

A Cytotaxonomic Study of Five Species of *Impatiens* (Balsaminaceae) in Java and Borneo, Malesia

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Somatic chromosome numbers of five species belonging to the genus *Impatiens* (Balsaminaceae) collected from Java and Borneo, Malesia are reported. *Impatiens chonoceras* Hassk. was found to be $2n = 14$, *I. javensis* Steud., *I. platypetala* Lindl., and *I. radicans* Zoll. were $2n = 16$, and *I. kinabaluensis* S. Akiyama & H. Ohba was $2n = 12$. Chromosome numbers of *I. chonoceras*, *I. javensis*, *I. radicans*, and *I. kinabaluensis* were examined for the first time. Karyotypes of *I. javensis*, *I. platypetala*, and *I. radicans* are similar and are thought to be closely related to each other. Taxonomical notes for each species are given.

Key words: Chromosome, cytotaxonomy, *Impatiens*, karyomorphology, Malesia.

Impatiens (Balsaminaceae) consists of 450 to 850 species distributed in tropical and temperate regions in the northern hemisphere (Grey-Wilson 1980, Cronquist 1981). Southeast Asia including Malesian region is one of the centers of diversity for *Impatiens* (Grey-Wilson 1980).

Chromosome numbers of *Impatiens* have been examined by many authors and various somatic chromosome numbers, $2n = 6, 8, 12, 14, 15, 16, 17, 18, 19, 20, 26, 28, 32, 34, 36, 40, 44, 48, 50, 56, \text{ca. } 60$, have been reported (see Federov 1969, Goldblatt 1981, 1984, 1985, 1988, Goldblatt and Johnson 1990, 1991, 1994, 1996, 1998, 2000, 2003). Thus

polyploidy and aneuploidy with different basic chromosome numbers might play an important role in the speciation of *Impatiens*.

Karyological studies have been made for species of *Impatiens* in several regions. For example, India (Govindarajan and Subramanian 1986, Rao et al. 1986), the Himalayas (Akiyama et al. 1992), and Southwest China (Sugawara et al. 1994, 1997). For Southeast Asian species of *Impatiens*, although several chromosome counts have been reported (Jones and Smith 1966, Arisumi 1973, 1987, Shimizu 1979, Larsen 1981, Okada 1989), no karyological study has been done.

Table 1. Species examined, localities and chromosome numbers of five species of *Impatiens*

Species	Localities (voucher specimens and their deposition)	Chromosome number (2n)
<i>I. chonoceras</i> Hassk.	Indonesia. West Java (Jawa Barat); Mt. (Gn.) Papandajan, 1950–2470 m alt. (Tsukaya & al. 104, 9 July 2003, TNS, BO)	14*
<i>I. javensis</i> Steud.	Indonesia. West Java (Jawa Barat): Gn. Gede-Pangorango National Park, 1840 m alt. (Tsukaya & al. 235, 6 March 2003, TNS BO)	16*
<i>I. platypetala</i> Lindl.	Indonesia. West Java (Jawa Barat); Gn. Gede-Pangorango National Park, 2120 & 1840 m alt. (Tsukaya & al. 259, 260, 8 March 2003, TNS, BO)	16
<i>I. radicans</i> Zoll.	Indonesia. West Java (Jawa Barat): Gn. Gede-Pangorango National Park, Gn. Gede/Gn. Pangorango, 2400 m alt. (Tsukaya & al. 240, 7 March 2003, TNS, BO)	16*
<i>I. kinabaluensis</i> S. Akiyama & H. Ohba	Malaysia. Sabah (Borneo Island); Kinabalu National Park, Mt. Kinabalu, Timpohon Gate–Layang-Layang, 1770 m alt. (Tsukaya & al. 0403201, 14 March 2004, TNS, BORH**)	12*

*New count for the taxon.

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This paper reports the chromosome numbers and karyotypes of five species of *Impatiens* collected from Java and Borneo, Malesia.

Materials and Methods

Materials were collected from their native habitat in West Java (Indonesia) and Mt. Kinabalu, Borneo (Sabah, Malaysia) in 2003 and 2004. Localities and voucher specimens are listed in Table 1.

For observation of somatic metaphase chromosomes, root tips were pretreated in the field with a 2 mM 8-hydroxyquinoline solution for about 3 hours and fixed with Newcomer's fluid (see Sharma and Sharma 1980, Wakabayashi 1988). The root tips were stained by Feulgen's nuclear reaction, and macerated in a mixture of 2 % pectinase and 2 % cellulase for 1.5–2 hours. They were again stained with 2 % lacto-propionic orcein and then squashed and observed using a light microscope.

Results and Discussion

Chromosome number of *I. chonoceras*

was $2n = 14$, *I. javensis*, *I. platypetala*, and *I. radicans* were $2n = 16$, and *I. kinabaluensis* was $2n = 12$, respectively (Table 1). Among them, chromosome numbers of *I. chonoceras*, *I. javensis*, *I. radicans*, and *I. kinabaluensis* were counted for the first time.

1. *Impatiens chonoceras* Hassk. (Fig. 1a)

The somatic chromosome number of *I. chonoceras* was $2n = 14$. This number is considered to be diploid, with basic chromosome number $x = 7$. Chromosomes are 1.5–3.1 μm long. Karyologically, the complement is monomodal, gradually reducing in size. Satellites were observed in two pairs of chromosomes: in the longest and second longest pairs.

Akiyama et al. (1992) reported chromosome numbers of 16 species in the Himalayas and reported those of *I. falcifer* and *I. serrata* as $2n = 14$. Sugawara et al. (1997) reported 15 species of *Impatiens* in Yunnan, SW China and chromosome numbers of *I. kamtilongensis* Toppin and *I. wuchengyihii* S. Akiyama, H. Ohba & S. K. Wu as $2n = 14$. All the species mentioned

above are similar in having characteristic floral morphology; the shape of the lower sepal is navicular tapering to the spur and the inflorescence usually has one or two flowers (Akiyama et al. 1991, 1996). Morphologically *I. chonoceras* is similar to these species. *Impatiens chonoceras* has a lower sepal with pubescence on the outer surface, like *I. kamtilongensis* and *I. wuchengyihii*, while the lower sepals of *I. falcifer* and *I. serrata* are glabrous on the outer surface. The inflorescence of *I. chonoceras* is axillary and pedunculate, and usually consists of two flowers with a basal bract. These two flowers appear in opposite. The inflorescences of *I. kamtilongensis* and *I. wuchengyihii* are racemose with two flowers (Akiyama et al. 1996), while those of *I. falcifer* and *I. serrata* are unique and have a middle bract (Akiyama and Ohba 2000). It is notable that *I. chonoceras* has the apparently most primitive form of inflorescence among Chinese species (i. e., *I. kamtilongensis* and *I. wuchengyihii*) and Himalayan species (i. e., *I. falcifer* and *I. serrata*).

Cytologically, all the species mentioned above have similar karyotypes. *Impatiens chonoceras*, *I. kamtilongensis*, and *I. wuchengyihii* have somatic chromosome numbers $2n = 14$, with monomodal complements. *Impatiens falcifer* and *I. serrata* also have somatic metaphase chromosomes $2n = 14$, and they show monomodal complements as judged from the plates (Akiyama et al. 1992).

Impatiens chonoceras was collected in Mt. Papandajan, a famous volcano located in SW Java. From the morphology and cytology, *I. chonoceras* is considered to be closely related to the species of the Himalayas and adjacent regions. *Potentilla polyphylla* Wall. ex Lehm. (Rosaceae), distributed mainly in the Himalayas and adjacent regions, was collected in this mountain (Kalkman 1968, 1993), suggesting that Mt. Papandajan had been a refugium for Himalayan plants in this

area.

2. *Impatiens javensis* Steud., *I. platypetala* Lindl. and *I. radicans* Zoll. (Figs. 1b–e)

The somatic chromosome numbers of *I. javensis*, *I. platypetala*, and *I. radicans* were $2n = 16$. These numbers are considered to be diploid, with basic chromosome number $x = 8$. Chromosomes are 0.9–1.8 μm long in *I. javensis*, 1.0–1.5 μm in *I. platypetala*, and 1.1–1.7 μm in *I. radicans*. Karyomorphologically they showed similar karyotypes. The complements were monomodal, gradually reducing in size. Satellites were observed in one pair of chromosomes.

Van Steenis (1948) recognized three subspecies in *I. platypetala*: subsps. *platypetala*, *aurantiaca*, and *nematoceras*. Among the subspecies, subsp. *platypetala* has the widest distribution range, throughout Java and Sumatra. Zinov'eva-Stahevitch and Grant (1984) reported chromosome numbers of *I. platypetala* subsp. *platypetala* as $n = 7$ and $2n = 14$ (from Mt. Gedé, Java), subsp. *aurantiaca* $2n = 14$ (from Sulawesi), and subsp. *nematoceras* $n = 8$ and $2n = 16$ (from Indonesia). Arisumi (1987) reported the chromosome number of *I. platypetala* (subsp.) *aurantiaca* as $2n = 8$. Okada (1989) counted chromosome numbers of *I. platypetala* from Sumatra, which may be subsp. *platypetala* based on distribution, as $2n = 16$. Our count for *I. platypetala*, refers to subsp. *platypetala*, coincides with the number counted by Okada (1989), but differs from the number given by Zinov'eva-Stahevitch and Grant (1984) for a collection from the same locality. For confirming the chromosome numbers of the species and the infraspecific taxa of *I. platypetala*, examination of ample materials collected from the entire range of distribution is necessary.

Impatiens platypetala, *I. javensis*, and *I. radicans* are closely related species judging from floral morphology and karyotypes. In Gunung (Mt.) Gede-Pangrango National

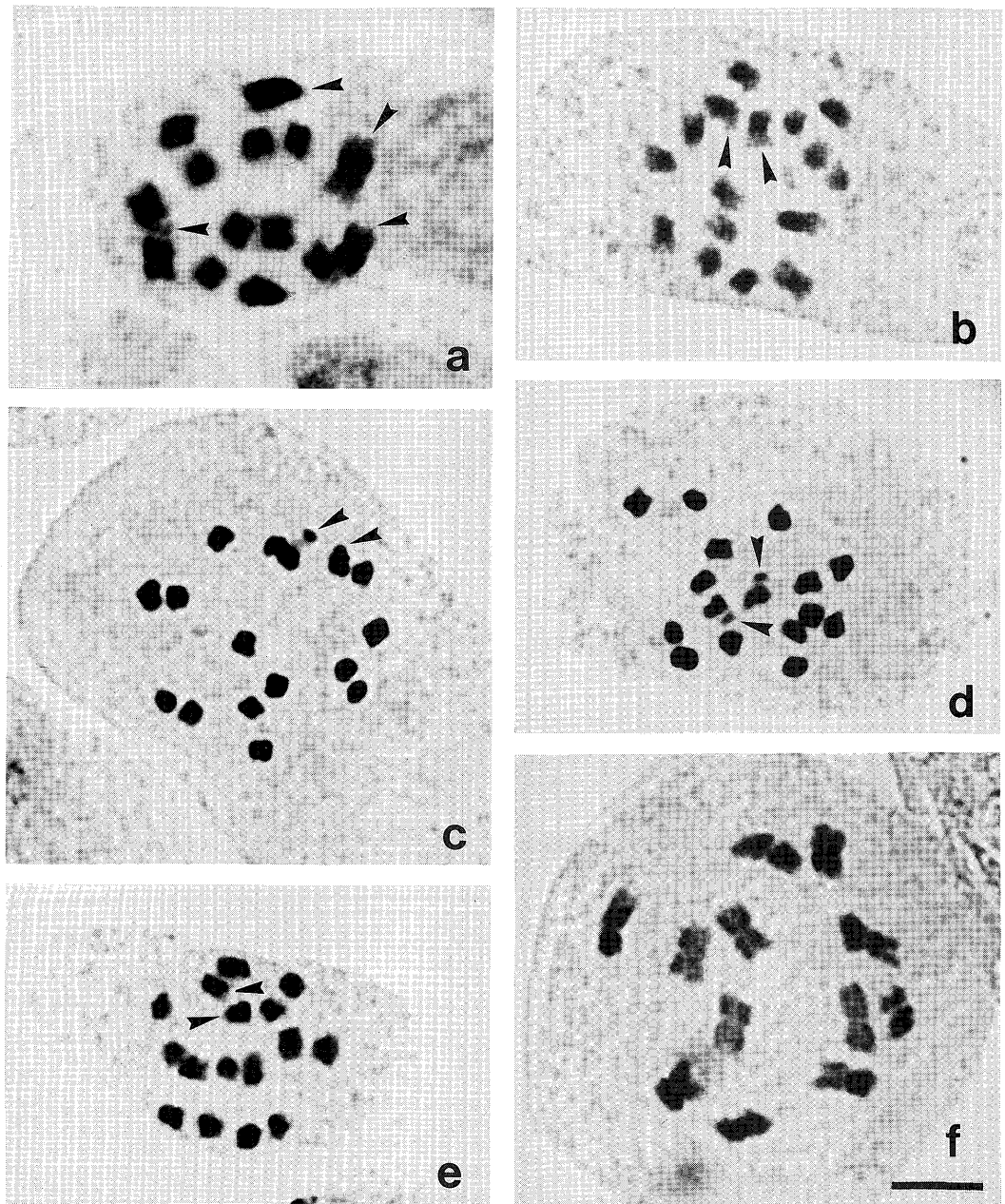


Fig. 1. Somatic metaphase chromosomes of five species of *Impatiens* in SE Asia. a: *I. chonoceras* ($2n = 14$). b: *I. javensis* ($2n = 16$). c, d: *I. platypetala* ($2n = 16$). e: *I. radicans* ($2n = 16$). f: *I. kinabaluensis* ($2n = 12$). Arrows indicate satellite chromosomes. Bar indicates $5 \mu\text{m}$.

Park, they inhabit different altitudes: *I. platypetala* at the lowest altitudes, less than 1800 m, *I. javensis* between 1800 m and 2400 m, and *I. radicans* above 2400 m. *Impatiens platypetala* is distributed widely in Malesia while *I. javensis* is found in Java and probably in Sumatra, and *I. radicans* is endemic to West Java (as far as known, from Mts. Pangrango, Tjikurai and Papandajan) (van Steenis 1972). *Impatiens javensis* and *I. radicans* are genetically close and it is known that they produce interspecific hybrid (Tsukaya 2004). It may be thought that *I. javensis* and *I. radicans* might be differentiated in diploid level after altitudinal isolation from an *I. platypetala*-like ancestor having 16 somatic chromosomes in Java Island.

3. *Impatiens kinabaluensis* S. Akiyama & H. Ohba (Fig. 1f)

The somatic chromosome number of *I. kinabaluensis* was $2n = 12$. The number is considered to be diploid, with basic chromosome number $x = 6$. Chromosomes are 2.4–3.8 μm long. Karyologically, the complement is monomodal, gradually reduced in size. Satellites were not observed.

Chromosome numbers of *Impatiens* in Southeast Asia including Malesia have been fragmentarily reported (Jones and Smith 1966, Arisumi 1973, 1987, Shimizu 1979, Larsen 1981, Zinov'eva-Stahevitch and Grant 1984, Okada 1989), with those of about 20 species so far revealed. Among these species, two species were reported as $2n = 12$; *I. chiangdaoensis* T. Shimizu from Thailand (Shimizu 1979, Larsen 1981) and "*I. harlandii* Dransfield ined." from Borneo (Zinov'eva-Stahevitch and Grant 1984) although we could not find any literature on "*I. harlandii*", and could not determine whether this is the same species as *I. kinabaluensis* or not.

Van Steenis (1948) noted the distribution of *I. platypetala* subsp. *platypetala* as "throughout Java, further in Sumatra and

probably also occurs in Borneo, Sulawesi and the Lesser Sunda Islands". Masamune (1942) listed *I. platypetala* among Bornean *Impatiens*.

After examining the morphology of *Impatiens* collected in Mt. Kinabalu, Borneo (Tsukaya & al. 0403201), it is concluded that this is not *I. platypetala*, but a new species, named *I. kinabaluensis* (Akiyama et al. 2005)

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ツリフネソウ属植物 (ツリフネソウ科) の細胞分
類学的研究

ジャワ島 (インドネシア) およびボルネオ島キ
ナバル山 (マレーシア) で採集されたツリフネソ
ウ科ツリフネソウ属 5 種の染色体数と核型を報告
した。

Impatiens chonoceras Hassk. は $2n = 14$, *I. javensis*
Steud., *I. platypetala* Lindl., *I. radicans* Zoll. は $2n =$
 16 , *I. kinabaluensis* S. Akiyama & H. Ohba は $2n =$
 12 であった。 *Impatiens chonoceras*, *I. javensis*, *I.*
radicans, *I. kinabaluensis* の染色体数は, 今回が初
めての報告である。 *Impatiens chonoceras* は, 花や
花序の形態と核型から, ヒマラヤや中国南西部に
分布する種と類縁があると考えられた。 *Impatiens*
javensis, *I. platypetala*, *I. radicans* の 3 種は, 花の
形態と核型から互いに近縁と考えられ, 分布がジャ

ワ島付近に限られる *I. javensis* と *I. radicans* は,
 $2n = 16$ の染色体数を持つ *I. platypetala* 類似の祖先
種から垂直的な棲み分けにより分化したものでは
ないかと推定された。 *Impatiens kinabaluensis* は,
形態と核型から, これまで記載されていなかった
新種であると考えられた。

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