

Collection of Wild Soybean (*Glycine soja*) and Wild Azuki Bean (*Vigna angularis* var. *nipponensis*) in Gunma and Nagano prefectures, Japan, 2014

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Summary

We have conducted a field survey for collecting the wild crop relatives of soybean (*Glycine max*) and azuki bean (*Vigna angularis*) in Gunma and Nagano prefectures, Japan, from 15th to 17th October, 2014. A total of 38 accessions, consist of 15 wild soybean (*Glycine soja*), 1 cultivated azukibean (*Vigna angularis* var. *angularis*) and 22 wild azuki bean (*Vigna angularis* var. *nipponensis*), was collected. The collected seeds were conserved in the National Institute of Agrobiological Sciences (NIAS) genebank. We plan to multiply the seeds and to evaluate their growth traits in 2015. The multiplied seeds will become available upon request from NIAS genebank for research and educational purposes.

KEY WORDS : *Glycine*, *Vigna*, wild crop relatives, genetic resources

Introduction

The genus *Glycine* and *Vigna* belong to the legume family (Leguminosae), and include crops such as soybean (*Glycine max*), cowpea (*Vigna unguiculata*), mung bean (*Vigna radiata*), and azuki bean (*Vigna angularis*). Their wild relatives are expected to have favorable characters for breeding biotic and abiotic stress tolerant crops (McCouch *et al.*, 2013, Palmgren *et al.*, 2015). Since wild soybean (*Glycine soja*) and wild azuki bean (*Vigna angularis* var. *nipponensis*) are cross compatible with cultivated soybean and cultivated azuki bean, respectively, their diversity should be conserved for the breeding program.

The NIAS genebank has been conducting collecting trips for the conservation of wild soybean and wild azuki bean germplasm distributed in Japan (Tomooka *et al.*, 2010, Vaughan *et al.*, 2010, see also Annual Report on Exploration and Introduction of Plant Genetic Resources, NIAS, https://www.gene.affrc.go.jp/publications.php#plant_report). However, wild soybean and wild azuki bean germplasm from Gunma and Nagano prefectures were very few in the NIAS genebank (Tomooka *et al.*, 1998, Weerasekera *et al.*, 2002). Therefore, we decided to conduct a field survey for collecting wild soybean and wild azuki bean plants in Gunma and Nagano prefectures, Japan.

Methods

A field survey was conducted by car in Gunma and Nagano prefectures, Japan from 15th to 17th October, 2014 (Table 1). When we saw wild soybean or wild azuki bean from the car or saw the environment where these wild legumes seemed to grow, we stopped our car and searched the area in order to collect bulk seeds from natural populations. As a passport data, the collection site name, latitude, longitude, altitude, sketch map of the collection sites and special characteristics of the habitats and plants we noticed were recorded. The latitude and longitude were measured by WGS84 world geodetic system using Garmin GPSmap 60CSx.

Table 1. Itinerary of a collection trip to Gunma and Nagano prefectures (Oct. 15-17, 2014)

群馬県および長野県における探索の日程

| Date | Itinerary | Stay |
|---------|---|-----------|
| Oct. 15 | Tsukuba (NIAS) - Maebashi - Shibukawa - Minakami - Numata - Shibukawa | Shibukawa |
| Oct. 16 | Shibukawa - Nakanojo - Higashiagatsuma - Naganohara - Kusatsu - Tsumagoi - Karuizawa - Miyota - Saku | Saku |
| Oct. 17 | Saku - Shimonita - Tomioka - Takasaki - Tsukuba (NIAS) | |

Results and Discussion

A total of 38 accessions consisted of 15 wild soybean, 1 cultivated azuki bean and 22 wild azuki bean were collected (Table 2). Passport information of each accession is summarized in Table 3. Locations of the collection sites are shown in Fig. 1. Wild soybean and wild azuki bean plants were found growing in a grassland of riverside, beside paddy rice fields, on the embankment of railway, or in abandoned paddy fields (see Exploration Photos; Photo 1 ~ Photo 16).

Collected samples could be divided into 3 groups based on the collection sites (Fig. 1, Table 2). Group 1 consists of samples collected at the central areas of Gunma prefecture (G-1 ~ G-13). Group 2 consists of samples collected in the western areas of Gunma and eastern areas of Nagano prefecture (G-14 ~ G-23). Group 3 consists of samples collected in the southern areas of Gunma prefecture (G-24 ~ G-38). Among the 18 collection sites of wild legumes, we could found both wild soybean and wild azuki bean growing sympatrically at 5 sites (Photos 1, 2, 6, 14 and 16).

In the collection areas of Group 1 (central Gunma) and the Group 3 (southern Gunma), we could find wild azuki bean populations more than wild soybean populations. On the contrary, we have experienced difficulty in finding wild azuki bean population in the Group 2 areas (western Gunma and eastern Nagano). This might be related to the environment of the habitats influenced by the altitude. Altitude of the collection sites of Group 1, 2 and 3 ranged from 217 m to 458 m, 650 m to 1050 m, and 72 m to 379 m, respectively. This suggests that wild soybean can survive at higher altitude habitat compared with wild azuki bean. The highest altitude of the collection site of wild azuki bean in the present survey was 650 m (G-14 in the Group 2). At altitude higher than 700 m, only wild soybean populations were found and collected (G-16 ~ G-23). The highest altitude of wild soybean collection site in the present survey was 1050 m. To confirm suggested altitudinal difference of their habitat, passport data of wild soybean and wild azuki bean collected in Japan were retrieved from the NIAS genebank database. Based on the passport data, altitude of collection sites ranged from 0 to 1050 m (average: 86 m) for wild soybean (1084 accessions) and ranged

Table 2. A summary of collected samples in Gunma and Nagano prefectures

群馬県および長野県における収集品の地域別内訳

| Group of samples | Collection areas | Range of altitudes among collection sites | No. of collected samples | | |
|------------------|--|---|--------------------------------------|--|--|
| | | | Wild soybean (<i>Glycine soja</i>) | Wild azuki bean (<i>Vigna angularis</i> var. <i>nipponensis</i>) | Cultivated azuki bean (<i>Vigna angularis</i> var. <i>angularis</i>) |
| Group 1 | Central areas of Gunma | 217 m ~ 458 m | 3 | 9 | 1 |
| Group 2 | western areas of Gunma and eastern areas of Nagano | 650 m ~ 1050 m | 8 | 2 | 0 |
| Group 3 | southern areas of Gunma | 72 m ~ 379 m | 4 | 11 | 0 |
| Total | | | 15 | 22 | 1 |

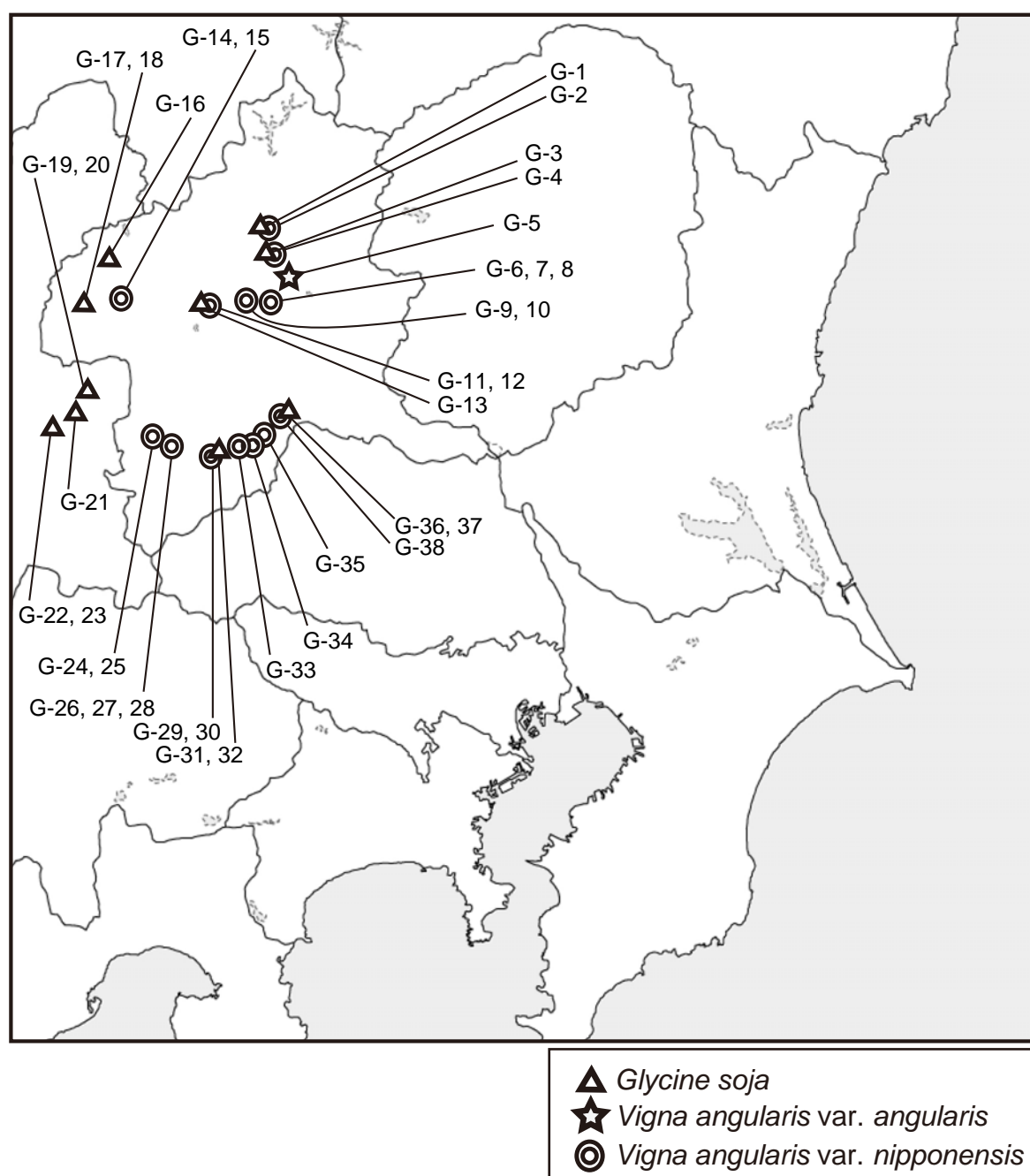


Fig. 1. A map of the collected materials in Gunma and Nagano prefectures, 2014

from 0 to 700 m (average: 113 m) for wild azuki bean (515 accessions), respectively. This might have some relationship with the fact that the northern limit of distribution area of wild azuki bean in Japan is Akita prefecture, while that of wild soybean is southern part of Hokkaido.

Variations in seed size were observed among collected samples of wild soybean (see Seed Photos). Large seeded wild soybean accessions were collected from southern Gunma (G-32, G-36 and G-37). In addition, we could not find intermediate (weedy) populations of azuki bean with tan seed coat color which were frequently found in Japan (Seed Photos of JP247253, collected in Saitama prefecture in Tomooka *et al.*, 2013, see also Seed Photos of NK-11 and NK-12-2 collected in Nagasaki and Kumamoto exploration report in this volume).

The collected seeds were conserved in the National Institute of Agrobiological Sciences (NIAS) genebank. We plan to multiply the seeds and to evaluate their growth traits in 2015. The multiplied seeds will become available upon request from NIAS genebank for research and educational purposes.

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群馬県および長野県東部におけるダイズおよび アズキ近縁野生種遺伝資源の探索収集 2014年

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摘要

本報告は、これまで保存点数が少なかった群馬県および長野県東部におけるダイズおよびアズキ近縁野生種の調査報告である。調査は2014年10月15～17日にかけて行った。その結果、ダイズ野生種 (*Glycine soja*) 15点、アズキ野生種 (*Vigna angularis* var. *nipponensis*) 22点、栽培アズキ1点、計38点の遺伝資源を収集した。これらの遺伝資源は、つくば市にある農業生物資源研究所で栽培し、特性評価と種子増殖を行う計画である。増殖種子は、農業生物資源研究所のジェンバンクで保存するとともに、研究や教育目的で利用するために配布可能な遺伝資源とする。

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

| JP No. | Coll. No. | Coll. Date | Species name | Status | Collection Site | Latitude | Longitude | Altitude (m) | Soil | 100 seed weight (g) | Remarks |
|--------|-----------|---------------|--|------------|--|--------------|---------------|--------------|------------|---------------------|---|
| 251548 | 2014G-1 | 15 Oct., 2014 | <i>Glycine soja</i> | wild | Kamimoku, Minakamimachi, Tonegun, Gunma | N36-44-06.1 | E138-58-53.5 | 425 | sand | 2.72 | river side embankment, many <i>Glycine soja</i> plants growing here |
| 251549 | 2014G-2 | 15 Oct., 2014 | <i>Vigna angularis</i> var. <i>nipponensis</i> | wild | " | " | " | " | " | 2.84 | river side embankment. a few <i>Vigna angularis</i> var. <i>nipponensis</i> plants growing here |
| 251550 | 2014G-3 | 15 Oct., 2014 | <i>Glycine soja</i> | wild | Suzuridamachi, Numata, Gunma | N36-39-04.4 | E139-01-33.3 | 330 | gravel | 2.12 | growing along small path beside paddy fields |
| 251551 | 2014G-4 | 15 Oct., 2014 | <i>Vigna angularis</i> var. <i>nipponensis</i> | wild | " | " | " | " | " | 2.27 | " |
| 251552 | 2014G-5 | 15 Oct., 2014 | <i>Vigna angularis</i> | cultivated | Tochikubo, Showamura, Tonegun, Gunma | N36-37-40.3 | E139-03-45.6 | 340 | clay | 18.60 | red seeded azuki bean planted on the ridge of terrace paddy fields |
| 251553 | 2014G-6 | 16 Oct., 2014 | <i>Vigna angularis</i> var. <i>nipponensis</i> | wild | Kawashima, Shibukawa, Gunma | N36-31-42.0 | E138-58-42.4 | 217 | gravel | 4.08 | growing on the embankment beside railway |
| 251554 | 2014G-7 | 16 Oct., 2014 | <i>Vigna angularis</i> var. <i>nipponensis</i> | wild | Kawashima, Shibukawa, Gunma | N36-31-40.36 | E138-58-46.44 | 217 | clay | 3.92 | growing beside a fallow paddy field |
| 251555 | 2014G-8 | 16 Oct., 2014 | <i>Vigna angularis</i> var. <i>nipponensis</i> | wild | Kawashima, Shibukawa, Gunma | N36-31-56.0 | E138-58-33.1 | 220 | clay | 3.48 | growing beside paddy fields |
| 251556 | 2014G-9 | 16 Oct., 2014 | <i>Vigna angularis</i> var. <i>nipponensis</i> | wild | Gochoda, Higashiagatsumamachi, Agatsumagun, Gunma | N36-33-11.22 | E138-54-25.83 | 290 | clay | 2.96 | growing in a road side grassland |
| 251557 | 2014G-10 | 16 Oct., 2014 | <i>Vigna angularis</i> var. <i>nipponensis</i> | wild | Gochoda, Higashiagatsumamachi, Agatsumagun, Gunma | N36-33-12.4 | E138-54-27.2 | 283 | clay | 3.40 | growing in a grassland beside paddy fields |
| 251558 | 2014G-11 | 16 Oct., 2014 | <i>Vigna angularis</i> var. <i>nipponensis</i> | wild | Iwashita, Higashiagatsumamachi, Agatsumagun, Gunma | N36-33-49.9 | E138-45-30.4 | 458 | gravel | 3.00 | growing in a grassland beside railway |
| 251559 | 2014G-12 | 16 Oct., 2014 | <i>Vigna angularis</i> var. <i>nipponensis</i> | wild | Iwashita, Higashiagatsumamachi, Agatsumagun, Gunma | N36-33-38.6 | E138-45-29.8 | 440 | gravel | 2.56 | a lot of <i>Vigna angularis</i> var. <i>nipponensis</i> plants growing here |
| 251560 | 2014G-13 | 16 Oct., 2014 | <i>Glycine soja</i> | wild | Iwashita, Higashiagatsumamachi, Agatsumagun, Gunma | N36-33-37.51 | E138-45-29.01 | 437 | gravel | 2.20 | a few <i>Glycine soja</i> plants with small leaflets growing |
| 251561 | 2014G-14 | 16 Oct., 2014 | <i>Vigna angularis</i> var. <i>nipponensis</i> | wild | Otsu, Naganoharamachi, Agatsumagun, Gunma | N36-33-05.2 | E138-37-07.1 | 650 | clay | 3.76 | growing in a grassland between paddy fields and railway, long pod |
| 251562 | 2014G-15 | 16 Oct., 2014 | <i>Vigna angularis</i> var. <i>nipponensis</i> | wild | Otsu, Naganoharamachi, Agatsumagun, Gunma | N36-33-03.5 | E138-37-07.8 | 645 | clay | 4.08 | growing in a grassland near paddy fields |
| 251563 | 2014G-16 | 16 Oct., 2014 | <i>Glycine soja</i> | wild | Maeguchi, Kusatsumachi, Agatsumagun, Gunma | N36-34-59.1 | E138-34-20.8 | 1050 | black soil | 1.96 | black soil habitat, growing in a grassland beside a farmer's house |
| 251564 | 2014G-17 | 16 Oct., 2014 | <i>Glycine soja</i> | wild | Kanbara, Tsumagoimura, Agatsumagun, Gunma | N36-30-15.9 | E138-32-53.1 | 988 | black soil | 1.44 | black soil habitat, growing in an abandoned field, a lot of <i>Glycine soja</i> plants covered the land |
| 251565 | 2014G-18 | 16 Oct., 2014 | <i>Glycine soja</i> | wild | Kanbara, Tsumagoimura, Agatsumagun, Gunma | N36-30-16.01 | E138-32-54.23 | 987 | black soil | 1.44 | " |
| 251566 | 2014G-19 | 16 Oct., 2014 | <i>Glycine soja</i> | wild | Oiwake, Karuizawamachi, Kitasakugun, Nagano | N36-19-52.18 | E138-33-04.61 | 934 | sand | 2.84 | growing between road and an abandoned fields |
| 251567 | 2014G-20 | 16 Oct., 2014 | <i>Glycine soja</i> | wild | Oiwake, Karuizawamachi, Kitasakugun, Nagano | N36-19-54.1 | E138-33-04.2 | 930 | sand | 1.76 | growing at an edge of an abandoned fields |

Table 3 (Continued).

| JP No. | Coll. No. | Coll. Date | Species name | Status | Collection Site | Latitude | Longitude | Altitude (m) | Soil | 100 seed weight (g) | Remarks |
|--------|-----------|---------------|--|--------|--|--------------|---------------|--------------|------|---------------------|---|
| 251568 | 2014G-21 | 16 Oct., 2014 | <i>Glycine soja</i> | wild | Housho, Miyotamachi, Kitasakugun, Nagano | N36-17-59.1 | E138-31-35.7 | 750 | clay | 3.00 | growing in a grassland dominated by <i>Pueraria</i> plants beside harvested paddy fields |
| 251569 | 2014G-22 | 16 Oct., 2014 | <i>Glycine soja</i> | wild | Kamihirao, Saku, Nagano | N36-16-52.06 | E138-29-29.77 | 700 | clay | 2.04 | growing in a fallow paddy field |
| 251570 | 2014G-23 | 16 Oct., 2014 | <i>Glycine soja</i> | wild | Kamihirao, Saku, Nagano | N36-16-53.0 | E138-29-30.4 | 700 | clay | 1.88 | " |
| 251571 | 2014G-24 | 17 Oct., 2014 | <i>Vigna angularis</i> var. <i>nipponensis</i> | wild | Minaminomaki, Shimonitamachi, Kanragun, Gunma | N36-14-24.9 | E138-42-9.3 | 377 | sand | 3.04 | growing in a grassland beside main road (R254) |
| 251572 | 2014G-25 | 17 Oct., 2014 | <i>Vigna angularis</i> var. <i>nipponensis</i> | wild | Minaminomaki, Shimonitamachi, Kanragun, Gunma | N36-14-24.6 | E138-42-12.2 | 379 | sand | 3.36 | growing in a fallow paddy field with <i>Solidago altissima</i> population |
| 251573 | 2014G-26 | 17 Oct., 2014 | <i>Vigna angularis</i> var. <i>nipponensis</i> | wild | Higashinomaki, Shimonitamachi, Kanragun, Gunma | N36-13-59.5 | E138-43-40.4 | 330 | sand | 4.40 | growing in a grassland beside terrace paddy fields, many <i>Vigna angularis</i> var. <i>nipponensis</i> plants growing but most of the pods were immature |
| 251574 | 2014G-27 | 17 Oct., 2014 | <i>Vigna angularis</i> var. <i>nipponensis</i> | wild | Higashinomaki, Shimonitamachi, Kanragun, Gunma | N36-13-56.2 | E138-43-42.3 | 334 | sand | 3.72 | growing in a grassland dominated by <i>Solidago altissima</i> population beside paddy fields |
| 251575 | 2014G-28 | 17 Oct., 2014 | <i>Vigna angularis</i> var. <i>nipponensis</i> | wild | Higashinomaki, Shimonitamachi, Kanragun, Gunma | N36-13-58.0 | E138-43-43.7 | 328 | sand | 3.44 | " |
| 251576 | 2014G-29 | 17 Oct., 2014 | <i>Vigna angularis</i> var. <i>nipponensis</i> | wild | Kannari, Tomioka, Gunma | N36-14-38.9 | E138-50-00.1 | 204 | clay | 3.08 | growing in a fallow paddy field, many <i>Vigna angularis</i> var. <i>nipponensis</i> plants were growing |
| 251577 | 2014G-30 | 17 Oct., 2014 | <i>Vigna angularis</i> var. <i>nipponensis</i> | wild | Kannari, Tomioka, Gunma | N36-14-39.7 | E138-50-08.6 | 202 | clay | 3.64 | growing in a grassland with <i>Solidago altissima</i> and <i>Typha latifolia</i> |
| 251578 | 2014G-31 | 17 Oct., 2014 | <i>Glycine soja</i> | wild | Kannari, Tomioka, Gunma | N36-14-39.3 | E138-50-08.6 | 202 | clay | 2.52 | " |
| 251579 | 2014G-32 | 17 Oct., 2014 | <i>Glycine soja</i> | wild | Kannari, Tomioka, Gunma | N36-14-41.0 | E138-50-17.7 | 201 | clay | 3.44 | growing beside an abandoned field, long peduncle |
| 251580 | 2014G-33 | 17 Oct., 2014 | <i>Vigna angularis</i> var. <i>nipponensis</i> | wild | Nanokaichi, Tomioka, Gunma | N36-15-16.1 | E138-52-57.1 | 150 | clay | 2.88 | growing just beside Kabura river, near "Tomioka Seishi Kojo" |
| 251581 | 2014G-34 | 17 Oct., 2014 | <i>Vigna angularis</i> var. <i>nipponensis</i> | wild | Shirakura, Kanramachi, Kanragun, Gunma | N36-15-16.8 | E138-56-22.1 | 142 | clay | 3.64 | growing at an edge of fallow paddy fields, near "Shirakura Shrine" |
| 251582 | 2014G-35 | 17 Oct., 2014 | <i>Vigna angularis</i> var. <i>nipponensis</i> | wild | Yoshiimachinakajima, Takasaki, Gunma | N36-15-24.8 | E139-00-28.5 | 93 | clay | 3.80 | growing in a fallow paddy field beside road, growing with <i>Solidago altissima</i> plants |
| 251583 | 2014G-36 | 17 Oct., 2014 | <i>Glycine soja</i> | wild | I-62 bridge, Negoyamachi, Takasaki, Gunma | N36-17-19.7 | E139-01-48.4 | 72 | clay | 4.04 | growing in a grassland beside a stream, a lot of <i>Glycine soja</i> plants growing also in a grassland in a stream |
| 251584 | 2014G-37 | 17 Oct., 2014 | <i>Glycine soja</i> | wild | I-62 bridge, Negoyamachi, Takasaki, Gunma | N36-17-20.39 | E139-01-49.18 | 73 | clay | 3.88 | " |
| 251585 | 2014G-38 | 17 Oct., 2014 | <i>Vigna angularis</i> var. <i>nipponensis</i> | wild | I-62 bridge, Negoyamachi, Takasaki, Gunma | N36-17-19.5 | E139-01-48.2 | 72 | clay | 2.88 | growing in a grassland beside a stream |



Photo 1. Habitat of *Glycine soja* (G-01) & *V. angularis* var. *nipponensis* (G-02), Kamimoku, Gunma



Photo 2. Habitat of *Glycine soja* (G-03) & *V. angularis* var. *nipponensis* (G-04), Tsukiyono, Gunma



Photo 3. Azuki bean (G-05) cultivated on a ridge of terrace paddy field, Numata, Gunma



Photo 4. Habitat of *V. angularis* var. *nipponensis* (G-06), Shibukawa, Gunma



Photo 5. Habitat of *V. angularis* var. *nipponensis* (G-10), Shibukawa, Gunma



Photo 6. Habitat of *V. angularis* var. *nipponensis* (G-12) & *Glycine soja* (G-13), Higashiagatsumamachi, Gunma



Photo 7. Habitat of *Glycine soja* (G-16), Kusatsumachi, Gunma



Photo 8. Habitat of *Glycine soja* (G-19) & *Glycine soja* (G-20), Karuizawamachi, Nagano



Photo 9. *Glycine soja* plants (G-19), Karuizawamachi, Nagano



Photo 10. *Glycine soja* plants (G-21) among *Pueraria* plants, Miyotamachi, Nagano



Photo 11. Habitat of *Glycine soja* (G-22) & *Glycine soja* (G-23), Saku, Nagano



Photo 12. Habitat of *V. angularis* var. *nipponensis* (G-25) Shimonitamachi, Gunma



Photo 13. Habitat of *V. angularis* var. *nipponensis* (G-27) Shimonitamachi, Gunma



Photo 14. Habitat of *V. angularis* var. *nipponensis* (G-30), Tomioka, Gunma



Photo 15. Habitat of *V. angularis* var. *nipponensis* (G-33), Tomioka, Gunma



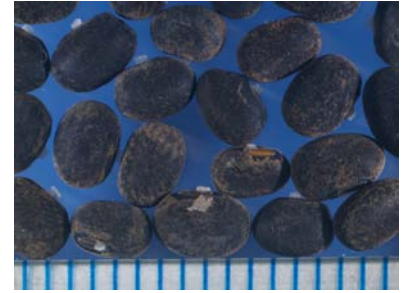
Photo 16. Habitat of *Glycine soja* (G-36), *Glycine soja* (G-37) & *V. angularis* var. *nipponensis* (G-38), Takasaki, Gunma



G-1, JP251548, *Glycine Soja*



**G-2, JP251549,
V. angularis var. *nipponensis***



G-3, JP251550, *Glycine Soja*



**G-4, JP251551,
V. angularis var. *nipponensis***



G-5, JP251552, *V. angularis*



**G-6, JP251553,
V. angularis var. *nipponensis***



**G-7, JP251554,
V. angularis var. *nipponensis***



**G-8, JP251555,
V. angularis var. *nipponensis***



**G-9, JP251556,
V. angularis var. *nipponensis***



**G-10, JP251557,
V. angularis var. *nipponensis***



**G-11, JP251558,
V. angularis var. *nipponensis***



**G-12, JP251559,
V. angularis var. *nipponensis***



G-13, JP251560, *G. soja*



**G-14, JP251561,
V. angularis var. *nipponensis***



**G-15, JP251562,
V. angularis var. *nipponensis***



G-16, JP251563, *G. soja*



G-17, JP251564, *G. soja*



G-18, JP251565, *G. soja*



G-19, JP251566, *G. soja*



G-20, JP251567, *G. soja*



G-21, JP251568, *G. soja*



G-22, JP251569, *G. soja*



G-23, JP251570, *G. soja*



**G-24, JP251571,
V. angularis var. *nipponensis***



**G-25, 251572,
V. angularis var. *nipponensis***



**G-26, 251573,
V. angularis var. *nipponensis***



**G-27, 251574,
V. angularis var. *nipponensis***



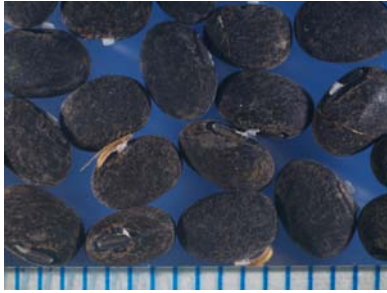
**G-28, JP251575,
V. angularis var. *nipponensis***



**G-29, JP251576,
V. angularis var. *nipponensis***



**G-30, JP251577,
V. angularis var. *nipponensis***



G-31, JP251578, *G. soja*



G-32, JP251579, *G. soja*



**G-33, JP251580,
V. angularis var. *nipponensis***



**G-34, JP251581,
V. angularis var. *nipponensis***



**G-35, JP251582,
V. angularis var. *nipponensis***



G-36, JP251583, *G. soja*



G-37, JP251584, *G. soja*



**G-38, JP251585,
V. angularis var. *nipponensis***