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CHARACTERIZATION OF INTRODUCTIONS OF GLYCINE JAVANICA L.

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SUMMARY

A collection of 127 introductions of *Glycine javanica* L. was established at the Kairi Research Station in north-eastern Queensland between 1965 and 1968.

A system of grouping introductions into Types is presented upon the basis of reported chromosome number and arbitrary categories of time of commencement of flowering and vegetative-reproductive periodicity. Ten morphological forms are described. Each introduction is assigned to a Type-morphological form.

The interbreeding behaviour of some introductions was also studied. Tetraploid forms exhibited hybridism involving heterosis in vigour and depression in fertility, while diploid forms interbred freely. Status as evolutionary entities is proposed for some Type-morphological form combinations.

I. INTRODUCTION

Glycine javanica L. (*G. wightii*) is a herbaceous perennial legume which plays a significant role in pasture improvement in areas of Queensland and northern New South Wales.

Three cultivars—Tinaroo, Cooper and Clarence—are currently available, and each has arisen from selection between introductions in different environments (Allen 1960; Edye 1967).

Pritchard and Wutoh (1964) reported the existence of diploid and tetraploid forms. Edye and Kiers (1966) described variation in seed yield, stolon development and frost resistance at Lawes, and Edye (1967) reported upon aspects of agronomic performance in a number of environments. Investigations into the nature of genetic differences between introductions were reported by Wutoh, Hutton, and Pritchard (1968 *a, b*).

A programme of varietal improvement was commenced at the Kairi Research Station in north-eastern Queensland in 1964. Improved cultivars are sought with respect to wider edaphic adaptation, longevity of growing season, density of stolon branching and rooting, and efficiency of *Rhizobium* symbiosis.

This self-pollinating species (Hutton 1960) is of very recent domestication and introductions can be regarded as random collections from natural populations rather than selections of known agronomic merit. A system of grouping the available material upon criteria relating to degree of genetic diversity and agronomic worth was sought.

II. METHODS

Field characterization of phenotype.—The Kairi Research Station is situated at 17°S. latitude at an elevation of 2,200 ft. The climate is of an elevated tropical nature. Mean annual rainfall is 50 in., with approximately three-quarters of this occurring between December and March. Storm rains precede the main rainy season and may occur from late October to early January. The period from April to June is characterized by long periods of very cloudy conditions and light rains. Light frosts occasionally occur between May and August. The dominant soil type is a deep, red brown clay loam derived from basalt under a vegetation of rain-forest.

Between 1965 and 1968, 127 introductions of *Glycine javanica* were observed in the field at the Research Station. Seeds were inoculated with a commercial inoculant and sown into 3 in. round jiffy pots. Month-old seedlings were transplanted into field plots of two rows 5 lk apart. Plots were maintained free of weeds and intermittently grazed. Particular introductions, designated as standards, were planted in plots randomly scattered among the other introductions.

The following observations were made on each introduction:—

(1) Time of commencement of flowering: observations were made once a week on the time when the first floret opened, using a linear scale commencing on November 30.

(2) Growth ratings: visual estimates were made on a 1-5 scale, each month, of total plant bulk and the vigour of vegetative and reproductive growth components.

(3) Morphology: variations in individual plant parts were described and used as the basis for description of distinct morphological forms.

The nature of the various planting groups and the seasons (from October 1 to September 30) during which the plants were observed are presented in Table 1. It should be noted that in the case of flowering time, which can interact with planting date, data from within the season of establishment were disregarded, except in the case of material planted in January 1968 (group IV).

TABLE 1

DETAILS OF PLANTING GROUPS AND RECORDING SEASONS FOR *Glycine javanica* INTRODUCTIONS AT KAIRI RESEARCH STATION

Planting Group				Recording Season (Oct. 1–Sept. 30)	
Identity	Planting Date	No. of Introductions		Flowering	Growth and Morphology
		New	Standard		
I	Feb. 1965	32	0	1965–66, 1966–67	1965–66, 1966–67
II	Mar. 1966	41	0	1966–67	1966–67, 1967–68
III	Jan. 1967	40	6	1967–68	1967–68
IV	Jan. 1968	14	6	1967–68	1967–68

Chromosome number.—Chromosome numbers were drawn from Pritchard and Gould (1964) and from data kindly provided by Dr. A. J. Pritchard, Division of Tropical Pastures, Commonwealth Scientific and Industrial Research Organization.

Interbreeding behaviour.—A number of flowering and morphological variants of both diploid and tetraploid chromosome numbers were grown in pots in a glasshouse. Crosses were made between Tinaroo and each of C.P.I. 30366, 23411 and 25422 and Cooper; between Cooper and each of C.P.I. 30366, 25918 and 23411; and between Clarence and each of C.P.I. 18070, 27020 and 28279.

Fine forceps were used to emasculate expanded buds from 12 to 14 hr before onset of anthesis. Standard, wing and keel petals were broken off to allow removal of anthers. Pollination was effected immediately by making gentle contact between ruptured anthers of the pollen parent and the stigma of the seed parent. Pollen was then obtained from florets collected at the time of anthesis earlier that day and stored in a glassine bag on a laboratory bench. Stamens bearing anthers with moist masses of semi-extruded pollen were dissected out with the aid of a binocular microscope. After pollination the entire potted plant was stood in a water-bath and covered with a sealed plastic cage for 18 hr.

F1 and F2 populations were successfully established in the field in 1967 and 1968 respectively as spaced plants, along with small parental populations. The hybrids Tinaroo x 23411 and those between Cooper and each of 30366, 25918 and 23411 were not grown as F2's.

Growth ratings were made as previously described. In addition, seed yield per plant was recorded.

III. RESULTS

(a) Time of Commencement of Flowering

Introductions commenced to flower each season from mid December until early June.

The distribution of flowering time of planting group I in each of two seasons included four groupings consisting of the same introductions which maintained the same relative flowering order. These groupings were taken as the basis for defining flowering categories. The mid-points of the discontinuities between adjacent groupings in each season were used to define the limits of four seasonal flowering categories. These categories were termed very early, early, mid-flowering and late flowering.

The definition of categories so defined for the 1966-67 season was then superimposed upon the flowering distribution of planting group II in that season.

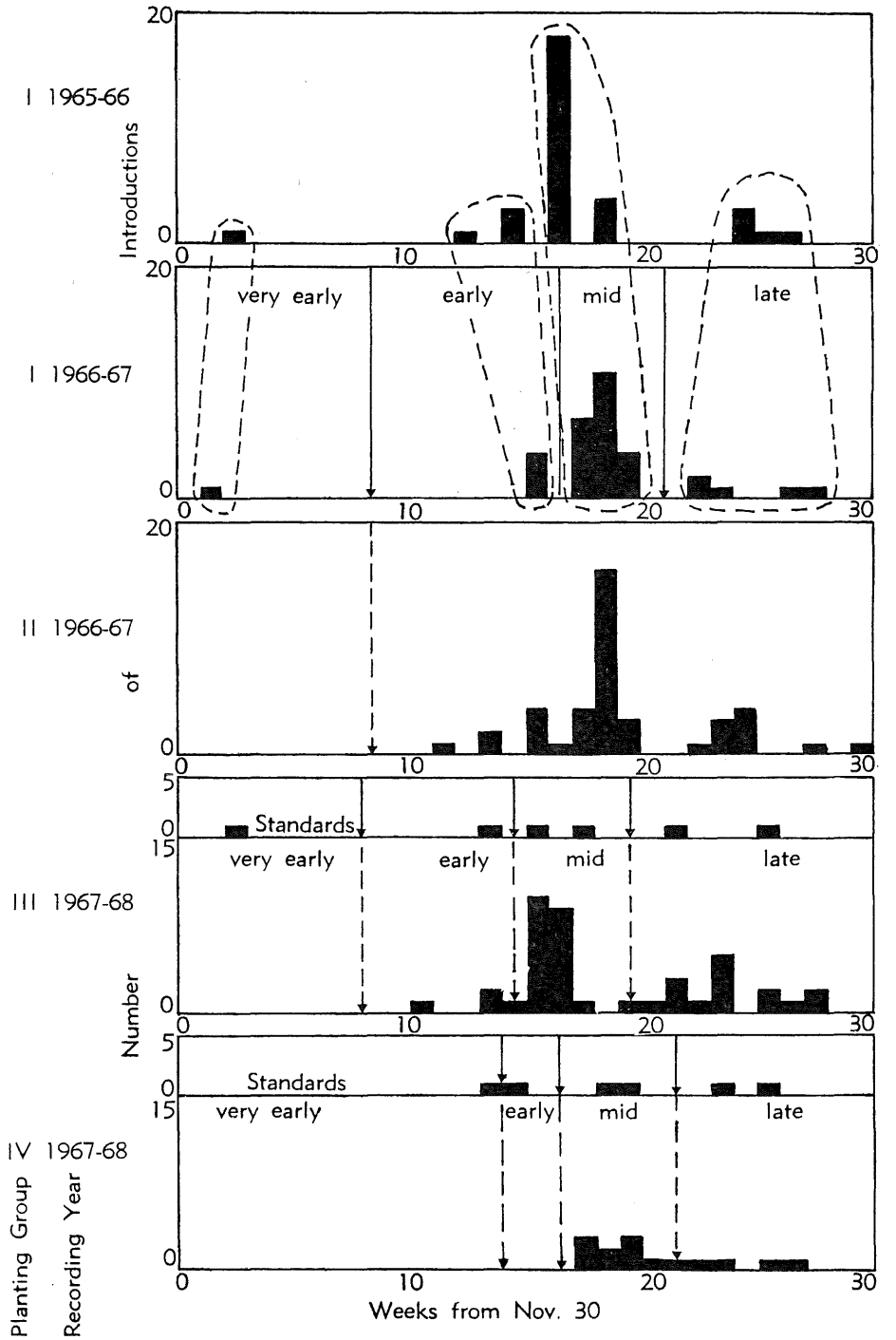

Fig. 1.—Flowering time distribution and category definition of individual planting groups.

Introductions from planting group I representing each category and termed standards were established with planting groups III and IV. In the 1967-68 season, as group IV was only in its establishment season, the mid-points between the times of flowering of these standards were used to define category limits separately for these planting groups.

Figure 1 illustrates the flowering time distribution and category definition for the various planting groups. By such means a category description was applied to each introduction.

(b) Growth

Seasonal vigour.—During the period December to March in each season all introductions made prolific growth. Some introductions exhibited a pronounced capacity for growth during other times of the season in the absence of temperature and moisture extremes.

In October 1966, early storms relieved moisture stress and C.P.I. 18103, 25703, 26263, 27834 and 29744 showed pronounced early-season vigour among the established introductions at that time.

In late June of 1967 and 1968, with adequate moisture and in the absence of frost, a high late-season vigour rating was applied to C.P.I. 18070, 20737, 27022, 28279, 30599, 30640 and 34129, Tinaroo, CQ-700, K50194, K51324 and K51394.

Vegetative—reproductive periodicity.—The overall growth pattern observed was that peak vegetative growth preceded reproductive growth, and upon flowering and fruiting, vegetative growth slowed down. However, in the absence of temperature and moisture extremes, and defoliation, two distinct categories of periodicity were recognized:

(*a*) Marked periodicity. Upon commencement of flowering a flowering peak is soon reached. Fruiting then becomes the major physiological function and vegetative growth and flowering are reduced to a low level.

(*b*) Moderate periodicity. Upon commencement of flowering a flowering peak is gradually attained. While fruiting proceeds, vegetative growth and flowering continue at a reduced but still appreciable rate.

In the Kairi environment the majority of introductions were assigned a marked periodicity. The introductions C.P.I. 32944 and 30366 and CQ-700 best illustrate the moderate category.

(c) Morphology

Diversity was recorded with regard to size, shape, coloration and vestiture of plant structures, including leaves, racemes, florets, pods and seeds. Some clearly distinguishable variation is described in Table 2.

TABLE 2

SOME MORPHOLOGICAL DIVERSITY AMONG *Glycine javanica* INTRODUCTIONS AT KAIRI RESEARCH STATION

Characteristic	Descriptive Standards			
Shape of terminal leaflet	(i) = Form a = ovate b = oblong c = orbicular	(ii) = Apex a = acute b = acuminate c = obtuse d = mucronate		
Vestiture of pods and stem	(iii) = Degree a = hirsute b = pubescent c = glabrous	(iv) = Inclination a = appressed b = ascending c = erect	(v) = Direction a = antrorse b = retrorse	(vi) = Colour a = white b = tawny
Raceme	(vii) = Density of florets a = compact b = spaced	(viii) = Floret size a = < 5 mm b = 5–7 mm c = > 7 mm	(ix) = Standard petal marking a = nil b = pink c = violet d = red e = purple	(x) = Calyx pigmentation a = nil b = slight c = intense
Pod	(xi) = Colour a = black b = dark brown c = light brown	(xii) = Shape a = cylindrical b = laterally flattened		

Ten distinct morphological forms can be recognized by different combinations of individual structural variations. These are described in Table 3, using the descriptive standards presented in Table 2. Each introduction was assigned to a morphological form.

TABLE 3

DESCRIPTION OF MORPHOLOGICAL FORMS OF *Glycine javanica* GROWN AT KAIRI RESEARCH STATION

Morphological Form		Shape of Terminal Leaflet		Pod Vestiture				Raceme				Pod	
No.	Example†	(i)*	(ii)	(iii)	(iv)	(v)	(vi)	(vii)	(viii)	(ix)	(x)	(xi)	(xii)
1	cv. Tinaroo ..	a*	a	a	a, b, c	a	b	b	a	c	a	b	a
2	30498 ..	a	b	a	a	a	b	b	a	a, b	a	c	b
3	cv. Cooper ..	b	a, c	a	b	a	a	b	b	d	b	a	a
4	cv. Clarence..	a	a	a	b	a, b	a	b	b	e	a	a	a
5	30363 ..	a	b	a	b	a	b	b	a	b	b	b	a
6	32944 ..	a	a, c	b	b	a	b	b	b	d	a	b	a
7	28279 ..	a, c	a, b, c	a	b	a, b	b	b	b, c	e	c	b	a
8	K51112 ..	a	b	c	—	—	—	b	a	c	a	b	b
9	32696 ..	a	a	a	b	b	b	b	a	e	c	b	a
10	30600 ..	a	a, c	a	b	a	b	a	b	e	b	a	b

† cv. = cultivar name; K = Kitale number; others are Commonwealth Plant Introduction numbers. * Descriptive standards used in Table 2.

(d) A Proposed System of Grouping into Types

Level of ploidy has been linked with categories of time of commencement of flowering and vegetative-reproduction periodicity. This resulted in 16 potential combinations. These combinations have been used as definition of Types, which are identified by the use of capital letters A to P in Table 4.

TABLE 4

DEFINITION OF TYPES OF *Glycine javanica* GROWN AT KAIRI RESEARCH STATION

Type Identity		Flowering Category	Periodicity Category
Diploid*	Tetraploid†		
A	I	Very early	Marked
B	J	Very early	Moderate
C	K	Early	Marked
D	L	Early	Moderate
E	M	Mid	Marked
F	N	Mid	Moderate
G	O	Late	Marked
H	P	Late	Moderate

* 2n = 22 chromosomes.
† 2n = 44 chromosomes. Tetraploids from Pritchard (personal communication).

Types can be further characterized by reference to their morphology. Type-morphological forms are identified by linking the respective capital letter referring to Type from Table 4 with the numerals describing the appropriate morphological form from Table 3.

The 127 introductions in this collection have been assigned into 10 Types and 25 Type-morphological forms. The distribution of introductions into Type-morphological forms is presented schematically in Table 5.

(e) Interbreeding Behaviour

F1 generation.—Progeny of all hybrids were quite normal in general plant form, with the exception of a proportion from the hybrid Clarence x 25279. The abnormal form was stunted and somewhat grotesque, with internode length and leaf size reduced. Growth of this form was never vigorous, but all plants were persistent at 20 months from planting.

All hybrids except the abnormal form of Clarence x 28279 showed heterosis for vigour when considered relative to mid-parent values, and some—e.g. Clarence x 27020, Clarence x 18070, Tinaroo x Cooper—and the normal form Clarence x 28279 showed true heterosis based upon increase over higher parent value.

TABLE 5

DISTRIBUTION OF INTRODUCTIONS OF *Glycine javanica* GROWN AT KAIRI RESEARCH STATION INTO TYPE-MORPHOLOGICAL FORM

Type-Morphological Form	Identity of Introductions Included	Type-Morphological Form	Identity of Introductions Included
A–		I–	
B–1	30366	J–	
C–2	25422, [28278], 30361, 30362, 30528, 30533, 30535, K609	K–4	25336, 31639
C–3	25918		
D–2	25421, 30496, 30498	L–	
E–4	16417, 16673, 18419, [34005]	M–4	13300, 13420, 16830, 17185, 17673, 24520, 25296, 25333, 25423, 25431, 25701,[25703],25919,25920,26263, 27835, 28277, 28320, 28826, 29308, 29804, 30077, 30079, 30080, 30359, 30367, 30471, 30495, (30499), 30529, 30530, 30531, 30532, 30638, 30639, 30815, 32020, 33159, 34589, 34830, (39085), (Q5097), (Q5098), Q7004, Q7009, Q7013, (Q10885), (Q10887), (Q10888), Mexico, Clarence
E–1	30214A, 33154		
E–3	18103, (25700), Cooper, 27834, 29744, 32942, (Q10886), (Q7008)		
E–5	30363, 30497, 30341, 30643		
F–6	32862, (32939), 32944, 33157	M–7	27020
F–8	CQ700	N–4	25337
G–4	34656	O–4	30360, K51212
G–7	27019, K50194	O–7	2702, 18070, 27021, 27022, 28279, 37922, K51324, K51394
G–1	15905, 20737, 30365, 30396, 30599, 30640, 32941, 34125, 34126, 34127, 34128, 34129, 39083, Tinaroo		
G–3	23411		
G–8	(Q10299), (K50101), K51112		
G–9	(32696), K52338		
G–10	12600, 30600, Q11394		
H–		P–	

Q = Queensland Department of Primary Industries number; K = Kitale number; others are Commonwealth Plant Introduction numbers.

( ) = Chromosome number as assumed.

[ ] = Introductions reported as including diploid and tetraploid forms. Here diploid is assumed.

Each hybrid set some seed. However, Clarence x 28279, Clarence x 27020 and Clarence x 18070 showed marked negative heterosis on both mid- and high-parent values for seed per plant. The hybrid involving Tinaroo and Cooper showed positive heterosis for seed set per plant based upon mid-parent values.

In view of the positive heterosis for vigour and negative heterosis for seed set in the hybrids, Clarence x 28279, Clarence x 27020 and Clarence x 18070, the actual fertility of these hybrids was further examined. Their fertility, when expressed as weight of seed set per unit of bulk rating, exhibited marked negative heterosis. While these bulky plants flowered profusely, there was a high degree of floret abortion (greater than 90% in the 3 days following anthesis), and the pods which developed were of reduced size (generally only 1–3 seeds). This behaviour pattern was consistent in the two years. Table 6 shows the nature of the heterotic response for vigour and fertility of these hybrids.

TABLE 6

HETEROSIS IN *Glycine javanica*, GROWN AT KAIRI RESEARCH STATION FOR VIGOUR AND FERTILITY, IN TWO YEARS, MEASURED AS PERCENTAGE INCREASE OVER HIGHER PARENT

F1 Identity	Heterosis			
	Vigour		Fertility	
	1967	1968	1967	1968
Clarence x 27020 ..	74	16	−90	−63
Clarence x 28279† ..	38	56	−99	−98
Clarence x 28279* ..	−98	−98	−99	−99
Clarence x 18070 ..	12	33	−99	−99

† = Normal plant form.
* = Abnormal plant form.

F2 generation.—All progeny from the normal F1 form of Clarence x 28279 were normal in the F2. In a population of three plants from the abnormal F1 form, two plants were normal and one abnormal. The abnormality was of a more extreme nature, and at 10 months the plant was below 1 in. in height and had not flowered.

As F1 and F2 populations were not grown under comparable circumstances, an assessment of depression in vigour due to inbreeding was restricted to a comparison of the relationship between mean mid-parent values and mean F1 values, and mean mid-parent values and mean F2 values. On this basis there was a general reduction in vigour in all hybrids from F1 to F2.

F2 populations of Tinaroo x Cooper, Tinaroo x 25422 and Tinaroo x 30366 were fully fertile and showed mean seed set values ranging about mid-parent values.

The populations of Clarence x 18070, Clarence x 27020 and Clarence x 28279 were again characterized by a high percentage of floret abortion, reduced pod size and low values for seed set per plant and for fertility. Some plants of each hybrid set no seed. Variance for seed was highest in Clarence x 27020

and lowest in Clarence x 28279. Plants normal in form derived from the abnormal F1 form of Clarence x 28279 were quite fertile. The seed set and fertility characteristics of these hybrids are presented in Table 7.

TABLE 7

SEED SET AND FERTILITY OF SOME *Glycine javanica* F2 HYBRIDS AND THEIR PARENTS GROWN AT KAIRI RESEARCH STATION

Parent or F2 Identity	Mean Seed Set (g/plant)	Mean Fertility (g seed/unit bulk)
18070	4·50	1·80
27020	21·10	8·93
28279	7·40	2·47
Clarence	10·95	7·30
Clarence x 27020	1·43	0·74
Clarence x 28279†	0·69	0·25
Clarence x 28279*	7·00	2·80
Clarence x 18070	0·32	0·17

† = Normal form plants from normal form F1.
* = Normal form plants from abnormal form F1.

IV. DISCUSSION

The polymorphic nature of *Glycine javanica* L. has been used as a basis of classification by other workers. The morphological forms numbered 1, 3, 4, 7 and 8 are recognized as Hermann's (1962) botanical taxa, subspecies *micrantha* (Hochst. ex A. Rich) F. J. Herm.; variety *moniliformis* (Hochst. ex A. Rich) F. J. Herm.; variety *claessensii* (De Wild) Hauman; subspecies *pseudo-javanica* (Taub.) Hauman respectively. The morphological forms 2, 4, 7, 8 and 9 are similar to Bogdan's (1966) "Flat seed Rhodesian Glycine", "Common East African Glycine", "Round Leaf Glycine" and "Fine Violet Glycine", respectively. The introduction C.P.I. 18103 is mentioned by Bogdan as conforming to his "Leathery Leaf Glycine". In this study, however, C.P.I. 18103 conformed to morphological form 3, and the description of "Leathery Leaf Glycine" is in accordance with form 4.

Clausen (1962) regards morphologic, ecologic and genetic relationships as criteria of basis evolutionary significance. The grouping of introductions into Type-morphological forms proposed here is the basis of chromosome number (genetic), flowering time and growth periodicity (ecologic), and morphology. When the nature of the interbreeding behaviour of particular introductions is also considered, status as evolutionary entities can be applied. Following the nature of such entities as presented by Clausen (1962), an ecospecies relationship is proposed between Clarence and each of C.P.I. 27020, 18070 and 28279. The relationship between Tinaroo and each of C.P.I. 23411 and Cooper, and between C.P.I. 30366 and Cooper, is proposed as that of subspecies. The relationship between Cooper and each of C.P.I. 25918 and 23411 and that between Tinaroo

and C.P.I. 30366 are proposed as ecotypic. These relationships could be tentatively extended to apply to the Type-morphological combinations these introductions represent.

Further evidence of ecological differences between Type-morphological forms may result from a consideration of the geographic distributions of component introductions, including the altitude aspect.

Speciation, according to Clausen (1962), may begin with either morphological, ecological or genetical differentiation, or more or less simultaneously in any two or three of these directions. This divergence syndrome has proceeded to the greatest extent betwen these introductions to which an ecospecies relationship has been assigned. Pritchard (personal communication) reports the tetraploid nature of each of the introductions so involved, i.e. Clarence and C.P.I. 18070, 27020 and 28279. The diploid condition is reported for those introductions interbred where divergence was confined to morphological or ecological aspects. This wider divergence in the tetraploid population could result from the effects of polyploidy upon different diploid pregenitors, which Stebbins (1950) referred to as intervarietal autopolyploidy. A more speculative inference would be that different adaptation was conferred upon different tetraploid forms, contributing to their respective isolation and accumulation of diversity.

This system of grouping introductions into Type-morphological forms has been used as a basis for nomination of candidate introductions into screening trials with various objectives. These can be for (*a*) profuse rooting and branching of stolons; (*b*) early and effective nodulation; (*c*) maximum length of growing season and high annual level of dry matter production; and (*d*) wide edaphic adaptation.

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REFERENCES

ALLEN, G. H. (1960).—Glycine—a pasture legume for Queensland. *Qd Agric. J.* 86:273.

BOGDAN, A. V. (1966).—*Glycine javanica* under experimental conditions in Kenya. *Trop. Agric., Trin.* 43:99.

CLAUSEN, J. (1962).—"The Evolution of Plant Species". (Hafner: New York.)

EDYE, L. A. (1967).—Yield comparisons of thirty-eight introductions of *Glycine javanica* in swards in three environments. *Aust. J. Exp. Agric. Anim. Husb.* 7:342.

EDYE, L. A., and KIERS, H. J. (1966).—Variation in maturity, stolon development and frost resistance of *Glycine javanica*. *Aust. J. Exp. Agric. Anim. Husb.* 6:380.

HERMANN, F. J. (1962).—A revision of the genus *Glycine* and its immediate allies. *Tech. Bull. U.S. Dep. Agric.* No. 1268.

HUTTON, E. M. (1960).—Flowering and pollination in *Indigofera spicata, Phaseolus lathyroides, Desmodium uncinatum,* and some other tropical legumes. *Emp. J. Exp. Agric.* 28:235.

PRITCHARD, A. J., and GOULD, K. F. (1964).—Chromosome numbers in some introduced grasses and legumes. *Tech. Pap. Div. Trop. Past. C.S.I.R.O. Aust.* No. 2.

PRITCHARD, A. J., and WUTOH, J. G. (1964).—Chromosome numbers in the genus *Glycine* L. *Nature, Lond.* 202:322.

STEBBINS, JR., G. L. (1950).—"Variation and Evolution in Plants". (Columbia University Press: New York.)

WUTOH, J. G., HUTTON, E. M., and PRITCHARD, A. J. (1968*a*).—Combining ability in *Glycine javanica. Aust. J. Agric. Res.* 19:411.

WUTOH, J. G., HUTTON, E. M., and PRITCHARD, A. J. (1968*b*).—Inheritance of flowering time, yield and stolon development in *Glycine javanica. Aust. J. Exp. Agric. Anim. Husb.* 8:317.

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