

Chromosomal Characters of Two *Cycas* Species (Cycadaceae, Cycadales)

Goro KOKUBUGATA¹, Ken D. HILL² and Katsuhiko KONDO³

國府方吾郎¹・Ken D. Hill²・近藤勝彦³：ソテツ属 2 種における染色体形質

The genus *Cycas* (Cycadaceae, Cycadales; Stevenson 1992) distributes in the Malaysian region, Japan and Southeast Asia, extending to Micronesia, Polynesia, Madagascar and East Africa, and is the largest in the Cycadales (Stevenson *et al.* 1995). Although taxonomical studies of *Cycas* have been advancing (*e.g.*, Hill 1992, 1994a, 1994b, 1996), its chromosomal studies have been poorly performed. The reason for the few accumulations of cytotaxonomic information in *Cycas* is that all species of the genus are regarded as endangered species in the world, and that it is difficult to observe too large chromosomes of *Cycas*. The aim of the present study is to investigate chromosome numbers and karyotypes of *C. ophiolitica* and *C. wadei*, which have been unreported cytotaxonomically, by the standard aceto-orcein staining method.

Materials and Methods

Cycas ophiolitica K. D. Hill and *C. wadei* Merrill cultivated in the experiment nursery of Tsukuba Botanical Garden were investigated in the present study (Table 1). Young leaflets were pretreated in 2 mM 8-hydroxyquinoline at 4°C for 20 hr, fixed in acetic ethanol (1:3) at 4°C for 20 hr, and stored in 70% ethanol at –20°C. The stored leaflets were macerated in 45% acetic acid at 60°C for 5 min and then stained in 2% aceto-orcein at 20°C for 12 hr. After being squashed with 45% acetic acid, the stained chromosomes at mitotic metaphase were classified with their arm ratio following Levan *et al.* (1964).

Results and Discussion

Cycas ophiolitica and *C. wadei* showed a chromosome number of $2n = 22$ (Fig. 1). The chromosome number of the two species is for the first time. The chromosome number of the two species corresponded with those of other *Cycas* species in the earlier reports (*e.g.*, Sax and Beal 1934, Kokubugata and Kondo 1994, Kondo *et al.* 1995). Chromosome lengths of *C. ophiolitica* ranged from 14.1 to 28.8 μm , and that of *C. wadei* from 16.6 to 32.9 μm .

Two species showed four subterminal-centromeric chromosomes from the 1st to the 4th chromo-

¹Tsukuba Botanical Garden, National Science Museum, Tsukuba, 305-0005. 国立科学博物館 筑波研究資料センター 筑波実験植物園.

²National Herbarium of New South Wales, Royal Botanic Gardens, Sydney. Mrs Macquaries Road, Sydney, NSW, 2000 Australia.

³Laboratory of Plant Chromosome and Gene Stock, Graduate School of Science, Hiroshima University, Kagamiyama, Higashi-Hiroshima, 739-8526. 〒739-8526 東広島市鏡山 1-4-3. 広島大学大学院理学研究科附属植物遺伝子保管実験施設.

Table. 1. Plant materials of two *Cycas* species observed in the present study

Species	Accession no.*	Origin	Chrom. no. (2n)
<i>C. ophiolitica</i>	TBG 136405	Australia: Rockhampton: Mt. Archer. 120 m alt., Queensland.	22
<i>C. wadei</i>	TBG 136391	The Philippines: Culion Island.	22

*TBG: Accession number of Tsukuba Botanical Garden.

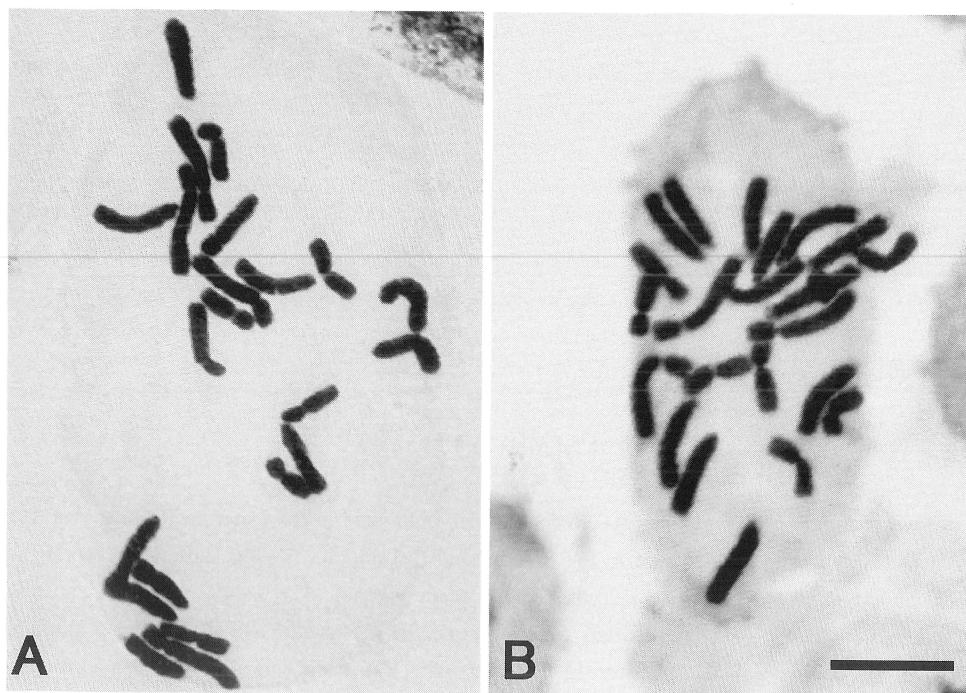


Fig. 1. Chromosome at mitotic metaphase of two *Cycas* species studied. A. *C. ophiolitica*. B. *C. wadei*. Bar shows 10 μm .

somes, twelve terminal-chromosomes from the 5th to the 16th, and four submedian- and two median-centromeric chromosomes from the 17th to the 22nd (Fig. 2). In conclusion, the present karyotypic data of two species were similar to each other and those of other *Cycas* species in the previous reports (e.g., Sax and Beal 1934, Kokubugata and Kondo 1994, Kondo *et al.* 1995).

Summary

Somatic chromosomes of *Cycas ophiolitica* and *C. wadei* were observed by the standard aceto-orcein staining method. Two species showed chromosome number of $2n = 22$, and karyotype consist of two median-, four submedian-, four subterminal- and twelve terminal-centromeric chromosomes. The karyotypes of the two species were similar to those of other *Cycas* species reported previously.

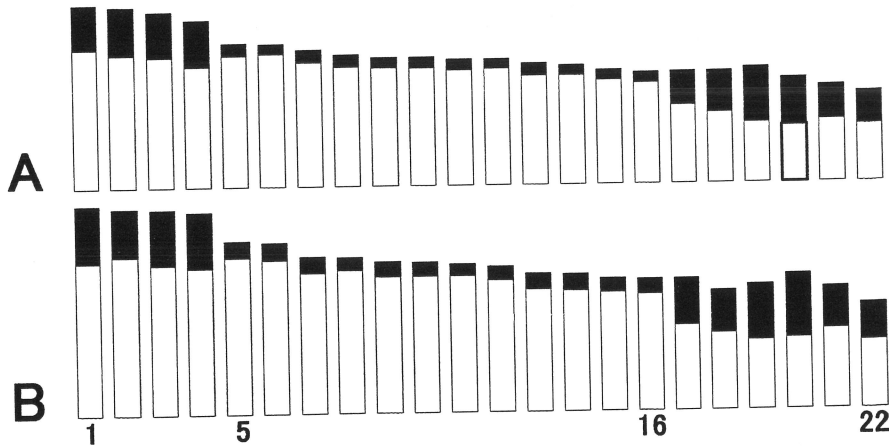


Fig. 2. Ideogram of two *Cycas* species studied. A. *C. ophiolitica*. B. *C. wadei*. Opened areas show long arm. Solid areas show short arm.

摘 要

ソテツ属（ソテツ科，ソテツ目）のこれまで染色体学的研究のなされていない2種，*Cycas ophiolitica* と *C. wadei* の2種の染色体と核型をアセトオルセイン染色法により観察した。2種ともに染色体数 $2n = 22$ を示し，核型は，2個の中部動原体型染色体，4個の長い次端部動原体型染色体，4個の次中部動原体型染色体，12個の端部動原体型染色体から構成されていた。これら2種の染色体数は過去の報告と一致し，核型は過去の報告におけるソテツ属他種の核型と類似していた。

Acknowledgements

The present study was financially supported in part by the Young Scientists Program B (No. 14740474; Representative: Goro Kokubugata) of Ministry of Education, Culture, Sports, Sciences and Technology, Japan.

References

- Hill, K. D., 1992. A preliminary account of *Cycas* (Cycadaceae) in Queensland. *Telopea* **5**: 177–206.
- , 1994a. The *Cycas media* group (Cycadaceae) in New Guinea. *Austral. Syst. Bot.* **7**: 527–541.
- , 1994b. The *Cycas rumphii* complex (Cycadaceae) in New Guinea and the Western Pacific. *Austral. Syst. Bot.* **7**: 543–567.
- , 1996. A taxonomic revision of the genus *Cycas* (Cycadaceae) in Australia. *Telopea* **7**: 1–64.
- Kokubugata, G. and K. Kondo, 1994. Quantitative variability in karyotype of *Cycas revoluta*. *La Kromosomo* **II-75-76**: 2613–2618.
- Kondo, K., G. Kokubugata, M. Hizume, R. Tanaka and T. Satake, 1995. A karyomorphological study of five species and one variety of *Cycas*. *Cytologia* **60**: 141–147.
- Levan, A., K. Fredga and A. A. Sandberg, 1964. Nomenclature for centromeric position on chromosomes. *Hereditas* **52**: 201–220.
- Sax, K. and J. M. Beal, 1934. Chromosomes of the Cycadales. *J. Arnold Arb.* **15**: 255–258.
- Stevenson, D. W., 1992. A formal classification of the extant cycads. *Brittonia* **44**: 220–223.
- , R. Osborne and K. D. Hill, 1995. The world list of cycads. In: Vorster P. (ed.), *Proc. 3rd Intl. Conf. on Cycad Biology*, Pretoria, South Africa. pp. 55–64.