

increased, plant respiration increased. Considering the reduction in plant sizes, dry wt, or wounding in itself, severely damaged plants probably had increased respiration to compensate for injury. The anatomical study showed that by day 2 the process of wound healing (cell division) had begun. The respiration needed to initiate wound healing was not measurable in this experiment. Todd and Saren (6, 13) reported that increased respiration rates in plants exposed to wind treatments decreased rapidly after exposure.

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Hybridization Among Diploid and Tetraploid Forms of New Guinea, Java, and Celebes *Impatiens* spp.¹

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Abstract. Interspecific hybrids were synthesized by intercrossing diploid and tetraploid of *Impatiens* spp. from New Guinea ($2x=32$), Java ($2x=16$), and Celebes ($2x=8$). Breeding analysis of the intra- and interploidy crosses showed that the species functioned as diploids in relation to each other. Diploid by tetraploid crosses produced little or no seed, and with few exceptions the triploid seedlings were sterile. All but 2 out of 43 selfs and reciprocal crosses among tetraploids produced viable seed. Amphidiploid and autotetraploid seedlings were pollen and seed fertile. Allotetraploids composed of 2 or 3 genomes in various unequal combinations and those with 4 genomes were pollen and seed sterile or nearly so. Most of the 3 and 4 allopolyploids were ornamentally inferior to the diploid and amphidiploid hybrids. New Guinea flower colors were recessive to those of Java and Celebes. The hybrid phenotypes were midparental for most traits and in some cases exhibited dosage effects. Some hybrids were superior to both parents in certain ornamentally important attributes.

Breeding experiments have shown that most New Guinea *Impatiens* spp. will hybridize among themselves to form fertile F₁ progenies, but sterile F₁ progenies occur in crosses between New Guinea and Java or Celebes spp. (2, 3). Exceptions to this have also been reported (12, 14). Crosses using natural diploids and artificial polyploids indicated that intraploidy crosses, i.e., $2x \times 2x$ or $4x \times 4x$, regardless of differences in base chromosome numbers were productive, but that interploidy crosses such as New Guinea ($2x=32$) \times Java ($4x=32$) and Java ($2x=16$) \times Celebes ($4x=16$) did not set seed (2). The implications of these initial results for plant breeding and evolutionary means for speciation in *Impatiens* prompted the extensive breeding tests

reported here.

In this study I conducted certain observations on 1) the viability and fertility of various genomic combinations among the hybrids, 2) the hybrid phenotypes, and 3) the use of interspecific hybrids in breeding programs.

Materials and Methods

Parental plants (Table 1) comprised a representative sample of the available species, hybrids, and unidentified plant introductions from New Guinea, Java, and Celebes (15). Except for N12, a seed sterile plant (1), and 'Tangerine', a generally poor seed parent, the parental plants were chosen from self- and cross-compatible types. This restriction to fertile types was necessary to reduce the chances of seed failure from specific incompatibility or sterility genes rather than from genome interactions. No substitutes for N12 or 'Tangerine' were available. Genomic identification follow those used previously (4);

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Table 1. Parental *Impatiens* spp. and hybrids.

Reference no.	Accession no.	Plant Intro. no.	Species and hybrids	Genomic formula ^z	Somatic chrom. no.	Origin
1	C26	366029	unidentified	CC	8	Celebes
2	C35		<i>I. platypetala aurantiaca</i> 'Tangerine' Steen	TT	8	"
3	J21C	349629C	<i>I. platypetala</i> Lindl	JJ	16	Java
4	J21D	349629D	" " "	JJ	16	"
5	N1A	349582A	unidentified	NN	32	New Guinea
6	N3	349586	"	NN	32	"
7	N3A	349586A	"	NN	32	"
8	N4	349588	"	NN	32	"
9	N4A	349588A	"	NN	32	"
10	N4C	349588C	"	NN	32	"
11	N5	354254	<i>I. schlechteri</i> Warb	NN	32	"
12	N6	354255	unidentified	NN	32	"
13	N12	354265	<i>I. schlechteri</i> Warb	NN	32	"
14	N13	349585	unidentified	NNNN	64	"
15	N15C	354252C	<i>I. mooreana</i> Schlect	NN	32	"
16	N15I	354252I	" " "	NN	32	"
17	N15K	354252K	" " "	NN	32	"
18	N15M	354252M	" " "	NN	32	"
19	N16	354253	<i>I. schlechteri</i> Warb	NN	32	"
20	N16D	354253D	" " "	NN	32	"
21	N17	349584	<i>I. linearifolia</i> Warb	NN	32	"
22	N33	367704	<i>I. herzogii</i> K. Schum	NN	32	"
23	N50		N6 × N16	NN	32	Lab. ^y
24	N51		N15 × N12	NN	32	"
25	N52		N3A × N5	NN	32	"
26	N53		N15C × N5	NN	32	"
27	N54		N19 × N15M	NN	32	"
28	N55		N4C × N16D	NN	32	"
29	N56		N3A × N12	NN	32	"
30	C26(4n)		colch. ^x C26	CCCC	16	"
31	C35(4n)		" C35	TTTT	16	"
32	J21C(4n)		" J21C	JJJJ	32	"
33	J21D(4n)		" J21D	JJJJ	32	"
34	N4A(4n)		" N4A	NNNN	64	"
35	N15I(4n)		" N15I	NNNN	64	"
36	N50(4n)		" N50	NNNN	64	"
37	N51(4n)		" N51	NNNN	64	"
38	N52(4n)		" N52	NNNN	64	"
39	N53(4n)		" N53	NNNN	64	"
40	N54(4n)		" N54	NNNN	64	"
41	N55(4n)		" N55	NNNN	64	"
42	N56(4n)		" N56	NNNN	64	"
43	CJ1(4n)		" (C26 × J21C)	CCJJ	24	"
44	CJ2(4n)		" (C26 × J21D)	CCJJ	24	"
45	CT1(4n)		" (C26 × C35)	CCTT	16	"
46	JN1(4n)		" (N1A × J21D)	JJNN	48	"
47	JN2(4n)		" (J21C × N5)	JJNN	48	"
48	JN3(4n)		" (N3A × J21d)	JJNN	48	"
49	NT1(4n)		" (N5 × C35)	NNTT	40	"
50	CJ1a(4n)		C26(4n) × J21C(4n)	CCJJ	24	"
51	CJ2a(4n)		C26(4n) × J21D(4n)	CCJJ	24	"
52	CN1a(4n)		N50(4n) × C26(4n)	CCNN	40	"
53	CN2a(4n)		N51(4n) × C26(4n)	CCNN	40	"
54	CN3a(4n)		C26(4n) × N50(4n)	CCNN	40	"
55	CT1a(4n)		C26(4n) × C35(4n)	CCTT	16	"
56	JN4a(4n)		N13 × J21C(4n)	JJNN	48	"
57	JN5a(4n)		N50(4n) × J21C(4n)	JJNN	48	"
58	JN6a(4n)		N51(4n) × J21C(4n)	JJNN	48	"
59	NT2a(4n)		N50(4n) × C35(4n)	NNTT	40	"
60	NT3a(4n)		N51(4n) × C35(4n)	NNTT	40	"
61	NT4a(4n)		N5(4n) × C35(4n)	NNTT	40	"

^zWhere C, J, N, and T represent Celebes (C26), Java, New Guinea, and 'Tangerine' (C35) species, respectively.^yFlorist and Nursery Crops Laboratory.^xTetraploid derived from colchicine treatment.

Table 2. Results of crosses for triploidy among New Guinea, Java, and Celebes *Impatiens*.

Genomic formula of crosses ^z	Reciprocal crosses (no.)	Parental plants (Reference numbers from Table 1.)	Avg seed (no.) ^y	Progeny	
				Genomic formula	Breeding behavior
CCCC × CC	1	1, 30	16	CCC	ns ^x
CCCC × JJ	2	3, 4, 32	6	CCJ	ns
CCCC × NN	4	21, 22, 26, 29, 30	11	CCN	ns
CCCC × TT	1	2, 30	2	CCT	sterile
JJJJ × CC	2	1, 32, 33	0	CJJ	
JJJJ × JJ	3	3, 4, 32, 33	0	JJJ	
JJJJ × NN	10	5, 6, 7, 11, 16, 32, 33	0	JJN	
JJJJ × TT	2	2, 32, 33	0	JJT	
NNNN × CC	5	1, 14, 34, 36, 37, 39	0	CNN	
NNNN × JJ	6	3, 14, 35, 38, 40, 41	0	JNN	
NNNN × NN	10	8, 9, 12, 13, 40, 41, 42	0	NNN	
NNNN × TT	4	2, 14, 35, 38, 40	0	NNT	
TTTT × CC	1	1, 31	0	CTT	
TTTT × JJ	2	3, 4, 31	0	JTT	
TTTT × NN	3	11, 26, 28, 31	3	NTT	sterile
TTTT × TT	1	2, 31	0	TTT	
CCJJ × NN	7	11, 20, 26, 28, 43, 50	0	CJN	
CCJJ × TT	3	2, 43, 44, 51	0	CJT	
CCNN × JJ	6	3, 4, 52, 53, 54	0	CJN	
CCNN × TT	3	2, 52, 53, 54	0	CNT	
CCTT × JJ	4	3, 4, 45, 55	0	CJT	
CCTT × NN	6	10, 11, 13, 15, 17, 18, 45	0	CNT	
JJNN × CC	2	1, 46, 47	0	CJN	
JJNN × TT	2	2, 46, 47	1	JNT	sterile
NNTT × CC	2	1, 49, 60	0	CNT	
NNTT × JJ	6	3, 4, 49, 59, 60, 61	0	JNT	
CCJJ × CC	2	1, 50, 51	0	CCJ	
CCJJ × JJ	4	3, 4, 50, 51	0	CJJ	
CCNN × CC	2	1, 52, 53	0	CCN	
CCNN × NN	6	24, 25, 26, 52, 53	0	CNN	
CCTT × CC	2	1, 45, 55	0	CCT	
CCTT × TT	2	2, 45, 55	1	CTT	sterile
JJNN × JJ	7	3, 4, 46, 47, 56, 57	0	JJN	
JJNN × NN	10	8, 10, 17, 23, 25, 57, 58	2	JNN	sterile

^zWhere C, J, N, and T represent Celebes (C26), Java, New Guinea, and 'Tangerine' (C35) species, respectively.

^yAvg seed per capsule.

^xNearly sterile; < 10% of pollinated flowers yielding seed.

i.e., N for New Guinea, J for Java, C for C26 (Celebes), and T for 'Tangerine' (Celebes).

About 15-50 pollinations were made for each reciprocal cross. Those that failed after 50 pollinations were considered difficult and discontinued. Parental stock and progenies were grown in the greenhouse under natural illumination at about 23.9°C (day)/18.3°C (night) temp from fall to late spring. Summer temp were modified by heavy shading and cooling pads. Immature capsules from abortive and normal crosses were examined periodically for embryo and ovule development. Plants were considered hybrids by visual inspection for parental traits and sometimes by chromosome counts made on root tip smears prepared by standard acetocarmine procedures.

Fertility was tested by choosing 3 or 4 seedlings at random from each progeny and then selfing, sibbing, backcrossing, or crossing them with other species and hybrids. A progeny was considered fertile if seed were obtained by self- or cross-pollination, nearly sterile if seed were set in less than 10% of the self- or cross-pollinated flowers, and sterile if every plant tested failed to set seed as pollen or as seed parent. Plants from progenies considered sterile were tested over a year for possible seasonal effects on fertility.

Results

Viability and fertility of triploids. Interploidy crosses were difficult, producing 8 kinds of progeny out of 34 possible

types of cross (Table 2). Capsules usually abscised within 1-2 weeks after pollination, and the ovules containing aborted embryos rarely enlarged. Aborted embryos, 10-15 days old, were at the globular stage and less than 0.1 mm across. Aborted embryos in the heart stage were seldom observed.

Of the 8 viable types, 5 were sterile (Table 2). Because triploids produced little or no pollen, pollen from parental plants and other diploid or tetraploid species was used in breeding tests. Three progeny types were classified as nearly sterile. They produced 1-2 seed per capsule and seedlings were sterile or nearly so. No seasonal influence on pollen or seed sterility was observed among triploids.

Viability and fertility of tetraploids. Intraploidy crosses to produce tetraploids resulted in seedlings representing 30 different genomic combinations (Table 3). Out of 43 types of selfs and crosses, 2 failed because the embryos aborted 1-2 weeks after pollination. About 2 or 3 out of 20-25 seedlings per progeny did not survive to maturity. Many of these resembled the chlorotic and slow-growing forms found in diploid populations of New Guinea spp. caused by recessive lethal genes.

In general, self-pollinations produced less seed than crosses. Also selfs produced 5-20% more deficient seedlings than crosses. Few parents consistently yielded more seed as one or the other parent in reciprocal crosses. Tetraploid 'Tangerine' averaged 3-5 seed per capsule as seed parent and 2-5 times more as pollen

Table 3. Results of crosses and selfs for tetraploidy among New Guinea, Java, and Celebes *Impatiens* spp.

Genomic formula of crosses ^Z	Self & reciprocal crosses (no.)	Parental plants (Reference numbers from Table 1)	Avg seed (no.) ^Y	Progeny	
				Genomic formula	Breeding behavior
CCCC × CCCC	1	30	15	CCCC	fertile
CCCC × JJJJ	2	30, 32, 33	4-20	CCJJ	"
CCCC × NNNN	5	14, 30, 35, 38, 39, 40	5-22	CCNN	"
CCCC × TTTT	1	30, 31	5-10	CCTT	"
JJJJ × JJJJ	1	32, 33	5-18	JJJJ	"
JJJJ × NNNN	6	32, 33, 37, 39, 40	7-16	JJNN	"
JJJJ × TTTT	2	31, 32, 33	3-13	JJTT	"
NNNN × NNNN	12	14, 34, 35, 37, 41, 42	4-27	NNNN	"
NNNN × TTTT	5	14, 31, 37, 38, 39, 40	3-20	NNTT	"
TTTT × TTTT	1	31	3	TTTT	"
CCCC × CCJJ	2	30, 43, 50	6-25	CCCJ	sterile
CCCC × CCNN	3	30, 52, 53, 54	9-19	CCCN	"
CCCC × CCTT	2	30, 45, 55	8-17	CCCT	ns ^X
CCCC × JJNN	6	30, 46, 47, 48, 56, 57, 58	6-34	CCJN	sterile
CCCC × NNTT	4	30, 49, 59, 60, 61	1-5	CCNT	"
JJJJ × CCJJ	3	32, 33, 50, 51	5-7	CJJJ	"
JJJJ × CCNN	3	32, 33, 52, 53, 54	1-6	CJJN	"
JJJJ × CCTT	2	32, 33, 55	13-15	CJJT	"
JJJJ × JJNN	4	32, 33, 46, 47	7-31	JJJN	"
JJJJ × NNTT	2	32, 33, 59	8-20	JJNT	"
NNNN × CCJJ	3	39, 40, 43	5-12	CJNN	"
NNNN × CCNN	4	37, 38, 41, 42, 52	0	CNNN	"
NNNN × CCTT	4	37, 38, 39, 40, 45	7-15	CNNT	sterile
NNNN × JJNN	7	34, 36, 37, 39, 46, 47, 48	10-21	JNNN	"
NNNN × NNTT	10	34, 35, 37, 41, 42, 49, 59	3-29	NNNT	"
TTTT × CCTT	2	31, 45, 55	4-12	CTTT	ns
TTTT × JJNN	2	31, 47, 56	0	JNTT	"
TTTT × NNTT	2	31, 49, 60	3-16	NTTT	sterile
CCJJ × CCJJ	3	43, 44, 50	10-16	CCJJ	fertile
CCJJ × CCNN	2	43, 51, 52, 53	4-8	CCJN	sterile
CCJJ × CCTT	2	43, 44, 45	10-20	CCJT	"
CCJJ × JJNN	3	43, 44, 46, 47	7-12	CJJN	"
CCJJ × NNTT	3	43, 44, 49, 59	0-10	CJNT	"
CCNN × CCTT	3	45, 52, 53, 54	5-9	CCNT	"
CCNN × CCNN	3	52, 53, 54	6-16	CCNN	fertile
JJNN × CCNN	1	46, 53	0-5	CJNN	sterile
JJNN × CCTT	8	45, 47, 48, 55, 57, 58	11-14	CJNT	"
JJNN × JJNN	1	46, 47	8-24	JJNN	fertile
JJNN × NNTT	4	46, 47, 49, 60	8-21	JNNT	sterile
CCTT × CCTT	1	45, 55	5-18	CCTT	fertile
CCTT × JJNN	6	45, 46, 47, 48, 55	6-23	CJNT	sterile
NNTT × CCTT	2	45, 49, 59	3-12	CNTT	"
NNTT × NNTT	3	49, 59, 60	5-33	NNTT	fertile

^ZWhere C, J, N, and T, represent Celebes (C26), Java, New Guinea, and 'Tangerine' (C35) species, respectively.^YAvg seed per capsule.^XNearly sterile; < 10% of pollinated flowers yielding seed.

parent (Table 3). Tetraploid C26, or CCCC, usually produced 2 or 3 times more as seed parent.

Among tetraploid progenies, autotetraploids and amphidiploids were fertile (Table 3). Sterile and nearly sterile progenies consisted of 4 different genomes (e.g., CJNT), or of 2 or 3 different genomes in unequal combinations (Table 3). The nearly sterile progenies, CCCT and CTTT, rarely set seed, but few selfs and backcrosses produced 1-4 seed per capsule. Pollen of CCCJ, CCCN, JJNN, etc., resembled pollen of the nearly sterile CCCT and CTTT and frequently appeared to be normal, but never produced viable seed. What apparently was the most defective or abortive pollen was produced among the sterile forms CJNT. Tetraploids made up of 3 different genomes, such as CNNT, CNTT, etc., also produced mostly defective pollen but their anthers usually had more pollen than CJNT. No seasonal differences in fertility were shown by the sterile and nearly sterile forms.

Chromosome numbers. Root tip smears of 3-6 seedlings

each of 8 triploid and 5 tetraploid progenies showed no deviation from the expected chromosome numbers. The triploids CCC, CCT, and CTT had 12 chromosomes, CCJ had 16, CCN and NTT had 24, JNT had 28, and JNN had 40. The tetraploids NNTT had 28, CJNT had 32, CCNN and NNTT had 40, and CNNN had 52. What appeared to be cytochimera root tissues were observed in several root samples (2).

Hybrid phenotypes. Dominant genes for certain floral and foliar characteristics served as markers of genomes carrying them. In triploids and tetraploids, genes for orange flowers from 'Tangerine' were dominant in every hybrid (Table 4). The intensity of the orange color was increased with increasing T genomes, but TT hybrids were sometimes equal to those of TTT or TTTT. The bright yellow flowers of the Celebes spp. was dominant in hybrids without the T genome (Table 4). T and C genomes together produced bright orange flowers in their hybrids. In hybrids composed of only Java and New Guinea spp., the magenta or purplish flower color of the Java

Table 4. Phenotypes and characteristics of ornamental importance among various genomic combinations of New Guinea, Java, and Celebes *Impatiens* spp.

Genomic formula ^z	Floral type ^y			Foliage type ^y			Plant type ^y		Ornamental value ^x
	Color	Size	Shape	Color	Size	Shape	Form	Size	
CCCC	C	C	C	C	C	C	C	C	average
CCCJ	C	C	CJ	C	CJ	CJ	CJ	C	average
CCCN	C	CN	CN	C	CN	CN	CN	CN	average
CCCT	C	C	C	C	C	CT	C	C	average
CCJJ	C	CJ	CJ	J	J	CJ	CJ	CJ	good
CCNN	C	CN	CN	N	CN	CN	CN	CN	good
CCTT	CT	CT	CT	CT	CT	CT	CT	CT	good
CCJN	C	CJ	CJ	JN	JN	JN	JN	JN	average
CCNT	CT	CT	CN	CN	CN	CN	CT	CT	average
CCJT	CT	CT	CT	CJ	CJ	CJ	CJ	CJ	average
CJJJ	C	J	J	J	J	J	J	J	average
CJJN	C	JN	JN	JN	JN	JN	JN	JN	average
CJJT	C	CJ	CJ	CJ	CJ	CJ	CJ	CJ	average
CJNN	C	JN	JN	JN	JN	JN	JN	JN	average
CJNT	CT	JN	JN	JN	JN	JN	JN	JN	average
CNNT	CT	CN	CN	N	N	N	N	N	good
CNTT	T	NT	NT	NT	NT	NT	NT	NT	average
JJJJ	J	J	J	J	J	J	J	J	good
JJJN	J	J	J	JN	JN	JN	J	J	average
JJNN	J	JN	JN	JN	JN	JN	JN	JN	good
JJNT	T	JN	JN	JN	JN	JN	JN	JN	average
JJTT	T	JT	JT	JT	JT	JT	JT	JT	good
JNNN	JN	N	N	JN	N	N	JN	JN	average
JNNT	T	JN	JN	JN	JN	JN	JN	JN	average
NNNN	N	N	N	N	N	N	N	N	average
NNNT	T	N	N	N	N	N	N	N	average
NNTT	T	NT	NT	N	NT	NT	N	N	good
NTTT	T	NT	NT	T	T	T	T	T	poor
TTTT	T	T	T	T	T	T	T	T	good

^zWhere C, J, N, and T represent Celebes (C26), Java, New Guinea and 'Tangerine' (C35) species, respectively.

^yClosest resemblance to genome listed ; CN, CJ, NT, etc., refer to mid-parental appearance.

^xCompared to diploids having similar plant characteristics.

genome was dominant. A few NJ progenies had the flower colors of the New Guinea parent. These exceptional seedlings had pale magenta, nearly white, pink, and rose colored flowers. About 60% of the Java hybrids had 2 or 4 distinct purplish spots near the throat of the flowers (Fig. 1). Because this trait was present only in Java hybrids, it served as a marker for the Java spp. where T or C was involved. A marker for certain New

Guinea parents was the phenotype for variegated foliage (Fig. 1). Variegated New Guinea spp. transmitted this trait to 20-100% of its hybrids. The gene or genes for variegation were unstable in several seedlings and somatic mutants giving rise to non-variegated shoots in variegated plants or variegated shoots in nonvariegated plants were observed (Fig. 2).

Size and shape of plants, leaves, and flowers seemed to be regulated polygenically, and in most cases progeny phenotypes



Fig. 1. Seedling of New Guinea x Java, showing variegated foliage from N, and purplish floral spots from J.



Fig. 2. 'Sweet Sue' seedling (NNTT) showing several branches that had mutated to the non-variegated form.

were intermediate to parental phenotypes (Table 4). Although these characteristics were not as distinct as the dominant traits for establishment of hybridity, the parental contribution was evident in most seedlings (Table 4). In some, the parental traits were determined in hybrid progenies by comparison with selfed progenies. Selfed seedlings of tetraploid Java, Celebes, and 'Tangerine' were practically identical to their parents. With the exception of N13, a natural tetraploid, selfed New Guinea tetraploids segregated for various floral, foliar, and plant characteristics.

Ornamental traits. Triploid progenies were ornamentally inferior to their diploid and tetraploid parents. The best triploids were not as attractive as the average diploid or tetraploid seedling from related progenies. Also, triploids generally had shorter bloom periods, more flower bud drops, malformed flowers, and distorted leaves than related diploids or tetraploids.

Variation among tetraploid hybrids was related to the diversity of their parents. The phenotypes of progenies made up of only J, C, and T combinations were limited because of the homozygous nature of these species and dominant genes mentioned earlier. All had orange or yellowish orange flowers with light lavender or bluish cast near the throat. Most were floriferous and some showed purplish spots near the throat. When Tetra 21-D was crossed with the Celebes or 'Tangerine' tetraploids, the hybrids had reddish stems and leaves and open growth habit. Crosses between Tetra 21-C with the same tetraploids produced seedlings with compact growth habit, green leaves and stems. Few floriferous selections of CCTT and JJTT amphidiploids were considered ornamentally superior to either parent. These also bred true from seed. In general, the unequal or unbalanced genomic combinations among the J, C, T tetraploids were ornamentally inferior to the amphidiploids (Table 4) but better than the triploids.

The most diverse tetraploid progenies were derived from the various New Guinea spp. Except for flower color, these hybrids exhibited a wide range of sizes and shapes of flowers and foliage. Hybrids ranged from slow-growing dwarfs to vigorous giants, and from compact to open growth forms. In general, amphidiploids were superior to the 2 or 3 genomic combinations with unequal dosages of various genomes. Selfed seedlings of colchicine-induced amphidiploids were less variable than those derived from amphidiploids created by crossing autotetraploids. This was probably due to gene differences in the basic sets making up the various species and hybrids within the New Guinea group. Colchicine-induced NNTT and NNCC generally bred true from seed. Depending on their parentage, certain colchicine-induced NNJJ hybrids did not breed true from seed due probably to crossovers among homologous chromosomes in certain J and N sets. Several NNTT, NNJJ, and CCTT that had attractive flowers and everblooming habit were selected for their potential use as seed-propagated cultivars.

Discussion

Polyploidy may have played an important role in extending the geographical boundaries of *Impatiens* (10). The diploid Java and New Guinea species could have originated as tetraploid and octoploid species of Celebes or other closely related species with $x=4$. Evolutionary changes in chromosome pairing, isolation, and accumulation of mutations over long periods must have converted these polyploids into functional diploids. In this respect the natural New Guinea tetraploid, N13, could be relatively new since its breeding behavior was like the artificially induced New Guinea tetraploids. Observations of somatic chromosomes showed that the New Guinea and Java chromosomes were quite similar, whereas the Celebes spp. were different. Beck et al. (6) and Pasutti et al. (12) have reported similar observations. Cytological studies of meiotic chromosomes and chemotaxonomic investigations (7, 8) should provide valuable information about speciation in these groups

of *Impatiens*.

My results indicated that interbreeding hybrid populations could be created and maintained as amphidiploids at the 16, 24, 40, or 48 chromosome levels. Interploidy crosses were difficult and whatever progenies resulted from these crosses were sterile or nearly sterile. Intraploidy crosses for tetraploids other than amphidiploids produced many progenies, but they were also sterile or nearly so. Thus, sterility is a major obstacle for inter-specific hybridization at the diploid level (3) and, to a lesser extent, at the polyploid level. Preliminary studies showed that the progression to higher polyploid series might not restore fertility (unpublished data). Several colchicine-induced octoploids studied so far have been sterile and morphologically defective.

Alternatively, it is possible to breed among diploid species within the New Guinea group. Variants among several identified and unidentified species within this group are ornamentally attractive and diverse (15, 16). Many useful and ornamental cultivars have been produced by breeders in commercial and research institutions from New Guinea diploids (5, 9). Ornamental attributes sought by breeders, such as variegated foliage, attractive flowers, and everblooming habit, have been found in this group. But breeding exclusively within this group has some disadvantages resulting from the general vulnerability of crops with narrow genetic bases. Also, seed-reproduced cultivars, such as inbreds and F₁ hybrids, will require more time and effort to produce than amphidiploids.

There are several practical reasons for widening the genetic base of the New Guinea group by hybridization with species from Java, Celebes, and islands around tropical India. Nearly 50% of over 500 identified *Impatiens* spp. grow in India (11). Species from some of these islands might function as bridges between the New Guinea and Indian groups. Ecotypes of *I. platypetala* Lindl. found in Java and Celebes, such as those described by Van Steenis (13), might have the requisite recessive or neutral floral characteristics for amphiploid production. Genes for tolerance to heat and drought required for outdoor plants or genes for tolerance to low levels of light and humidity required for indoor plants might be found in the untested species from these areas. Early flowering and tolerance to the hot summers of certain areas in midwestern United States are necessary in order to grow *Impatiens* in these regions, and attempts to breed among Java spp. for these characteristics have been reported by Pasutti et al. (12).

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Growth and Yield of Crops Treated with Triacontanol¹

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Abstract. The growth of several vegetable and field crops in the greenhouse was increased by applications of 1-triacontanol to the foliage, soil, or seed. Neither the seed nor soil treatments increased the yield of crops in the field. However, foliar sprays ranging from 5 to 500 mg/ha significantly increased the marketable yield of 7 of 10 crops tested. The average yield increase was based on comparisons of all the different rates and time of 1-triacontanol applications with untreated controls. The response of tomato, carrot and wheat seed treatments with 1-triacontanol was shown to be positively correlated with temperature at time of germination and early growth.

Alfalfa (*Medicago sativa* L.) hay (4) and 1-triacontanol, [CH₃(CH₂)₂₈CH₂OH] isolated from alfalfa hay (5) has been shown to stimulate the growth of several crop species (5) as well as tobacco callus tissue (3). Triacontanol was previously shown to be the principal long-chain alcohol component of wax derived from alfalfa leaves (2).

Triacontanol applied in nutrient solutions to rice seedlings at 2.3×10^{-8} M (10 µg/liter) increased seedling dry wt and protein (nitrogen) content within 3 hr in the light or dark (6). Growth analysis indicated that the direct effect of triacontanol occurred within the first 24 hr after treatment (6). Treated plants continued growing larger than controls, but did not gain more dry wt per cm² of leaf area. This increase in dry wt had a V_{max} of 4.9 mg/hr and a K_{dose} of 25 min in the dark (1). This response was affected by both CO₂ and O₂ concn with the optimum response occurring at 200-300 µl/liter of CO₂ and 5% of O₂. Dark grown seedlings treated with triacontanol did not fix more atmospheric CO₂ than did controls (1).

The consistent increase in growth due to triacontanol observed in these studies indicated that triacontanol may be useful in crop production. The objective of this research was to determine the best rates, methods, and times of triacontanol application for increasing the growth of several crops under greenhouse conditions and the yield under field conditions.

Materials and Methods

Greenhouse experiments. Studies were carried out using a sterilized soil mix consisting of equal volumes of peat, sand and a sandy loam soil. Clay pots 10 and 18 cm in diam were used for small and large seeded crops, respectively. After seedlings emerged they were thinned to a uniform stand of 3 to 5 plants per pot. Plants were assigned to blocks based on uniformity of size. A random number table was used to assign treatments. They were grown in a greenhouse with a night temp maintained at about 25°C. In the temp studies, plants were grown

under an 8 hr night and a 16 hr day with an irradiance of 270 µEs/cm² (Lambda LI 185 with quantam sensor) at the soil level. After thinning, plants were watered twice a week with a soluble 20N – 8.6P – 16.6K fertilizer at the rate of 1 g/liter with 100 ml and 250 ml per pot for the 10 and 18 cm pots, respectively.

Triacontanol was applied to seeds by soaking the seed in dichloromethane containing triacontanol for 1 hr. Controls consisted of untreated seed and seed soaked in dichloromethane, except for the temp study where dichloromethane treated seed was the only control. A 2.5% emulsifiable concn of triacontanol (American Cyanamid, Princeton, New Jersey) was used for foliar applications. This formulation was still so concentrated that the rates used required the addition of Tween 20 at 0.1% by vol as a surfactant. Studies conducted in the greenhouse with the emulsion blank and Tween 20 used alone showed that neither influenced plant growth. The controls in these tests were all sprayed with either the commercial emulsion or chloroform and Tween 20. The sprays were applied with an atomizer until the liquid dripped from the leaves. The triacontanol solutions for the soil drenches were made up from dilutions of a 0.1 mg/liter stock solution. This was obtained by adding the pure chemical to water and stirring over heat for several days to get it into solution. The 1.0 mg/liter treatment was applied by dissolving the triacontanol in chloroform (1.0 mg/0.5 ml). The chloroform solution was added to water, rapidly shaken and added to the soil. The soil drenches were applied in 100 ml and 250 ml of water for the 10 and 18 cm pots, respectively. Plants were harvested at the soil surface and dried in an oven at 80°C for at least 2 days. Randomized complete block designs with 4 to 6 blocks were used for all tests. Means were compared by the LSD procedure.

Field experiments. Field tests were conducted at 3 locations in central Michigan on a Spinks sandy loam, Miami clay loam, or Park Hill clay loam. Normal cultural and pest control practices for crop production in Michigan were utilized in all field studies.

The foliar treatments were applied in the field with 1 or 2 flat fan nozzles with the same formulations used in the greenhouse. The Tween 20 was applied to controls when there was

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