BIOLOGIA PLANTARUM 36 (1): 37-45, 1994

Cytological relationships of selected species of *Panicum* L.

M.A. HAMOUD, S.A. HAROUN, R.D. MACLEOD*, and A.J. RICHARDS*

Botany Department, Faculty of Science, Tanta University, Tanta Egypt
*Department of Biology, University of Newcastle upon Tyne, Newcastle, England**

Abstract

The cytological investigation of 12 taxa of *Panicum* L. revealed that the vast majority of them have the basic number x = 9 at different ploidy levels. The basic number x = 8 was recorded only in the tetraploid species *P. maximum* with 2n = 32. The diploid number 2n = 18 was encountered in *P. capillare*, *P. laevifolium*, *P. antidotale* and *P. coloratum* (2) with 3B-chromosomes recorded in the latter species. The tetraploid chromosome number 2n = 36 was found to exist in *P. miliaceum*, *P. miliare*, *P. coloratum* (1) and *P. virgatum*. The hexaploid number 2n = 54 was recorded in *P. bulbosum*, *P. dichotomiflorum* and *P. esculentum*. The karyotypes of all accessions were mostly symmetrical and mainly comprised of meta-and submeta-centric chromosomes with little variation in length among them within each karyotype. Investigation of chromosome association during metaphase I of meiosis revealed that the frequency of bivalents/cell was the highest among all investigated diploid, tetraploid and hexaploid accessions. Univalents were also frequently encountered in various accessions. These results may indicate that segmental alloploidy has been the major process by which polyploid species have originated.

Introduction

The genus *Panicum* L. comprises more than 500 species distributed in both tropical and subtropical warm regions. Cytological studies in this genus have been restricted mostly to the determination of chromosome numbers, which have been reported for almost one third of the total number of species of *Panicum* L. The chromosome numbers reported are in multiples of 7, 8, 9, 10 or 11 suggesting up to five basic numbers for this genus (Darlington and Wylie 1955, Gould 1960, Bolkhovskikh *et al.* 1969, Christopher and Abraham 1976, Bouton *et al.* 1981, Kumatsu and Suzuki 1987). However, the basic numbers 9 and 10 are predominate in the genus, and numbers which are not multiples of them may have evolved by cytological aberrations (Tateoka 1965, Jauhar and Joshi 1965, 1969).

Received 20 April 1993, *accepted* 11 May 1993.

In the present study, attempts are made to use cytological features to analyse and add some information to help in the understanding of species relationship in the genus *Panicum* L.

Material and methods

Materials of the investigated taxa were obtained from various sources (Table 1). Plants were grown under glasshouse conditions and their identification was verified by comparing them with herbarium material of voucher specimens kept in the Herbarium of the Royal Garden, Edinburgh, Great Britain.

Table 1. Cytological features of some species of *Panicum* L. (chr - chromosome).

Source	Taxon	Chr no. 2n	Basic no. x	Ploidy level	Mean chr length [μm] ± S.E	Mean arm ratio ± S.E.
H.	1 - *P. capillare*	18	9	2x	1.20 ± 0.11	1.17 ± 0.28
D.	2 - *P. laevifolium*	18*	9	2x	1.32 ± 0.15	1.72 ± 0.43
D.	3 - *P. antidotale*	18	9	2x	1.05 ± 0.13	1.13 ± 0.06
G.	4 - *P. miliaceum*	36*	9	4x	0.96 ± 0.10	1.04 ± 0.02
M.	5 - *P. miliare*	36	9	4x	1.16 ± 0.07	1.13 ± 0.05
F.	6 - *P. coloratum* 1	36*	9	4x	1.53 ± 0.11	1.36 ± 0.12
D.	7 - *P. coloratum* 2	18+3B	9	2x	1.08 ± 0.12	1.30 ± 0.18
D.	8 - *P. maximum*	32*	8	4x	0.91 ± 0.03	1.23 ± 0.09
H.	9 - *P. virgatum*	36	9	4x	1.96 ± 0.22	1.31 ± 0.12
G.	10 - *P. bulbosum*	54	9	6x	1.21 ± 0.07	1.29 ± 0.04
J.	11 - *P. dichotomiflorum*	54	9	6x	0.87 ± 0.05	1.08 ± 0.03
J.	12 - *P. esculentum*	54	9	6x	1.02 ± 0.08	1.07 ± 0.02

*Satellite

Sources Codes:
- H. Hortus Botanicus Nationalis (Belgium).
- D. Directorate of plant and seed control (South Africa).
- J. Jardin Botanique de Bordeaux (France).
- F. Field collections (Egypt).
- G. Garten der Universität Basel (Switzerland).
- M. Musée d'Histoire Naturelle (France).

For studies of mitotic chromosomes, root-tips of about one month old plants were pretreated with 0.4 % colchicine for 3 h. Roots were then washed with distilled water and fixed in freshly prepared mixture of ethanol : glacial acetic acid (3:1 v/v) for 24 h at room temperature. Roots were kept in 70 % ethanol in refrigerator at 4 °C for up to four weeks. Washed root-trips were hydrolysed in 1 N HCl at 60 °C for 14 min, transfered to distilled water for 2 min and stained in leuco-basic fuchsin for 1 - 3 h at room temperature.

The terminal 1 mm of deeply stained root apex was squashed in a drop of 45 % acetic acid on a slide. Cells with fully contracted and well spreading metaphase

chromosomes were photographed using an automatic exposure *Olympus* camera and *Timax* 100 film (100 ASA). Photographic prints were constructed by arranging chromosomes in order of their relative length and arm ratios as measured from prints. Variation in chromosome length and arm ratios within the karyotype were measured by calculating their means and standard errors (± S.E).

Chromosome associations during diakinesis and metaphase I of meiosis were also recorded from temporary preparations. Buds were fixed in 3:1 ethanol:glacial acetic acid mixture for 24 h and anthers were stained and squashed in 0.75 % aceto-carmine solution.

Results and discussion

The cytological features, including chromosome number, basic number, ploidy level, mean chromosome length and arm ratio of the investigated accessions of *Panicum* L. are given in Table 1. In addition, mean frequencies of chromosome association during diakinesis and metaphase I of meiosis are given in Table 2. Also, karyotypes of various taxa are shown in Figs. 1 - 12. These are comprised mostly of meta- and submeta-centric chromosome types. However, they differed in many other cytological details.

Table 2. Mean frequencies of chromosome assocciation during metaphase I of meiosis in various accessions of *Panicum* L.

Species	2n	Mean number of chromosomes assocciation			
		I	II	III	IV
P. capillare	18	1.00	8.50	0.00	0.00
P. laevifolium	18	2.00	8.00	0.00	0.00
P. antidotale	18	1.20	8.40	0.00	0.00
P. miliaceum	36	1.77	18.11	0.07	0.00
P. miliare	36	3.00	16.50	0.00	0.00
P. coloratum 1	36	2.44	14.60	0.66	0.66
P. coloratum 2	18+3B	2.51	6.70	0.00	0.00
P. bulbosum	54	3.85	18.85	1.28	2.28
P. dichotomiflorum	54	1.20	24.20	0.95	0.85
P. esculentum	54	6.22	19.55	0.88	1.55

I: univalent, II: bivalent, III: trivalent, IV: quadrivalent.

P. capillare (Fig. 1): The samples investigated of this species show a diploid chromosome number of 2n = 18. This number has been also reported by Avdulov (1928), Gould (1960), and Sokolovskaya and Probatova (1972). The karyotype of this species consists mainly of metacentric and submetacentric chromosomes in addition to few acrocentric ones. The variation among chromosomes in length and arm ratio is noticible as indicated by the relatively high values of S.E. (± 0.11 and ± 0.28 for mean length and arm ratio, respectively).

Examination of the first metaphase of meiosis revealed a high similarity between chromosome pairs where bivalents showed a proportion of 8.5/cell (Table 2). However, the presence of some univalents (1.0/cell) indicates that some inter-chromosome differences occur.

P. leavifolium (Fig. 2): The chromosome number 2n = 18 was also found in this species. Similar counts have been reported by Nath and Swaminthan (1957) and Kumatsu and Suzuki (1987). However, tetraploid number of 2n = 48 was recorded in this species by Hutchinson and Bashaw (1964). The karyotype of this species is similar to that of the previous one. However, it showed satellited pair of chromosomes and higher mean arm ratio ((1.72 ± 0.43). The high value of S. E. indicates a considerable variation among chromosomes in centromere position. In addition, the chromosomes showed lower proportion of bivalents (8.0/cell) and higher univalents (2.0/cell) at metaphase I of meiosis. This may indicate higher chromosomal changes such as within arm translocations and paracentric inversions which could prevent normal pairing at meiosis.

P. antidotale (Fig. 3): The chromosome number 2n = 18, recorded for this species in the present study, is confirmed by Burton (1942), Jauhar and Joshi (1965) and Kumatsu and Suzuki (1987). The tetraploid number 2n = 36 has been reported also by Narayan (1962).

The karyotype of this species, being comprised of metacentric chromosomes with little variation in centromere position among chromosomes, could be considered more primitive than those of the aforementioned two species. However, the frequency of chromosome association at metaphase I for this species was very close to that of *P. capillare*.

P. miliaceum (Fig. 4): This species has been found tetraploid with 2n = 36. The same number has been recorded by Hunter (1934), Chandula (1959) and Singh and Godward (1960). However, chromosome count of 2n = 42 was reported by Rau (1929) and Avodulov (1931) whereas 2n = 40 was reported by Church (1929) in this species.

The karyotype is comprised mainly of metacentric chromosomes with one pair satellited. The variability among chromosomes of the karyotype was mainly due to differences in mean length (S.E .± 0.10) rather than in arm ratio (S.E .± 0.02). The high frequency of bivalent formation (18.0/cell) during metaphase I of meiosis indicates an allotetraploid origin for this species. The variation between the two genomes, however, must be subchromosomal one since the chromosomes are morphologically alike.

P. miliare (Fig. 5): This species have also shown chromosome count of 2n = 36. This number has been reported earlier by Rau (1929) and more recently by Christopher and Abraham (1976).

Being less variable in lengths of chromosomes, the karyotype of this species is more symmetrical than that of the previous one. The bivalent frequency (19.5/cell)

recorded at metaphase I of meiosis indicates that this species is also allotetraploid. However, the high value of univalent formation (3.0/cell) may indicate a high degree of subchromatid changes in some chromosomes that prevent their synapsis during meiosis.

P. coloratum (Figs. 6, 7): The two accessions investigated of this species have diploid and tetraploid chromosome numbers of 2n = 18 and 36. These and other chromosome numbers have been reported by many authors. The chromosome numbers encountered are 2n = 18, 36, 45 and 63 (Pritchard and Delacy 1974), 2n = 32 and 54 (Joshi *et al.* 1959, Hutchinson and Bashaw 1964), 2n = 44 (Moffett and Hurcombe 1949) and 2n = 54 (Jauhar and Joshi 1968, Kumatsu and Suzuki 1987). However, different accessions varied widely in growth form and morphology. This variability has been considered normal for species which are normally sexual (Brown and Emery 1958, Hutchinson and Bashaw 1964) and cross pollinated (Codd 1939).

The chromosomes of the tetraploid accession (Fig. 6) with one pair satellited, are almost one and half times as long as those of the diploid one. However, the diploid accession has shown three accessory fragment chromosomes (Fig. 7). The karyotypes of both accessions are asymmetrical in terms of both chromosome lengths and arm ratios as indicated by the value of S.E. (Table 1). The study of chromosome association during first metaphase of meiosis revealed frequencies of bivalents (6.7/cell for diploid and 14.6/cell for tetraploid) that were lower than those of species with comparable chromosome number in the present study. This may suggest that many intra-chromosomal changes, especially paracentric inverions localized within arms, occur in both accessions and may have no physiological effect or adaptive significance.

P. maximum (Fig.8): It was the only species that exhibited the basic number x = 8 in the tetraploid condition with 2n = 32. This and many other numbers have been reported in *P. maximum* by various authors. The numbers 2n = 18 have been reported by de Wet (1954) and Raman *et al.* (1959), 2n = 32 by Chen and Hsu (1961), Jauhar (1967), Christopher and Abraham (1976) and Savidan (1980), 2n = 44 by Moffett and Hurcombe (1949), 2n = 48 by Warmke (1951). These counts may indicate that *P. maximum* is dibasic with x = 8 and 9. The basic number x = 8 has not been reported in any other species of *Panicum* so far. Therefore, the number 2n = 32 could have derived from 2n = 18 through sequential steps involving chromosomal changes that prevent normal pairing of homologous chromosomes causing univalent formation. These univalents could have been misdirected towards poles resulting in aneuploid production. Hence two double monosomics (2n = 18 – 1 – 1) or duplicated double monosomics could produce 2n = 32. Therefore, the basic number x = 8 might have been derived from x = 9. The number 2n = 44 reported earlier could be considered aneuploid also on basis of x = 9 since it is now known that these apomictic species, with cyclic ploidy, shows a good deal of causual aneuploidy.

The karyotype of the investigated sample of *P. maximum* is comprised of meta- and submeta-centric chromosomes, two pairs of which are satellited. The

chromosomes are obviously short and show little variation in length and arm ratio within this symmetrical karyotype.

P. virgatum (Fig. 9): Cytological investigation of this species revealed that it has a tetraploid chromosome number of 2n = 36. This, as well as many other numbers have been reported in this species. The numbers 2n = 36 and 72 were recorded by Burton (1942), 2n = 18, 36, 54, 72, 90 and 108 were reported by Nielsen (1944), and 2n = 21, 25, 30 and 32 were mentioned by Brown (1948). All these numbers could be considered either euploids or aneuploids that have originated from accessions with x = 9.

The karyotype of this species is comprised mainly of meta-and submeta-centic chromosomes. It showed the highest mean chromosome length (1.96 ± 0.22 μm) among all species investigated in the present study. The high value of S. E. of mean length indicates a considerable variation in length of chromosomes within the karyotype. In addition, some variability in centromere position is observed.

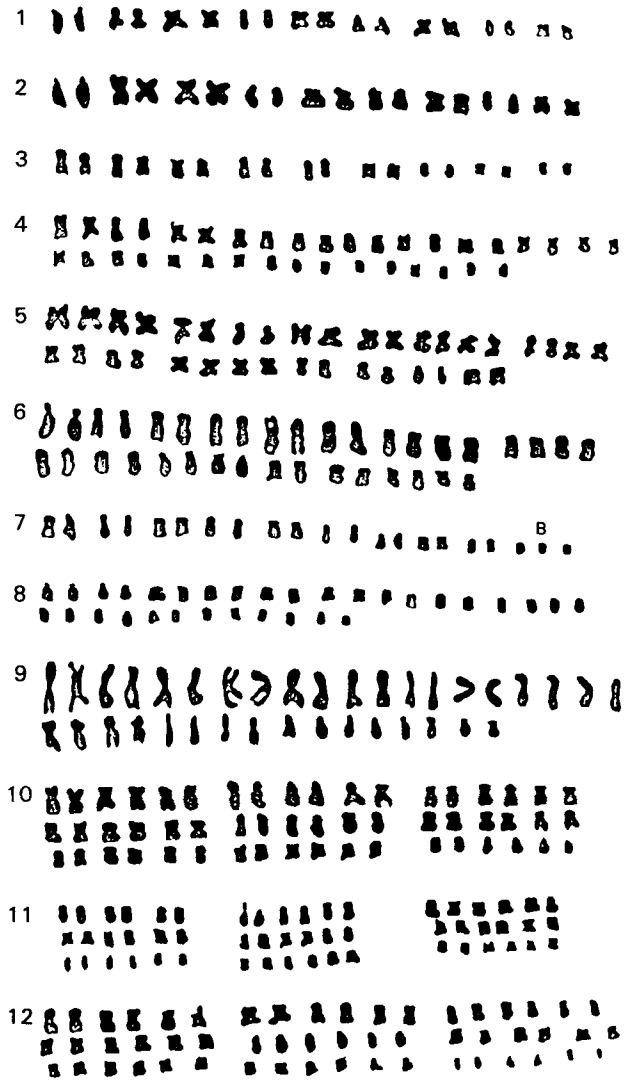
P. bulbosum (Fig. 10): This species exhibited the hexaploid number of 2n = 54. This chromosome number has been reported also in this species by Brown (1951) and Singh and Godward (1960). These authors also recorded 2n = 72 whereas 2n = 70 was recorded by the latter authors. However, tetraploid number of 2n = 36 was reported earlier by Krishnaswamy (1940). These findings indicate that the basic number for this species is also x = 9 which occasionally persists in an aneuploid form.

The karyotype of this species is symmetrical and consists of chromosomes which are relatively thick and have median and submedian centromeres. The highest frequency of chromosomes association during metaphase I of meiosis is recorded for bivalents (18.85/cell). Univalents (3.85/cell), trivalents (1.28/cell) and quadrivlaents (2.28/cell) were also encountered. These results point to the segmental alloploid origin of this species with some similarity existing among the genomes forming its karyotype. It also explains the occasional existance of aneuploid forms.

P. dichotomiflorum (Fig. 11): The chromosome number of this accession also has been found 2n = 54. This number was previously recorded by Church (1929) and Gould (1958). Tetraploid number of 2n = 36 was also recorded by Brown (1948).

This species exhibited the lowest mean chromosome length (0.87 + 0.05 μm) amongest all the investigated taxa of *Panicum*. Its karyotype is symmetrical and composed of metacentric chromosomes. The high frequency of bivalents (24.2/cell) indicates that it is allohexaploid. However, the very low proportion of trivalents (0.95/cell) and quadrivalents (0.85/cell) indicates low similarity to be existing among its genomes.

P. esculentum (Fig. 12): The hexaploid chromosome number of 2n = 54 has been found also in this species. This number has been reported by Krishnaswamy (1940), who also recorded 2n = 58. The karyotype of this species is symmetrical and



Figs. 1-12: Karyotypes of different species of *Panicum* L.
1 - *P. capillare*, 2 - *P. laevifolium*, 3 - *P. antidotale*, 4 - *P. miliaceum*, 5 - *P. miliare*, 6 - *P. coloratum 1*, 7 - *P. coloratum* 2, 8 - *P. maximum*, 9 - *P. virgatum*, 10 - *P. bulbosum*, 11 - *P dichotomiflorum*, and 12 - *P. esculentum.*

comprised of metacentric, morphologically similar, short chromosomes. Although this presumes an autoploid origin for this species, the frequency of bivalent formation during meiosis has been found 19.55/cell and that of univalents has been found 6.22/cell, being the highest among all the investigated species. These results suggest segmental alloploidy as the process by which this species was originted. In addition, it indicates high interchromosomal dissimilarity within the karyotype which may have resulted from paracentric inversions.

References

Avdulov, N.P.: Systematic Karyology of the Family Gramineae. - Proc. All-Russ. bot. Kongr. Pp. 65-67. 1928.

Avdulov, N.P.: Karyo-systematische Untersuchung der Familie Gramineen. - Bull. appl. Bot. Plant. Breed. **44** (Suppl.): 428, 1931.

Bolkhovskikh, Z., Grif, V., Matveeva, T., Zakharyeva, O.: Chromosome Number of a Flowering Plants. - Academy of Sciences of the USSR. V.L. Komarov Botanical Institute, Leningrad 1969.

Bouton, J.H., Brown, R.H., Bolton, I.K.: Photosynthesis of grass species differing in carbon dioxide fixation pathways. - Plant Physiol. **67**: 433-437, 1981.

Brown, W.L.: Cytological studies in Gramineae. - Amer. J. Bot. **35**: 382-396, 1948.

Brown, W.L.: Cytological studies of some Texas Gramineae. - Bull. Torrey bot. Club. **77**: 63-76, 1951.

Brown, W.L., Emery, W.H.P.: Apomixis in the Gramineae Panicoideae. - Amer. J. Bot. **45**: 253-263, 1958.

Burson, B.L., Bashaw, E.C.: Behaviour, transmission, and morphological effect of chromosomes in *P. coloratum*. - J. Crop Sci. **9**: 176-179, 1969.

Burton, G.W.: A cytological study of some species in the tribe Paniceae. - Amer. J. Bot. **29**: 355-361, 1942.

Chandola, R.P.: Cytogenetics of millets. - Cytologia **24**: 115-137, 1959.

Chen, C., Huss, C.: Cytological studies on Taiwan grasses I. tribe Paniceae. - Bot. Bull. Acad. Chin. Sci. **2**: 101-110, 1961.

Christopher, I, Abraham A.: Studies on the cytology and phylogeny of south Indian grasses III. Subfamily 6. Panicoideae tribe I. The Paniceae. - Cytologia **41**: 621-634, 1976.

Church, G.L.: Meiotic phenomena in certain Gramineae II. Paniceae and Andropoganeae. - Bot. Gaz. **88**: 63-84, 1929.

Codd, L.E.W.: Seed setting and strain building in grasses with special reference to makarikari grass. - Forming S. Afr. **14**: 277-279, 1939.

Darlington, C.D., Wylie, A.B.: Chromosome Atlas of Flowering Plants. 3rd. Ed. - George Allen and Unwin,. London 1955.

De Wet, J.M.J.: Chromosome numbers of a few South Africa grasses. - Cytologia **19**: 97-103, 1954.

Gould, F.W.: Chromosome number in southwestern grasses. - Amer. J. Bot. **45**: 757-768, 1958.

Gould, F.W.: Chromosome number in southwestern grasses II. - Amer. J. Bot. **47**: 873-877, 1960.

Hunter, A.W.S.: A karyosystematic investigation in the Gramineaee. - Can. J. Res. **11**: 213-224, 1934.

Hutchinson, D.G., Bashaw, E.C.: Cytology and reproduction of *P. coloratum* and related species. - Crop Sci. **4**: 151-153, 1964.

Jauhar, P.P.: Occurrence of accessory chromosomes in *P. maximum*. - Curr. Sci. **36**: 244-245, 1967.

Jauhar, P.P., Joshi, A.B.: Cytological studies in some species of *Panicum*. - Cytologia **31**: 153-159, 1965.

Jauhar, P.P., Joshi, A.B.: Accessory chromosomes in a new hexaploid species of *Panicum*. - Cytologia **21**: 105-110, 1968.

Jauhar, P.P., Joshi, A.B.: Cytotaxonomic investigation in the *P. maximum* complex III. - Cytologia **34**: 222-233. 1969.

Joshi, A.B., Patil, B.D., Manchanda, P.L.: Chromosome numbers in some grasses. - Curr. Sci. **28**: 454-455, 1959.

Krishnaswamy, N.: Untersuchungen zur Zytologie und Systematik der Gramineen. - Beih. bot. Centralbl. **60**: 1-50, 1940.

Kumatsu, T., Suzuki, S.: Chromosome numbers, behaviour and mode of reproduction in introduced strain of *Panicum* species. - J. jap. Soc. Grassl. Sci. **33**: 56-61, 1987.

Moffett, A.A., Hurcombe, R.: Chromosome numbers of South African grasses. - Heredity **3**: 369-373, 1949.

Narayan, K.N.: Apomixis in some species of *Pennisetum* and in *Panicum antidotale* in plant embrology. - A Symposium. Council of Scientific and Industrial Research. Pp. 55-61. New Delhi 1962.

Nath, J., Swaminathan, M.S.: Chromosome numbers of some grasses. - Indian J. Genet. Plant Breed. **17**: 120, 1957.

Nielsen, E.L.: Analysis of variation in *P. virgatum*. L. - J. agr. Res. **69**: 327-353, 1944.

Pienaar, R.V.: The chromosome number of some indigenous South African and introduced Gramineae - In: Meredith, D. (ed.): The Grasses and Pastures of South Africa. Pp. 551-570. Central News Agency, 1955.

Pritchard, A.J., Delacy, I.H.: The cytology, breeding system and flowering behaviour of *P. coloratum*. - Aust. J. Bot. **22**: 57-66, 1974.

Raman, V.S., Chandrasekharan, P., Krishnaswany, D.: Note on some chromosome numbers in Gramineae. - Curr. Sci. **28**: 127-128, 1959.

Rau, N.S.: Further contributions to the cytology of some crop plants of South India. - J. indian bot. Soc. **8**: 201-206, 1929.

Savidan, Y.: Chromosome and embryological analysis in sexual apomictic hybrids of *P. maximum*. - Theor. appl. Genet **57**: 153-156, 1980.

Singh, D.N., Godward, M.B.E.: Cytological Studies in the Gramineae. - Heredity **15**: 139-199, 1960.

Sokolovskaya, A.P., Probatova, N.S.: [Karyological investigation of grasses (Poaceae) in the south part of the Soviet Far-East.] - Bot. Zh. **62**: 1143-1153, 1972. [In Russ.]

Tateoka, T.: Contributions to biosystematic interactions of East African grasses. - Bull. nat. Sci. **8**: 161-173, 1965.

Warmke, H.E.: Cytotaxonomic investigations of some varieties of *P. maximum* and *P. purpurascens* in Puerto Rico. - Agron. J. **43**: 143-149, 1951.

Communicated by T. GICHNER