Crossability of Italian Ryegrass (*Lolium multiflorum* LAM.) with Tall Fescue (*Festuca arundinacea* SCHREB.)

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Three experiments were carried out to gain more detailed information on the crossability between Italian ryegrass and tall fescue in order to develop an effective breeding program of these intergeneric hybrids. In the first experiment three Italian ryegrass cultivars, as female parents, and 12 tall fescue cultivars and strains, as male parents, were used. There were wide ranges of variations in the crossability between cross combinations. The number of hybrids per spike (crossability) ranged from 16.4 to 103.7 and 17.3 to 145.0 in the cross combinations of 1980 and 1983, respectively. A specific tall fescue cultivar showed a high crossability with Italian ryegrass in both years. In the second experiment genotypic effects of Italian ryegrass on crossability were investigated in crosses of four Italian ryegrass clones with six tall fescue clones. There were significant differences in the crossability among the female Italian ryegrass clones. Among the male tall fescue clones the differences in the crossability were present, but they were not significant at the 5% level. In the third experiment the inheritance of the high crossability of a clone of Italian ryegrass cv. Waseaoba, W-2, was investigated using its selfed progeny plants. Most of the selfed progeny plants of the W-2 clone showed high levels of crossability (percentage of seed set) with a tall fescue clone. The mean seed set was 84.5% with a range from 61.5% to 95.7%.

KEY WORDS: *Festuca arundinacea* SCHREB., *Lolium multiflorum* LAM., crossability, intergeneric hybridization.

Introduction

Tall fescue, *Festuca arundinacea* SCHREB., and Italian ryegrass, *Lolium multiflorum* LAM., were introduced into Japan more than 100 years ago. Tall fescue is adapted to a wide range of climatic conditions in Japan. Its winter-hardiness, summer tolerance and persistency are desirable characteristics, but the plant is less acceptable to animals than many other cool season grasses. Italian ryegrass, on the other hand, is highly nutritious and palatable, and the most important grass in the southern part of Japan.

There is a close relationship between these species and many hybrids between them have been reported (CROWDER, 1953, LEWIS 1959 and 1966, BUCKNER 1960, BUCKNER *et al.* 1961, NAGAMATSU 1964, SULINOWSKI 1964, BEDDOWS 1965, ZWIERZYKOWSKI 1980 and TERADA 1983). Since 1980, we made numerous crosses between these species to produce a forage grass with combined favorable characteristics of both Italian ryegrass and tall fescue. At the time when seed sets of these crosses were scored, a wide range of variation in crossability was detected. BUCKNER (1960) and LEWIS and THOMAS (1986) reported that there were genotypic differences in the crossability between Italian ryegrass and tall fescue.

The present study was undertaken to gain more detailed information on the crossability between Italian ryegrass and tall fescue in order to develop an effective breeding program of these intergeneric hybrids.

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Materials and Methods

(i) Experiment 1: Crossability of Italian ryegrass cultivars with tall fescue cultivars and strains

The cultivars of Italian ryegrass and tall fescue, and strains of tall fescue used in this experiment are shown in Table 1. The Italian ryegrass cultivars used in the crosses were all Japanese ones. The tall fescue cultivars consisted of Japanese, European and American cultivars. The strains were ecotypes collected in Tunisia and France. Plants used in the investigations were grown in both an unheated greenhouse and in the field to prolong the crossing period. All hybridizations with tall fescue were carried out with Italian ryegrass as the female parent. The crossings were made by enclosing inflorescences of both parents in pollination bags before anthesis. Emasculation was not carried out since the self-fertility of Italian ryegrass is usually low. The seeds from each spike of the Italian ryegrass plants were harvested at the maturity stage, and sown in seedling pots in the unheated greenhouse. Seedlings were transplanted into the field in fall where hybrids were identified by the type of inflorescence during the following spring. The crossability was expressed by the number of hybrids obtained per spike.

(ii) Experiment 2: Female effect on the crossability of Italian ryegrass with tall fescue

Four genotypes of the Italian ryegrass cv. Waseaoba as female and six genotypes of the tall fescue cvs. Manade, Ky. 31, and Yamanami as male were used in this experiment. All the genotypes used were selected at random from plants grown in pots in the greenhouse and in the field, with the exception of one Italian ryegrass genotype (W-2), which was selected to confirm the presence of a genotype with high crossability, since it showed a high crossability with tall fescue in Experiment 1. The pollination method, hybrid identification and scoring procedures of crossability were similar to those described in Experiment 1.

(iii) Experiment 3: Crossability of selfed progeny plants of a highly crossable Italian ryegrass plant (W-2) with tall fescue

In order to determine whether the high crossability of Italian ryegrass plant with tall fescue was inherited in the next generation, 14 selfed progeny plants of the plant W-2 were used. They were grown together with the clone M-9 of the tall fescue cv. Manade, which was used in Experiment 2.

Prior to flowering, spikes of the selfed progeny of the plant W-2 were emasculated by hand and pollinated under controlled conditions using detached inflorescences of the clone M-9. For each cross combination the percentage of seed set was recorded. The crossability was expressed by the percentage of seed set in this experiment.

Results and Discussion

(i) Experiment 1

Results of crossing of Italian ryegrass cultivars with tall fescue cultivars and strains are shown in Table 2. In 1980, 59 crosses were made and 3,212 seeds were produced, 2,233 of which were hybrids. Analysis of variance revealed significant differences ($P < 0.05$) in the

Table 1. Species, cultivar or strain name, chromosome number and origin

Species	Cultivar or strain name	Chromosome number (2n)	Origin
<i>Lolium multiflorum</i>	Nasuhikari	14	Japan
	Waseaoba	14	Japan
	Yamaaoba	14	Japan
<i>Festuca arundinacea</i>	Aronde	42	Netherlands
	Bn271	42	UK
	Bn954	42	UK
	Jebel	42	UK
	Kasba	70	UK
	Kenwell	42	USA
	Ky.31	42	USA
	Ludion	42	France
	Manade	42	France
	Pastuca	42	Netherlands
	Tallmo	42	Netherlands
	Yamanami	42	Japan

Bn271 and Bn945 are ecotypes collected in Tunisia and France, respectively.

crossability between cross combinations. Waseaoba $\times$ Kasba showed a significantly higher crossability than the other cross combinations. It produced 103.7 hybrids per spike. However, the least successful cross, Waseaoba $\times$ Ludion, produced only 16.4 hybrids per spike. In 1983, a total of 54 crosses were attempted and 7,218 seeds were obtained; the seeds gave 4,714 mature plants, 4,491 of which were hybrids. There was no significant difference between the results of the same cross combinations, except for Waseaoba $\times$ Aronde, in 1980 and 1983. Waseaoba $\times$ Aronde produced 18.8 and 145.0 hybrids/spike in 1980 and 1983, respectively. Whether this difference is caused by genotypic differences in the crossability between the plants used in 1980 and 1983 or environmental differences during flowering and seed development in two years could not be determined in this experiment.

Analysis of variance of the data obtained in 1983 indicated significant ($P < 0.05$) differences in crossability among the 9 cross combinations. Three cross combinations, Waseaoba $\times$ Aronde, Waseaoba $\times$ Bn945 and Waseaoba $\times$ Kasba, showed a significantly higher crossability than the remaining cross combinations. Waseaoba $\times$ Kasba showed the highest crossability in both 1980 and 1983, indicating that the cv. Kasba is a highly compatible population. The cross combination of Yamaaoba $\times$ Kenwell was the lowest-ranking in crossability and the five other cross combinations, Waseaoba $\times$ Bn271, Waseaoba $\times$ Jebel, Waseaoba $\times$ Ky.31, Waseaoba $\times$ Manada and Waseaoba $\times$ Tallmo, were intermediate. These results indicate that there are genetic variations in crossability between plants of Italian ryegrass and tall fescue.

Table 2. Crossability (number of hybrids/spike) of three *L. multiflorum* cultivars with 12 *F. arundinacea* cultivars and strains

Cross combination	1980						1983			
	No. of spikes crossed	No. of seeds ob-tained	No. of hybrids ob-tained	No. hybrids/spike		No. of spikes crossed	No. of seeds ob-tained	No. of hybrids ob-tained	No. hybrids/spike	
				Range	Mean ^{a)}				Range	Mean ^{a)}
Waseaoba × Aronde	5	208	135	9-32	18.8b	6	1603	870	118-162	145.0a
Waseaoba × Bn945	—	—	—	—	—	6	990	836	4-207	139.3a
Waseaoba × Kasba	6	724	622	65-195	103.7a	6	1066	818	85-197	136.3a
Waseaoba × Bn271	—	—	—	—	—	6	670	494	4-177	82.3b
Waseaoba × Jebel	—	—	—	—	—	6	708	474	45-118	79.0b
Waseaoba × Yamanami	8	358	257	5-73	32.1b	6	634	358	34-129	59.7bc
Waseaoba × Ky.31	6	260	185	1-60	30.7b	6	767	320	11-100	53.3bc
Waseaoba × Ludion	8	278	156	2-29	16.4b	—	—	—	—	—
Waseaoba × Manade	6	324	203	6-29	25.5b	6	452	217	0-98	36.2bc
Waseaoba × Pastuca	5	246	168	4-75	32.8b	—	—	—	—	—
Waseaoba × Tallmo	9	565	304	3-98	31.7b	—	—	—	—	—
Yamaaoba × Kenwell	—	—	—	—	—	6	328	104	1-40	17.3c
Yamaaoba × Pastuca	2	130	99	48-51	49.5b	—	—	—	—	—
Nasuhikari × Pastuca	4	119	104	0-60	26.0b	—	—	—	—	—
Total or mean	59	3212	2233	—	36.7	54	7218	4491	—	83.2

^{a)}Any two means followed by the same letters do not differ significantly at the 5% level.

(ii) Experiment 2

Results of crossing of 4 clones of the Italian ryegrass cv. Waseaoba with six tall fescue clones are presented in Table 3. Analysis of variance revealed significant ($p < 0.05$) differences in crossability of Italian ryegrass clones, but no significant difference in the case of the tall fescue clones. The clone Waseaoba-2 (W-2) exhibited a significantly higher crossability with tall fescue than the other Waseaoba clones, indicating the presence of genotypic variations in Italian ryegrass in relation to crossability with tall fescue. LEWIS and THOMAS (1986) observed a significant difference in crossability between two Italian ryegrass cultivars in crosses of two Italian ryegrass cultivars with one tall fescue cultivar. In crosses between perennial ryegrass (*Lolium perenne* L.) and meadow fescue (*Festuca pratensis* HUDS.), REUSCH (1959) observed genotypic variations in perennial ryegrass in relation to the crossability between them. In the crosses between Waseaoba-8 and 6 tall fescue clones, two cross combinations showed a high level of crossability, while the others a low one, suggesting the presence of interactions in crossability between Italian ryegrass and tall fescue plants.

(iii) Experiment 3

Table 4 shows the results of the crossing of 14 selfed progeny plants of the W-2 clone with a clone of the tall fescue cv. Manade (M-9). Most of the selfed progeny plants of W-2 showed high levels of crossability (percentage of seed set) with the tall fescue clone, as in the case of the mother plant. The mean percentage of seed set was 84.5% with a range from 61.5% to 95.7%. On the basis of the results of the present study it is apparent that the high level of crossability of the W-2 clone with tall fescue is inherited in a high frequency in selfed progeny. This observation indicates that the gene(s) of Italian ryegrass is, at least, one of factors controlling the crossability with tall fescue.

The present study clearly demonstrated for the first time that the Italian ryegrass-tall fescue crossability is strongly influenced by Italian ryegrass genotypes and that the high intergeneric crossability of Italian ryegrass with tall fescue is heritable in a high frequency in selfed progeny. Based on the results of the cross Waseaoba $\times$ Kasba, it is suggested that genotypes or ploidy of tall fescue also influence the crossability with Italian ryegrass. It is considered that the development of Italian ryegrass and tall fescue lines with high crossability could facilitate the production of intergeneric F_1 hybrids between Italian ryegrass and tall fescue, and make it possible to develop an effective breeding program of the intergeneric hybrids.

Table 3. Crossability (number of hybrids/spike) of four clones of *Lolium multiflorum* cv. Waseaoba with six clones of *Festuca arundinacea*

		Manade		Yamanami		Ky.31		Mean
		6	9	11	18	4	18	
Waseaoba	2	114	182	291	138	294	117	189.3
	7	21	63	88	77	60	57	61.0
	8	11	9	107	115	61	39	57.0
	9	68	67	27	31	69	20	47.0
Mean		53.5	80.3	128.3	90.3	121.0	58.3	88.6

Table 4. Seed set in crosses of 14 selfed progeny plants of W-2 clone with a clone of cv. Manade

Clone No.	No. of flowers		Seed set	
	Pollinated	No.	%	
W-2/S-1	135	83	61.5	
2	101	75	74.3	
3	123	111	90.2	
4	114	99	86.8	
5	132	108	81.8	
7	113	95	84.1	
9	124	116	93.5	
10	49	37	75.5	
11	134	121	90.3	
12	148	123	83.1	
13	104	84	80.8	
14	90	78	86.7	
15	117	112	95.7	
16	125	111	88.8	
Total or mean	1609	1353	84.5	

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イタリアンライグラスとトールフェスクの交雑親和性

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寒地型イネ科牧草のイタリアンライグラス (*Lolium multiflorum* LAM.) とトールフェスク (*Festuca arundinacea* SCHREB.) との属間交雑育種を効果的に推進するために、両種の交雑親和性に関する3通りの実験を行った。

最初の実験では、イタリアンライグラス3品種とトールフェスク12品種・系統間で2か年(1980, 1983)にわたって交雑を行った。その結果、交雑親和性(雑種個体数/穂)は2か年とも交雑組合せ間で大きな変異が認められ、1980年には16.4~103.7, 1983年には17.3~145.0であった。また、トールフェスクの中に、両年とも高い交雑親和性を示す品種が認められた。

第二の実験では、交雑親和性に及ぼすイタリアンライグラスの遺伝子型の影響をみるために、イタリアンライグラス4クローン(遺伝子型)とトールフェスク6クローン(遺伝子型)との交雑を実施した。交雑親和性は、母親のイタリアンライグラスの遺伝子型によって有意に異なったが、トールフェスクの遺伝子型では有意な差は認められなかった。

第三の実験では、イタリアンライグラスの品種ワセアオバのW-2クローンは交雑親和性が非常に高かったので、その遺伝性をみるために、W-2クローンの自殖次代植物(W-2/S)とトールフェスクのクローンとの間で交雑を行った。その結果、14個体のW-2自殖次代植物とトールフェスククローンとの稔実率は平均84.5%, 範囲62-96%で、大部分の個体が高い交雑親和性を示した。