## "Haibushi", a New Variety of Snap Bean Tolerant to Heat Stress

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Received February 10, 1997

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#### **Abstract**

Snap bean is a heat-sensitive crop. In order to develop heat tolerant snap bean varieties, snap bean germplasm accessions were introduced from tropical countries. A total of 323 breeding lines provided by CIAT (Centro Internacional de Agricultura Tropical, Colombia), 20 varieties introduced from the Philippines, Sri Lanka and Indonesia, 6 varieties reported to be heat tolerant and provided by USDA (United States Department of Agriculture) etc. and 37 accessions collected in Malaysia and Thailand were evaluated for heat tolerance by open culture in summer at JIRCAS Okinawa Subtropical Station located in Ishigaki island, one of the sub-tropical islands of Japan. Some plants in one of the accessions from Malaysia were the most heat tolerant among the accessions tested. Therefore, one plant from this accession was chosen for further pure line selection. The line thus obtained showed a higher heat tolerance and higher yield potential than commercial varieties in local adaptability tests carried out in summer in several areas of southern Japan. Critical temperature for pod setting of the line was examined in a phytotron. The line showed a high tolerance not only to heat treatment for several days but also to heat treatment over a longer period of time like one month. When the line was exposed to high temperature for one month, the critical daily mean air temperature for pod setting ranged from 28°C to 29.5°C. Judging from the meteorological data of the cities in southern Japan, heat tolerance of the line is high enough for summer culture in southern Japan. The line was registered as a new snap bean variety and designated as Haibushi by the Ministry of Agriculture, Forestry and Fisheries, Japan in 1995.

Additional key words: evaluation, germplasm, high temperature, Phaseolus vulgaris

#### Introduction

The subtropical islands of Japan (Nansei islands) are situated at latitudes 24° to 31° along the Eastern China Sea and Pacific Ocean (Fig. 1). Monthly mean air temperature exceeds 25°C from May to October in the southernmost islands, Ishigaki island (Fig. 2). As a result, shortage of vegetables is a serious problem in agricultural production in this area. Due to the lack of heat tolerance, temperate vegetables can not be produced in summer, and heat tolerant tropical vegetables are scarcely distributed there.

Japan International Research Center for Agricultural Sciences (JIRCAS), Okinawa Subtropical Station has initiated a program for the collection, introduction and evaluation of plant germplasm from tropical countries. The center has used germplasm accessions for breeding crop varieties adapted to the soil and agro-climatological conditions of this area. Development of heat tolerant vegetables has been one of the main targets in the breeding program. In 1985 the center released a variety of winged bean (Psophocarpus tetragonolobus) named Urizun. It was the first registered variety of tropical vegetables in Japan whose production was promoted by the Ministry of Agriculture, Forestry and Fisheries, Japan (MAFF). Although the winged bean variety was accepted by farmers and consumers in the area, it could not satisfy the demand for vegetables in the hot season there. Therefore, we attempted to develop heat tolerant snap bean varieties (varieties of Phaseolus vulgaris for use of young green pods as vegetables) from germplasm accessions collected in tropical countries. Okinawa is a main producing area of snap beans in the cool season in Japan. Farmers apply advanced techniques to cultivate them. However, they do not produce snap beans in summer due to the heat susceptibility of the crop<sup>5)</sup>. If a heat tolerant variety could be released, it could be cultivated in this area. As a result, breeding work for heat tolerance was initiated.

#### **Materials and Methods**

#### 1) Germplasm introduction and collection

Breeding materials were introduced or collected during the period 1985 to 1993, from tropical countries including Malaysia, USA, the Philippines, Sri Lanka and CIAT (Centro Internacional de Agricultura Tropical, Colombia), Thailand, Indonesia.

In 1985 plant germplasm exploration by JIRCAS was carried out in the Philippines and Malaysia. Some varieties of snap bean which were expected to be mixtures were collected in Malaysia<sup>5)</sup>. In 1992, four varieties were given by the college of agriculture, UPLB. Four varieties

from Sri Lanka Plant Genetic Resources Center and two commercial varieties were given by Sri Lanka. Two heat tolerant varieties were given by USDA (United States Department of Agriculture). CIAT provided us with a set of breeding lines for international nursery for warm region (1991). In 1993 CIAT provided us again with 12 breeding lines for warm region and four varieties whose high heat tolerance had been reported previously. Some germplasm accessions of snap bean were collected in Thailand<sup>5)</sup>. In 1994 ten accessions were given by Lembang Horticultural Experiment Station, Indonesia.

# 2) Germplasm evaluation and breeding procedures of Haibushi

Pod setting potential of introduced accessions under high temperature conditions was evaluated by open culture in summer at JIRCAS, Okinawa Subtropical Station (JIRCAS Okinawa) located in Ishigaki island, Okinawa at lat. 24° 20′N (Fig. 1). They were sown in early June in order to have the period of flower formation and main harvest season coincide with the hottest period (Fig. 2). They

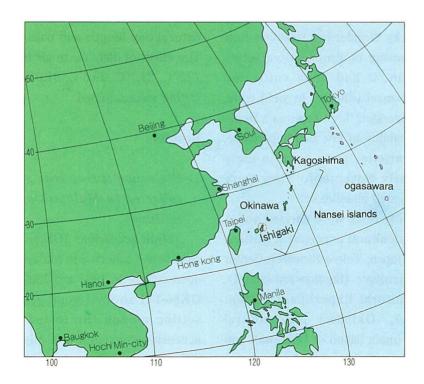


Fig. 1. Nansei islands and the southernmost island, Ishigaki island where JIRCAS Subtropical Station is located

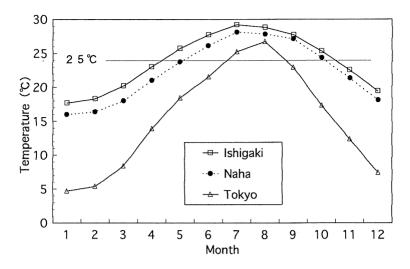


Fig. 2. Monthly mean air temperature at the meteorological station in Tokyo, Naha (Main island of Okinawa) and Ishigaki island

were cultivated according to the standard cultural practices applied for snap bean in southern Japan. Accessions selected for heat tolerance and yield potential were examined for genetic homogeneity and heterogeneous populations were subjected to pure line selection. Yield potential of selected lines, which belonged to the vinytype varieties, was tested in open culture also using poles and net. It was compared to that of a control variety, Kentucky Wonder. As Kentucky Wonder is a relatively heat tolerant variety among commercial varieties cultivated in Japan, it had been cultivated extensively in the Nansei islands. In yield tests, young pods were harvested three times in a week. In the test for adaptability to low air temperature, tested lines and varieties were sown in early October and in early November. They were cultivated in a non-heated glasshouse.

Local adaptability tests were carried out in several regional agricultural experiment stations located in southern Japan; Tokyo Research Center of Subtropical Agriculture (Ogasawara island), Kagoshima Agricultural Experiment Station (Kagoshima city), Okinawa Agricultural Experiment Station (main island of Okinawa), and Yaeyama Sub-station of Okinawa Agricultural Experiment Station (Ishigaki island). In the tests, Haibushi was sown in July except at Okinawa

Agriculture Experiment Station where sowing was performed in April. Sweetness and taste of boiled pods of Haibushi were evaluated by about ten persons for pods harvested in the summer of 1993 and 1994.

Heat tolerance of Haibushi was examined in a phytotron where the air temperature was controlled in a 12/12-h thermoperiod from 7:00 a.m. to 7:00 p.m./7:00 p.m. to 7:00 a.m. under natural day length and natural light conditions. The effect of daily mean air temperature (DMAT) 23.5°C, 28.1°C, 29.6°C, 31.0°C and 32.5°C on pod setting was examined.

### Results

1) Evaluation of snap bean germplasm accessions collected in Malaysia and development of Haibushi

Eight accessions (OK86-126 to OK86-133) of snap beans collected in Malaysia were evaluated in open culture in 1987 and 1989<sup>5)</sup>. Several plants of OK86-131 showed good growth and good pod setting among the tested accessions. The accession consisted of a mixture of phenotypes differing in pod shape and pod size. The plant with optimum pod setting which was expected to be highly heat -tolerant was selected from ten plants

cultivated in 1989. From that year until 1992, pure line selection was carried out through open culture in summer as shown in Fig. 3. In 1992 it was assumed from the observation of phenotypic characters that the line (OK86-131-1-1-1-1) developed from OK86-131 was fixed genetically. From that year yield potential tests and local adaptability tests had been carried out with the line. In 1995, OK86-131-1-1-1 was registered as a new variety by the Ministry of Agriculture, Forestry and Fisheries under the name of "Haibushi". Haibushi means southern star in the local language of Nansei islands.

# 2) Evaluation of varieties introduced from other countries

Besides Haibushi, a total of 311 testing lines for warm region given by CIAT were evaluated for heat tolerance in the summer of 1992 (Fig. 3). As shown in Fig. 4, in some of them vegetative growth was poor and they died. Although only 98 lines set flowers, the flowers sometimes showed an abnormal shape. Fifteen lines set immature small pods which did not grow further and dropped down. Only one line set a few fully enlarged pods

but the pods did not contain developed seeds. On the other hand, Haibushi set many normal pods under the same climatic conditions in this year (Fig. 5). Four accessions from UPLB (A537, A541, A550 and A554) and varieties from USDA (PI271997 and PI549536) reported to be heat tolerant<sup>3)</sup> did not set pods. Within A#00670, A#01277, A#01368, A#01383 and two local

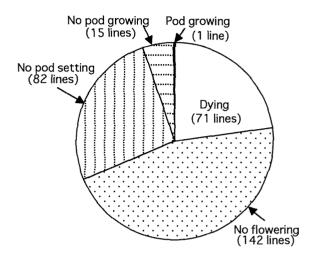


Fig. 4. Classification of 311 lines provided by CIAT by growth and pod setting potential in open culture in summer

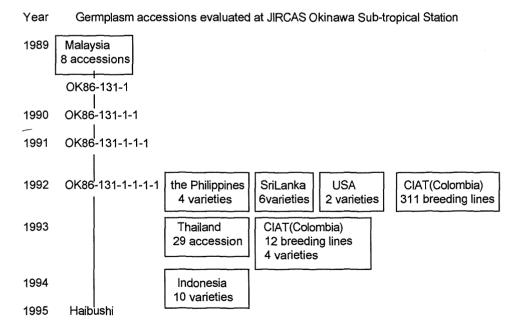


Fig. 3. Evaluation of introduced germplasm accessions of snap bean for heat tolerances and procedure for breeding of Haibushi

commercial varieties introduced from Sri Lanka, only A#00670 set several pods. However, the pod yield was far lower than that of Haibushi<sup>5)</sup>.

In 1993, mean air temperature of July and August measured at JIRCAS OKinawa was higher than in 1992 and 1994 (Fig. 6). Even in the case of Haibushi pod setting was poor in 1993 (Fig. 5).

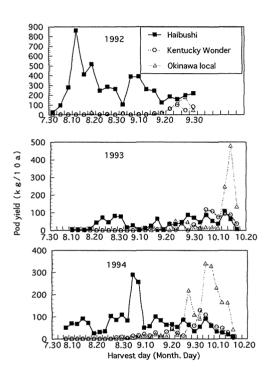


Fig. 5. Pod yield of Haibushi, Kentucky Wonder (control variety) and Okinawa local (another reference variety) in open culture in summer at JIRCAS Okinawa Subtropical Station

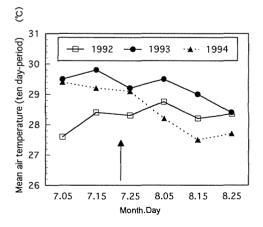


Fig. 6. Mean air temperature measured at JIRCAS Okinawa Subtropical Station. Arrow indicates the time of the onset of flowering of Haibushi.

Four heat tolerant varieties (Tamalanca, Negro hausteco, G2525 and PI271998)<sup>4,7)</sup> and 11 breeding lines given again by CIAT (764HAB472 to 784HAB492) and 29 accessions collected in Thailand (OK93-006 to OK93-034) were tested. No materials given by CIAT showed a good pod setting ability during this period. Some accessions from Thailand were heat tolerant but their pod setting ability was lower than that of Haibushi. In 1994, ten varieties introduced from Indonesia were tested but they did not set pods. No promising lines could be obtained from these accessions.

## 3) Yield potential and agronomic characteristics of Haibushi

### (1) Yield potential

From 1992 to 1994, the yield potential of Haibushi was examined in summer open culture (Fig. 5). The daily mean air temperature (DMAT) was very high in July and August, 1993 and July, 1994 which coincide with the time of flower formation and pod setting for Haibushi (Fig. 6). During these periods, pod setting was very poor and the quality of harvested pods was also low, as pods were short and bent. However, even in these years, Haibushi showed a higher heat tolerance than the control variety, Kentucky Wonder and another reference variety, OKinawa local (Fig. 5). 1992, DMAT measured at Ishigaki Meteorological Station showed a higher value than the average from mid-July to the end of August. Although the period corresponded to the duration from flowering to main pod setting in Haibushi and other tested varieties in yield potential tests, only Haibushi was able to set pods well during this period (Fig. 5). In that year, total pod yield of Haibushi reached 5000kg/ha. This yield was comparable to or higher than the average of pod yield in snap bean cropping in Japan.

#### (2) Local adaptability

In local adaptability tests, Haibushi was compared with Kentucky Wonder and reference varieties commonly produced in the areas where local adaptability tests were carried out. As DMAT of the hottest season is lower in most of areas in

southern Japan than in Ishigaki island where JIRCAS Okinawa is located and Iriomote island which is close to Ishigaki island (Fig. 7), Haibushi showed an adequate heat tolerance and higher productivity in summer open culture than Kentucky Wonder and other reference varieties at locations where local adaptability tests were

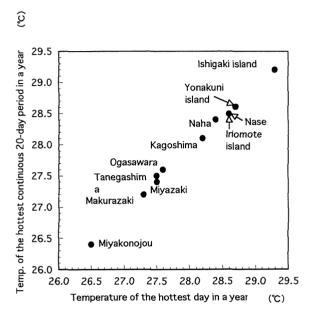


Fig. 7. Mean air temperature of the hottest 20 days and the hottest DMAT in this period in a year recorded at meteorological stations and meteorological observatories in southern Japan. Values are averages for 30 years.

carried out (Fig. 8).

#### (3) Adaptability to low air temperature

The monthly mean air temperature decreases to 17°C in January at JIRCAS Okinawa in Ishigaki island (Fig. 2). In winter culture, harvest of pods of Haibushi and other varieties started from December in the October sowing or from January in the November sowing. The harvest lasted until the end of May, except for Haibushi sown in October whose pod harvest was terminated in mid-March. In winter culture, plant growth of Haibushi was poorer than that of Kentucky Wonder and Okinawa local and pod yield of Haibushi was also lower than that of these varieties (Fig. 9).

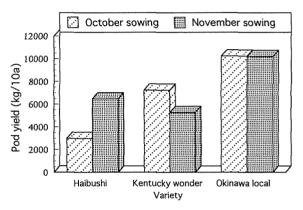


Fig. 9. Pod yield of Haibushi compared with that of Kentucky Wonder and Okinawa local sown in October and November for winter culture

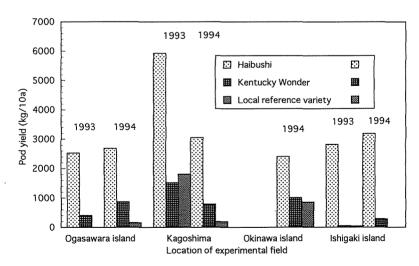


Fig. 8. Pod yield of Haibushi, Kentucky Wonder and local reference variety in local adaptability tests. As a local reference variety, "Kentucky Blue" was used in Ogasawara island and "Stayer" in other locations. The local reference variety was not cultivated in Ogasawara island in 1993. Pod yield of the local reference variety was 0kg/10a in Ishigaki island in 1994.

## (4) Morphological and agronomic characteristics of Haibushi

Morphological and agronomic characteristics of Haibushi are shown in Table 1. Haibushi belongs to the indeterminate type in growth habit (Fig. 10). Leaflet is small and pale green. Hypocotyl is reddish purple and the flower is also reddish purple. Seed coat color pattern is plain and seed coat color is black. Young pods are pale

Table 1. Morphological and agronomic characteristics of Haibushi

Hypocotyl color	Reddish purple
Growth habit	Viny, Indeterminate
Flower color	Red purple
Pod length	11.5cm
Pod width	1.1cm
Fresh weight per pod	5.3g
Number of seeds per pod	6.6
Seed coat color pattern	Plain
Seed coat color	Black
Seed shape	Oval
Days to 50% flowering	40
Photoperiodic sensitivity	Insensitive
100 seed weight	27.9g

green and they have a string. They have flat shape and are slightly bent (Fig.10). They are 10.3 to 13.3 cm long and 0.9 and 1.2cm wide. The fresh weight ranged from 4.1g to 6.5g. Boiled young pods of Haibushi are sweeter and tastier than those of Kentucky Wonder and Okinawa local (Fig. 11).

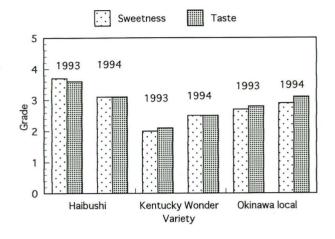


Fig. 11. Sweetness and taste of boiled pods of Haibushi compared with those of Kentucky Wonder and Okinawa local. They were scored into five grades from 1 (not sweet, not tasty) to 5 (sweet, tasty) by testers.

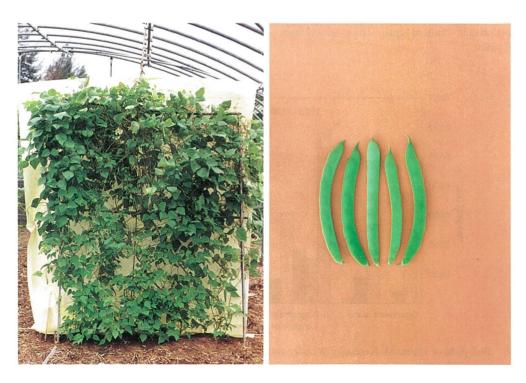


Fig. 10. Plant shape at flowering stage (left) and young pods (right) of Haibushi

#### (5) Resistance to pests and diseases

In yield tests carried out at JIRCAS Okinawa and in local adaptability tests, root rots were the main diseases in snap beans. Susceptibility to the plant diseases of Haibushi was not different from that of Kentucky Wonder and other reference varieties in the tests.

#### 4) Heat tolerance of Haibushi

In Japan, the hottest season occurs from July to August. Mean air temperature of the hottest 20 days and the hottest DMAT during this period increases to around 28.5°C or less at most of the meteorological stations and meteorological observatories located in southern Japan (Fig. 7). Exceptional high value of air temperature measured at Ishigaki Meteorological Station must be attributed to radiant heat from buildings surrounding the station which is located in an urban area. Therefore, air temperature in agricultural fields in Ishigaki island should be similar to that measured in Iriomote Island Meteorological Observatory. Because Iriomote island is only 20km's distant from Ishigaki island and the meteorological observatory is located in rural area without the influence of surrounding buildings. Therefore, in Japan, if a snap bean variety can set pods at 28.5°C, the heat tolerance of the variety is suitable for summer open culture in any locations in southern Japan. In open culture carried out at JIRCAS Okinawa in 1992, Haibushi was able to set pods well at the DMAT of 28.5°C where air temperature of the test field was observed by ourselves (Figs. 5 and 6).

Heat tolerance of Haibushi was examined under precisely controlled temperature conditions in a phytotron and it was compared to that of Kentucky Wonder. The pod setting ratio (ratio of numbers of pods to numbers of flowers) was not different between Haibushi and Kentucky Wonder at a low air temperature (DMAT 23.5°C) (about 80% for both varieties) (Fig. 12). As DMAT increased from 23.5°C to 32.5°C, the pod setting ratio decreased in the flowers which opened during the first five days of the heat treatment in both

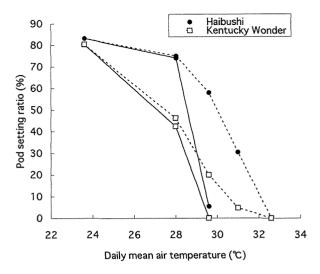


Fig. 12. Pod setting ratio of flowers which opened during the first 5 days (-----), and that of flowers which opened from 25 days to 29 days after the beginning of heat treatment (———)

varieties. However, the differences in the pod setting ratio were obviously significant between Haibushi and Kentucky Wonder at temperature from 28.1°C to 31.0°C (Fig. 12). Pod setting ratio of the flowers which opened from 25 days to 29 days after the beginning of the heat treatment (DMAT) 28.1°C) was also higher in Haibushi than in Kentucky Wonder (Fig. 12). This result indicated that Haibushi is more tolerant than Kentucky Wonder to heat treatment of both types, short period (5 days) and long period (up to 29 days). However, even in the case of heat tolerant Haibushi, pod-setting was nor observed during the long exposure to DMAT 29.6°C (Fig. 12). This result indicates that the critical temperature for pod setting ranges from 28.1 to 29.6°C under prolonged treatment in Haibushi.

#### Discussion

Breeding for heat tolerance of snap bean has been mainly carried out in the USA<sup>2)</sup>. Hot spell which lasts several days is a constraint on pod setting there. Previous physiological studies on heat tolerance of snap bean were also concentrated on the hot spell<sup>1, 6, 8)</sup>. However, in tropical and subtropical countries, hot days last longer, at least

more than one month (Fig. 2). Snap bean varieties cultivated in summer must be tolerant to this type of heat stress. In the evaluation of snap bean germplasm for heat tolerance in the present study, all the lines introduced from CIAT, the varieties introduced from tropical countries and even the varieties previously reported to be heat tolerant could not set pods (Fig. 4). It is assumed that they were not tolerant enough to long periods of hot days. Haibushi was found to be tolerant to both short and long periods of hot days (Figs. 5, 8 and 12)

The critical temperature for pod setting of Haibushi was around 28°C to 29.5°C during a long period of heat treatment (Fig. 12). Judging from the meteorological data, the heat tolerance of Haibushi was sufficient to get a good pod vield in summer open culture in all the locations of southern Japan. However, DMAT may exceed the critical temperature for pod setting of Haibushi there. The heat tolerance of Haibushi is not always sufficient to obtain a stable and good yield in summer culture (Fig. 5). Breeding for heat tolerance of snap bean should be further promoted. We should collect germplasm accessions that are more heat tolerant for heat tolerance breeding. We should collect snap bean or common bean (grain type varieties of *Phaseolus vulgaris*) accessions from many tropical countries, especially from South Asia, Africa and Central America, since snap beans are widely produced and heat tolerance is essential for cultivars to be planted in hot seasons in these areas.

On the other hand, greenhouse cultivation in winter showed that the adaptability of Haibushi to cool conditions was lower than that of reference varieties (Fig. 9). It appears that the yield reduction in Haibushi was caused by poor shoot growth under cool conditions. It is recommended that the cropping season of Haibushi should be restricted to warm seasons in the subtropical regions.

Regarding other characteristics of Haibushi, pale green color of young pods is not appreciated by Japanese consumers. We should improve the pod color by crossing with varieties with a dark green color. Cross-breeding should also be carried out between Haibushi and varieties resistant to root rots and other diseases. It was reported that the heat tolerance of snap bean to hot spell is a hereditary character<sup>4</sup>. However, the genetic aspects of tolerance to a long period of hot days remain to be investigated. Furthermore, a convenient screening method for the tolerance to a long period of heat stress has to be developed in order to carry out selection effectively with many progenies. Haibushi is anticipated to contribute to basic studies on genetics of heat tolerance and to development of a breeding method for heat tolerance in snap beans.

#### Acknowledgements

The authors appreciate the assistance of Dr. H. Yoshikawa, Director of Vegetables Breeding Division, National Research Institute of Vegetables, Ornamental Plants and Tea and staff of the Division. We thank the institutes which provided us with snap bean germplasm and also the staff of regional agricultural experiment stations for carrying out the local adaptability tests.

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## 若莢用インゲンマメ新品種「ハイブシ」の育成

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### 摘要

南西諸島を中心に、我が国の亜熱帯地域においては、 高温障害が夏期の野菜栽培における大きな隘路になって いる。この問題を克服する方策の一つは野菜の耐暑性の 向上である。南西諸島の中心部である沖縄県は、我が国 でも有数の冬期の若莢用インゲンマメ(サヤインゲン) の栽培地域である。冬期にはサヤインゲン栽培が盛んに 行われているが、インゲンマメは耐暑性に乏しいため、 沖縄県のサヤインゲン消費は6月から10月の間は県外か らの移入に頼っているのが現状である。そこで、熱帯諸 国から遺伝資源を導入し耐暑性サヤインゲンの育成をは かった。1985年にフィリピン、マレーシアで遺伝資源収 集調査を行い遺伝資源8点を収集して以来,1994年まで フィリピン、スリランカ、インドネシアから合計20品種 の遺伝資源を導入した。1993年にはタイで29点収集した。 また合衆国のUSDA(合衆国農業局)等から過去に耐 暑性と評価されたものを6品種導入した。コロンビアの CIAT(国際熱帯作物研究センター)から高温地帯向 けの配布系統を計323系統譲り受けた。これらを、国際 農林水産業研究センター・沖縄支所圃場(石垣市)で夏

期に露地栽培し耐暑性を評価した。その結果、マレーシ アで収集した遺伝資源の中に有望なものが見いだされた ので、雑駁な種子集団から純系分離を行い、耐暑性と収 量性に優れる系統を育成した。生産力検定試験や各地の 系統適応性試験等を経て、1995年この系統は農林命名登 録され「ハイブシ」と名付けられた。ファイトトロンを 用いた調査で、ハイブシの着莢のための高温限界は、連 続一ヶ月の高温処理条件下では28~29.5℃の間にあるこ とが判明した。我が国、西南暖地や亜熱帯島嶼部の高温 期の気温データから判断すると、ハイブシの耐暑性は我 が国の夏期栽培用品種としてはほぼ満足できると考えら れた。従来、サヤインゲンの耐暑性育種は、合衆国を中 心に一週間程度の短期の高温に対する耐性を目標にすす められてきた。本研究では、そのような育種研究で耐暑 性と評価された品種が着莢できない長期の高温下で良好 に着莢するハイブシを育成した。ハイブシは高温が長期 間続く熱帯、亜熱帯向けの耐暑性品種であると考えられ る。

キーワード:遺伝資源、インゲンマメ、高温ストレス、熱帯、Phaseolus vulgaris

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