm containing 10 - 11 seeds.

Azuki bean (*Vigna angularis*), a close relative of rice bean, is less common and less popular in Wenshan district. Two accessions (Y-7, 16) were collected. Both have a greenish white seed coat. In Xiao Guang Nan (小广南) village, it was called “Lit” (Y-7). In Guang Nan (广南) market, it was called “Mei Dao (米豆)”. In Bai Mei village (坝美村) a farmer told that she preferred rice bean to azuki bean, because it is more tasty and yields higher. In Ge Tong village (草庸村), we found azuki bean cultivated near a stream among paddy fields. It has an erect plant type with pale brown mature pods. Pod length is 8 - 10cm containing 8 - 12 seeds.

Soybean (*Glycine max*) is common in Wenshan district. Four accessions were collected. All accessions have small seeds with a yellow seed coat. In a road side market of Xi Yang (西洋), dried soybean seeds (黄豆) as well as young green pods were sold.

Yard long bean (*Vigna unguiculata*) is also common. Three accessions were collected in Zhuang and Miao villages. The seed coat color was brown.

Only one sample of hyacinth bean (*Lablab purpureus*) was collected, although it is commonly cultivated in home gardens as a vegetable. The collected material has very round shaped seeds with brown and black mottled colors.

## Northern Laos (November 6 to 22, 2004)

A list of collected materials in northern Laos is shown in Table 2. A locality map of the collection sites is shown in Fig. 1. Altitude of surveyed area ranged from 270 m to 1,300 m. Lowland river basin areas are used as paddy fields, while mountainous areas are mainly used for shifting cultivation. Diverse ethnic groups are living in northern Laos.

Rice bean (*V. umbellata*) is very common and is cultivated on the mountain sides with maize and other crops under shifting cultivation. Many ethnic groups grow this crop under their specific names. Tai Dam people call rice bean “Yo Ye” (two cases), Akah call it “Nun Shye” (one case), Khmu call it “Nyo Nye” (one case), Phunoi call it “Cong Pat” (three cases) or “Cong Shi (two cases), Ikoh call it “Nung Gong” (five cases). Seed size is somewhat larger and seed color is more diverse compared with samples observed in Yunnan, China. It shows very late maturity and viny plant type. It is usually cooked with meat or rice in a soup. In some villages, it is used to prepare “Khao Tom” (Lao language). Khao Tom is a similar food to “Zong Zi (Chinese : 粽子)” in Yunnan, China where it was used for celebration. However, it has no relationship with celebration in Laos. Unlike Yunnan, China, azuki bean (*V. angularis*) could not be found in northern Laos. Beside rice bean, winged bean (*Psophocarpus tetragonolobus*), soybean (*Glycine max*), cowpea (*Vigna unguiculata*) and hyacinth bean (*Lablab purpureus*) were collected in northern Laos.

In addition to cultivated legumes, several wild *Vigna* plants were found and collected together with their root nodules (Table 2). An accession of *V. reflexo-pilosa* (2004L-1) was found growing at the same site as the previous year. The altitude of this site is 270 m. In 2003 no seeds could be found, but a few seeds could be collected this year. Four accessions of *V. minima* were found and collected in Udom Xai and Luang Nam Tha provinces. The altitude of collection sites ranged from 718 m to 1,165 m (average 904 m). They were growing on rather shady road sides of mountain areas or on sunny places around paddy fields. Nine accessions of *V. hirtella* were found in Udom Xai, Luang Nam Tha and Phongsali provinces. The altitude ranged from 689 m to 1,300 m (average 929m). They were always found at wet river sides or wet road sides in mountain areas where many leeches were also found. Two samples of *V. tenuicaulis* were found in Phongsali province. The altitude ranged from 740 m to 949 m (average 845 m). They were found in sunny habitats around paddy fields.

## Discussion

This was the first systematic survey of cultivated and wild legumes in Yunnan China and northern Laos. Rice bean (*V. umbellata*) is the most important legume in these areas and landraces of this species are still grown. Rice bean is considered as an important crop for the future, and therefore it is necessary to collect more genetic variation of this species (Kashiwaba *et al.*, 2003). It is now difficult to collect landraces of this species in Thailand (Tomooka, 1995), because Thai farmers prefer to grow improved varieties instead of landraces. Considering this situation, it is important to collect landraces and farmers traditional knowledge before they disappear. As for wild legumes, we found 4 *Vigna* species, *V. reflexo-pilosa*, *V. hirtella*, *V. minima* and *V. tenuicaulis*. The scientific importance and the potential as breeding materials of each wild species are discussed below.

### ***V. hirtella* Ridley**

*V. hirtella* was first described by Ridley in 1920 based on a material collected on the Malay peninsula. After this, living materials had not been collected until recently, and the identity of the species has been unclear. We have collected many accessions considered to be *V. hirtella* from Malaysia, Thailand and Myanmar (Tomooka *et al.*, 1993, 1997, 2000a, 2003b). Accessions of *V. hirtella* collected in Thailand are cross compatible with azuki bean and rice bean. It shows a high level of powdery mildew resistance. Further studies regarding to its potential use as breeding materials are important (Tomooka *et al.*, 2000b, 2003a). Based on the DNA analyses using Thai and Myanmar materials, a very high level of genetic variation has been found and taxonomic revision may be necessary (Tomooka *et al.*, 2002a, Doi *et al.*, 2002, Seehalak *et al.*, 2005). Further analysis including Yunnan, China and Laos materials will be important to clarify the diversity in this important species (Tomooka *et al.*, 2002b).

### ***V. reflexo-pilosa* Hayata**

*V. reflexo-pilosa* is the only tetraploid species in the genus *Vigna* (Tomooka *et al.*, 2003). This legume (cultivated form, var. *glabra*) has a potential to become a “new” crop considering its robust nature. The number of available living materials of this crop in the world genebanks is very limited. Therefore, it is necessary to survey and collect this crop before it disappears. It seems probable that this crop is still cultivated in Laos. *V. reflexo-pilosa* var. *reflexo-pilosa* (the wild ancestor) can be used for broadening the genetic base of var. *glabra* (cultigen). For this reason, the wild *V. reflexo-pilosa* accession collected in this survey is considered to be a valuable genetic resource.

### ***V. minima* (Roxb.) Ohwi & Ohashi**

*V. minima* can cross with azuki bean and rice bean and is considered to be a useful gene source for these crops (Yoon *et al.*, 2000, Tomooka *et al.*, 2002a). Up to now, we have collected accessions of *V. minima* in Thailand and Myanmar.

Judging from the growth pattern of *V. minima* in Laos, this species seems to have a high potential as a cover crop to protect paddy bunds. After rice harvest, they can be used as forage. Further studies regarding symbiotic nitrogen fixation ability, allelopathic effects and water logging resistance are important research targets for studying *V. minima* genetic resources.

### ***V. tenuicaulis* N.Tomooka & Maxted**

This is the first record of this species in Laos. Formerly it was known only from Thailand and Myanmar (Tomooka *et al.*, 2002b, 2003). This species can cross with azuki bean and rice bean. A Thai accession showed complete resistance to bruchid beetles.

## **Acknowledgements**

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## 和文摘要

中国雲南省から東南アジア大陸部北部山岳地帯は、照葉樹林文化の中核に当たる地帯であり、古くから焼畑農耕を生産基盤にした多様な民族が暮らしてきた場所である。農耕の長い歴史のもとに、多様な民族がそれぞれに独自の栽培品種を成立させてきたものと考えられるため、在来作物の高い遺伝的多様性が期待できる。本調査では、中国雲南省文山壮族苗族自治州およびラオス北部のLuang Prabang 県, Udomxai 県, Luang Nam Tha 県, Phongsali 県を探索し、伝統的マメ科作物、その近縁野生種および共生している根粒菌の収集保全を行った。

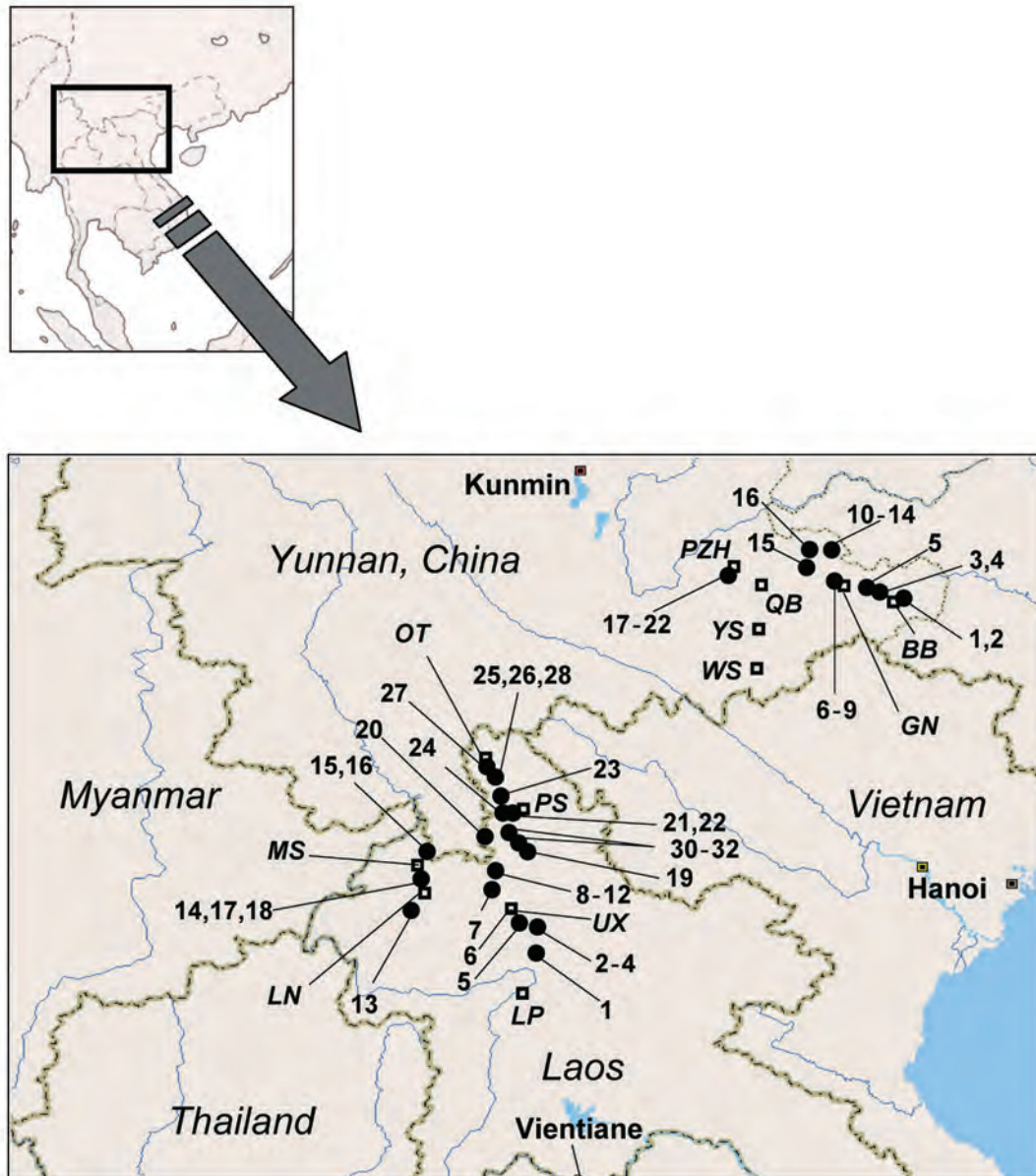


Fig.1 Collection sites of legume genetic resources in Yunnan Province, China and northern Laos. Numbers on the map correspond to accession no. in China (Table 1) and in Laos (Table 2).

City name abbreviation

China: BB(Babao), GN(Guangnan), PZH(Pu Zhe Hei), QB(Qiubei), AS(Aenshan)

Laos: LN(Luang Nam Tha), LP(Luang Prabang), MS(Muang Sing), OT(Ou Thai), PS(Phongsali), UX(Udom Xai)

Table 1. Legume materials seen in Wenshan District of Yunnan Porvince, China

## 中国雲南省文山壮族苗族自治州におけるマメ類遺伝資源

Date	Acc. No.	Species	Local name	Collection site, Altitude (m)	Ethnic group
15-Oct	2004Y-1	<i>Vigna umbellata</i>	Fan Dou (飯豆)	ca. 10km NE of Babao (八宝), 1,036m	Zhuang (壮族)
"	2004Y-2	<i>Lablab purpureus</i>	Daudei	Jin Ji Cun (金鳥村), ca. 5km E of Babao, 1,030m	Miao (苗族)
16-Oct	2004Y-3	<i>Vigna umbellata</i>	Fan Dou (飯豆)	Xi Yang Shi Chang (西洋市場), 645m	Sa group
"	2004Y-4	<i>Glycine max</i>	黄豆	"	—
"	2004Y-5	<i>Vigna umbellata</i>	Fan Dou (飯豆)	Bao Zhea Shi Chang (宝朕市場: 南甲産), 1,276m	—
"	2004Y-6	<i>Vigna umbellata</i>	Luoli	Xiao Guang Nan (小広南), 1247m	Hei Zhuang (黒壮族)
"	2004Y-7	<i>Vigna angularis</i>	Lit	"	"
"	2004Y-8	<i>Vigna unguiculata</i>	—	"	"
17-Oct	2004Y-9	<i>Vigna umbellata</i>	Fan Dou (飯豆)	Guang Nan Shi Chang (広南市場: 白泥塔産), 1,200m	—
"	2004Y-10	<i>Vigna umbellata</i>	—	Bai Mei Cun (坝美村), N of Guang Nan (広南), 847m	Zhuang (壮族)
"	2004Y-11	<i>Vigna unguiculata</i>	—	"	"
"	2004Y-12	<i>Glycine max</i>	—	"	"
"	2004Y-13	<i>Vigna umbellata</i>	Fan Dou (飯豆)	"	"
"	2004Y-14	<i>Vigna hirtella</i>	—	"	"
18-Oct	2004Y-15	<i>Vigna umbellata</i>	—	Tong Jian Cun (同剪村), NE of Guang Nan (広南), 1,600m	"
"	2004Y-16	<i>Vigna angularis</i>	—	Ge Tong Cun (草庸村), NE of Guang Nan (広南), 1,536m	"
20-Oct	2004Y-17	<i>Glycine max</i>	Tua Da	Cai Hua Cun (菜花村), near Pu Zhe Hei (普者黑), NE of Qiubei (邱北)	Miao (苗族)
"	2004Y-18	<i>Vigna unguiculata</i>	—	"	"
"	2004Y-19	<i>Phaseolus vulgaris</i>	—	"	"
"	2004Y-20	<i>Vigna umbellata</i>	Tu Tei	"	"
"	2004Y-21	<i>Glycine max</i>	Tu Da	"	"
"	2004Y-22	<i>Phaseolus vulgaris</i>	—	"	"



Table 2. A list of legume genetic resources collected in northern Laos

## ラオス北部におけるマメ類遺伝資源

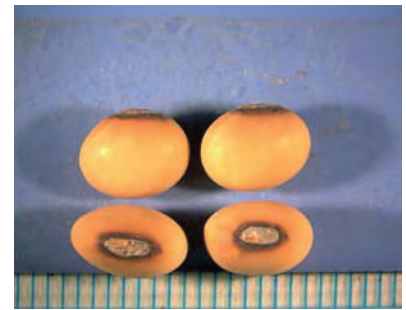
	Acc. No.	Species	Status	Local name	Collection site, Province	Altitude (m)	Ethnic group
11 Nov.	2004L-1	<i>Vigna reflexo-pilosa</i>	wild		ca. 50km N of Luang Prabang, Luang Prabang	270	
	2004L-2	<i>Psophocarpus tetragonolobus</i>	cultivated	Tua Pu	B. Kajed, S of Udom Xai, Udom Xai	740	Khmu
	2004L-3	<i>Lablab purpureus</i>	cultivated	Ple Dap	"	"	"
	2004L-4	<i>Vigna unguiculata</i>	cultivated	Tua Ring Suang	"	"	"
	2004L-5	<i>Vigna minima</i>	wild		S. of Udom Xai, Udom Xai	1,165	
12 Nov.	2004L-6	<i>Vigna umbellata</i>	cultivated	Nyo Nye	Udom Xai market (from China)	600	
	2004L-7	<i>Vigna hirtella</i>	wild		N. of Udom Xai, Udom Xai	778	
	2004L-8	<i>Vigna umbellata</i>	cultivated	Yo Ye	B. Ay, Namong Dist., Udom Xai	785	Yang (Tai Dam)
	2004L-8.5	<i>Vigna umbellata</i>	cultivated	Yo Ye	"	"	"
	2004L-9	<i>Vigna minima</i>	wild		"	"	"
	2004L-10	<i>Vigna hirtella</i>	wild		B. Muteun, Namong Dist., Udom Xai	949	Akah muteun
	2004L-11	<i>Vigna minima</i>	wild		"	"	"
	2004L-12	<i>Vigna hirtella</i>	wild		"	"	"
13 Nov.	2004L-13	<i>Vigna hirtella</i>	wild		B. Chalernsuk, Luang Nam Tha	689	Khmu
14 Nov.	2004L-14	<i>Vigna hirtella</i>	wild		between Luang Nam Tha and Muang Sing, Luang Nam Tha	1,060	
	2004L-15	<i>Vigna minima</i>	wild		N. of Muang Sing, Luang Nam Tha	718	
	2004L-16	<i>Vigna umbellata</i>	cultivated	Nung Gong	N. of Muang Sing, Luang Nam Tha	750	Ikoh
	2004L-17	<i>Vigna umbellata</i>	cultivated	Nung Gong	between Luang Nam Tha and Muang Sing, Luang Nam Tha	762	Akah
	2004L-18	<i>Vigna hirtella</i>	wild		"	"	"
16 Nov.	2004L-19	<i>Vigna hirtella</i>	wild		S. of Boun Tai, Phongsali	1,300	
	2004L-20	<i>Vigna umbellata</i>	cultivated	Guang Shi Gong Pat	B. Luangkhoun, near Chinese border, Phongsali	750	Phunoi
17 Nov.	2004L-21	<i>Vigna umbellata</i>	cultivated	Gong Shi Gong Pat	near Phongsali, Phongsali	1,213	"
	2004L-22	<i>Vigna umbellata</i>	cultivated	Gong Shi	B. Bakolong, Phongsali	1,074	"
	2004L-23	<i>Vigna umbellata</i>	cultivated	Nung Gong	B. Huay Yen, Phongsali	1,011	Ikoh
	2004L-24	<i>Vigna tenuicaulis</i>	wild		Boun Neua, Phongsali	949	
18 Nov.	2004L-25	<i>Vigna hirtella</i>	wild		N. of Boun Neua, Phongsali	797	
	2004L-26	<i>Vigna hirtella</i>	wild		S. of Ou Tai, Phongsali	1,078	
	2004L-27	<i>Vigna tenuicaulis</i>	wild		Ou Tai, Phongsali	740	
	2004L-28	<i>Vigna umbellata</i>	cultivated	Nung Gong	B. Pa Hok Gao, N. of Boun Neua, Phongsali	947	Ikoh
19 Nov.	2004L-30	<i>Vigna umbellata</i>	cultivated	Nung Gong	B. Sano Mai, Phongsali		Ikoh
	2004L-31	<i>Glycine max</i>	cultivated		"		"
	2004L-32	<i>Vigna unguiculata</i>	cultivated	Nung Tao	B. Aya, Phongsali	1,304	Hmon



2004Y-1  
(*Vigna umbellata*)



2004Y-2  
(*Lablab purpureus*)



2004Y-4  
(*Glycine max*)



2004Y-7  
(*Vigna angularis*)



2004Y-8  
(*Vigna unguiculata*)



2004Y-22  
(*Phaseolus vulgaris*)



2004L2  
(*Psophocarpus tetragonolobus*)



2004L4  
(*Vigna unguiculata*)



2004L9  
(*Vigna minima*)



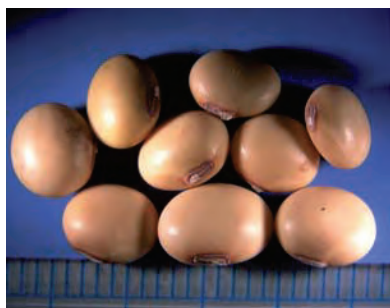
2004L17  
(*Vigna umbellata*)



2004L19  
(*Vigna hirtella*)



2004L20  
(*Vigna umbellata*)



2004L31  
(*Glycine max*)



2004L32  
(*Vigna unguiculata*)



2004L3  
(*Lablab purpureus*)

Photo 1. Some examples of legume genetic resources collected.