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Breeding for the Heat Resistant Rhododendrons

IV. The Reliability of Early Selection of Seedlings on to the Heat Resistance

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Introduction

In the course of their experiments for the heat resistant rhododendrons, one of the authors (K.A.) noticed the trend that the survival rate of the larger sized seedlings appeared to be consistently higher than that of the smaller sized ones within the same species or the same cross combination. Therefore, the preliminary experiment¹⁾ was conducted, using the three years old seedlings of various species and hybrids divided into three different sizes, large, medium and small, and it was found that the above mentioned observation was substantially valid.

In the practical breeding programme, an introduction of some reliable measure for the early selection of hybrid seedlings is instructive to promote the systematic and efficient breeding, especially in the plant species with long juvenile period or long life span. The present experiment was, therefore, carried out to confirm whether the above mentioned trend was still to be found in the tinier one year old seedlings, in order to make clear the feasibility of early, reliable selection for the efficient breeding in rhododendrons.

Materials and Methods

The artificial crosses were done in the spring of 1983, and the seeds were harvested and sown in the autumn of that year. The seedlings grown under long day at 20°C were divided, based on the size and the total number of seedlings, into either large, medium and small groups or large and small groups, and were transplanted into the plastic boxes (36×45×10cm) in middle March of 1984. Because of the difference in seed yields, the number of seedlings which could be examined was largely different according to the cross combination, but the mean number of seedlings per cross combination at the start of measurement, done on May 5 and mentioned just below, was either 233.4 in the former group or 96.0 in the latter group. The media used were the mixtures of pumice, weathered pumice and red clay, 3:1:1 by volume. They were grown in the same plastic house as in the previous experiment²⁾, covered with a sheet of black cheese cloth and left under natural day length. The liquid fertilizer containing N, P and K, each 100 ppm, was given twice a week from early May to late October.

The measurements of seedling growth, *i.e.*, survival rate, vigour, plant height and leaf size (width×length) were done *ca.* every 45 days from May 5 when the transplanted seedlings appeared

to have been fully established, to December 14, the termination of experiment, but in the present paper, only the survival rate will be described.

Results and Discussion

Table 1 is the final survival rate at the termination of experiment. Fig.1 is the consecutive changes in the survival rate during the experimental period.

As seen in both table and figure, the final survival rate in the large sized seedlings was extremely high, ranging from 100% to 58%. Of 49 cross combinations, those with higher survival rate than 85% were 38 combinations (77.6%) and that with lower rate than 60% was only one combination (2.0%); 'Vulcan's Flame' × *R.yakushimanum*. On the other hand, in the small sized seedlings the cross combinations with higher survival rate than 85% were only 5 combinations (10.2%), and the majority or 34 combinations (69.4%) were lower than 60% survival.

Moreover, the survival rate decreased with the decrease of seedling size, although some discrepancies were met with in 3 combinations; 'Blue Ensign' × *R.metternichii* var. *hondoense* f. *brevifolium* 1, 'Blue Ensign' × *R.simiarum* 2 and *R.metternichii* var. *hondoense* f. *brevifolium* 5 × *R.formosanum* 2, because the reversal of survival rate between seedling sizes was observed. As seen in Fig. 1, however, these discrepancies were so small that they did not seem to be due to the fundamental causes. In other words, the seedlings showing consistently vigorous growth immediately after seed germination were genetically the superior individuals and *vice versa*, and the reliable early selection for heat resistance might be feasible even in the one year old seedlings. The overall trend in the features of decrease both in the size of seedlings and during the experimental period was summarized in the right-bottom corner of Fig.1, which indicated clearly the above mentioned generalization.

The acceleration of breeding programme, or the feasibility of reliable early selection has been one of the important problems, especially in the plant species with long juvenile period or long life span.

The rhododendrons are, in general, slow growing plants and before attaining flowering they require much longer time than the azaleas. Under the long day condition at 15 to 20°C, Doorenbos⁵⁾ succeeded in earlier flowering in the seedlings of rhododendron, deciduous azalea and their hybrid in half the normal time of those under the natural conditions.

In 1961, Stuart²⁵⁾ applied the growth retardants, phosfon and CCC, to azalea cv. 'Coral Bells' and revealed that they caused the suppression of vegetative growth and the prompt initiation of flower buds. Similar hastening of flower bud initiation in azaleas by growth retardants was also confirmed by other workers^{18,36)}, and being combined with other treatments the procedures for the year-round production of flowering azaleas were developed¹⁷⁾.

In rhododendrons the growth retardants, phosfon and B-Nine, induced the flower bud formation in a way similar to that in azaleas⁴⁾. Recently, Kunishige *et al.*¹⁶⁾ found the more pronounced effect in Sumi-7, a growth retardant, on the induction of flower buds in rhododendrons. The foliar application of 250 ppm of this chemical to the 2 years old cuttings of cv. 'Jean Marie' brought forth the successive flowerings, beginning from the next season, whereas the untreated plants remained vegetative and the first flowering occurred 3 years later in the 6 years old cuttings. In 250 ppm, this chemical induced the heavy stunted growth, extending for several years, but in many cultivars it was disclosed to be effective even in 50 or 25 ppm. Although no attempt appeared to have been made so far for hastening the flowering of the hybrid seedlings with this chemical, it might

be a matter worthy to be considered, once they attained a certain size, for hastening the flowering and thus shortening the breeding cycles in rhododendrons.

In other woody plants, the top working has been widely employed for fastening the flowering and fruit setting in the breeding of fruit trees^{13,21,33,37,38)}. In citrus, the tongued-approach graft was revealed to stimulate strongly the growth of the seedlings as well as the shoot-tip grafted saplings²⁸⁾. The girdling also promoted the flowering and fruiting of juvenile citrus seedlings, although its effect was generally inferior to that of top working and the seedlings of such species as *C. grandis* showed almost no response²⁹⁾.

Based on the careful reading of 185 literatures, Zimmerman³⁹⁾ evaluated the effects of genotype, environment, grafting, girdling and so on, and recognized the following items to be effective on the earlier flowering, *i.e.*, (1) selection of genotypes with precocious flowering, (2) any procedure to attain large seedling size as rapidly as possible, (3) grafting seedlings onto dwarfing rootstocks or onto bearing trees, (4) girdling, scoring, bark inversion or root pruning and (5) sprays with growth retardants. The similar aspects concerning the juvenility, maturation and rejuvenation in woody plants were also reviewed by Hackett¹⁰⁾.

As for the early selections for the other characteristics in woody plants, the extensive co-working was conducted from 1968 to 1972 by Japanese researcher group for breeding of woody crops on fruit trees, forest trees, tea plant and mulberry²²⁾. Their results regarding the development and application of the methods for early diagnosis were published in 1973, in which were included the various outcomes on the resistance for disease, insect pest and freezing or frost injury, the quality of product and the yield. In tea plant, Toyao *et al.*³⁰⁾ investigated the bark splitting injury in late autumn and the frost hardiness in midwinter, and developed the methods of reliable selection, in the former case by freezing a few small stem pieces or cuttings and in the latter case by measuring the electric conductivity of exosmosis of frozen leaves, which, especially the former was applied extensively to the early selection of practical tea breeding.

In citrus with polyembryonic seeds, the identification of hybridity in embryo or seedling was done by the comparison of colour of mature cotyledon³¹⁾ or by the gas-chromatographic analysis of leaf oils of the seedling¹¹⁾. In Japanese pear and apple, the early selection for pear black spot¹⁵⁾ and apple rust²³⁾ was done by the inoculation onto young hybrid seedlings. The similar applications of early diagnosis for disease^{3,6,14,20,26,27,35)} and pest^{7,8)} resistance and other characteristics¹⁹⁾ to practical breeding were also known in vegetables.

In ornamental plants, the seedlings of tulip require *ca.* 6 years to reach an adult phase under the natural environment. However, under the controlled environment, especially for thermoperiodicity, Shisa and Higuchi²⁴⁾ disclosed that the juvenile phase might be shortened to *ca.* 3.2 years. Eijk *et al.*⁹⁾ studied the parameter for early selection in the forcing ability in tulip, and revealed that both time of emergence and time of dying down during the juvenile phase were closely correlated with the flowering date after forcing in the glasshouse and could be used as the parameter of early selection. The similar studies for early selection of hybrid seedlings were also made in rose for the cut stem length³²⁾ and in chrysanthemum for the disease resistance^{12,34)}.

So far as the authors are aware, no investigation has been conducted on the method of reliable early selection for the heat resistance in rhododendron seedlings. Except in the northern regions where the summer is cool, the usual practices of Japanese gardeners to grow rhododendron seedlings have been to transplant all of the tiny seedlings (*ca.* 6 or 3 months old) from seed bed to appropriate pots or boxes in early spring (when seeds were sown in the previous autumn) or early rainy season (when seeds were sown in early spring), and raise them elaborately thereafter. The underlying idea

Table 1. The final survival rate of seedlings at the termination of experiment (Dec. 14, 1984)*¹

Cross combination		Survival rate (%)			Cross combination		Survival rate (%)			
		L	M	S* ²			L	M	S	
BB	× MBC* ²	93		51	HC · 2	× psc	86		71	
	× fm · 1* ³	100		50	HC · 3	× mb · 4	99		53	
	× fm · 2	100		100	MF	× hp · 1	100	84	51	
	× sm · 1	93		73		× hp · 2	96	77	57	
BE	× fm · 1	99		75	× mb · 2	70	61	44		
	× fm · 2	88		63	× mb · 3	93		43		
	× fm · 3	95		89	× mr · 1	85		58		
	× hp · 1	73		41	× psc	88		41		
	× mb · 1	85	91	60	× sm · 2	100	86	20		
	× sm · 2	90		100	MR	× fm · 2	100	94	77	
DM	× fm · 2	100	75	53		× mb · 1	100		90	
	DOC	× mb · 2	98		39	PW	× fm · 1	98	60	17
		× mb · 3	82		15		× fm · 2	98		42
		× met	100		80		× fm · 3	100		62
× sm · 2		100		16	× hp · 1		80	55	47	
EL	× sm · 1	67		33	RH	× fm · 2	79		21	
FD	× fm · 1	74		0	VF	× yak	58		33	
	× fm · 2	100		58		hp · 3	× mr · 1	97		36
	× sm · 3	81		30		hp · 4	× fm · 2	100		73
HC · 1	× GT	100	98	58	× sm · 1	96		73		
	× fm · 1	96	90	28	mb · 2	× sm · 2	74	53	14	
	× mb · 4	100	63	13		mb · 3	× fm · 2	100		70
	× mr · 1	95	94	33	mb · 5	× fm · 2	100	92	96	
	× mr · 2	98		57						
	× psc	74	56	52						
	× sm · 4	98	64	12						

*1. The survival rates (%) were calculated based on the number of seedlings at the start of measurement (May 5, 1984).

*2. Abbreviation: L; large, M; medium, S; small, BB; Baden Baden, BE; Blue Ensign, DM; Dusty Miller, DOC; Doc, EL; Elizabeth, FD; Firedance (RM11), GT; Golden Torch, HC; *R. hyperythrum* × *Cynthia*, MBC; *R. metternichii* var. *hondoense* f. *brevifolium* × *Cynthia*, MF; Mrs. Furnival, MR; Mrs. Roosevelt, PW; Percy Wiseman, RH; Unnamed *R. forrestii* var. *repens* hybrid, VF; Vulcan's Flame, fm; *R. formosanum*, hp; *R. hyperythrum*, mb; *R. metternichii* var. *hondoense* f. *brevifolium*, met; *R. metternichii*, mr; *R. morii*, psc; *R. pseudochrysanthum*, sm; *R. simiarum*, yak; *R. yakushmanum*.

*3. The numerical suffix means different individual within the same species or cross combination.

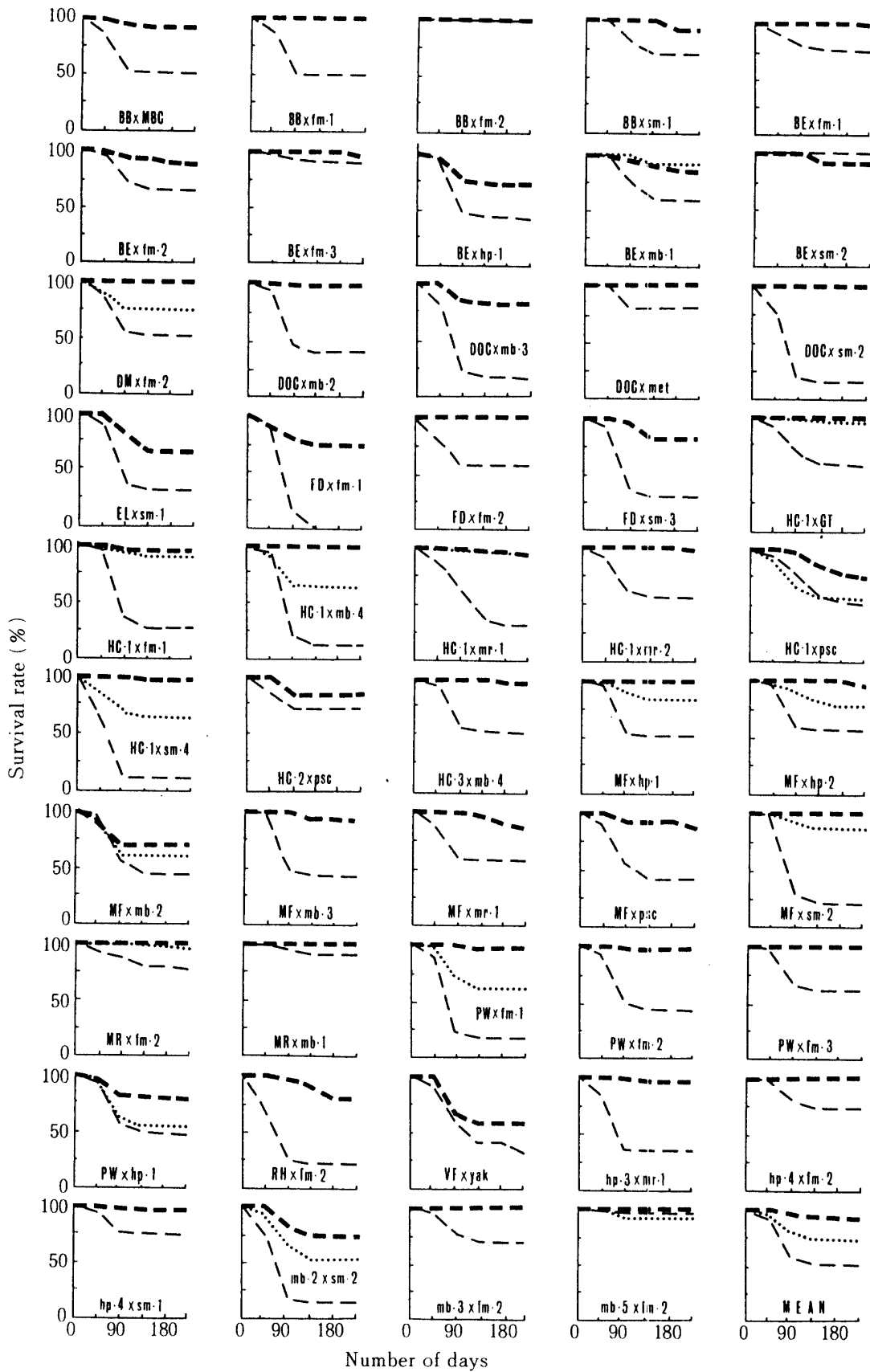


Fig. 1. The consecutive changes in the survival rate of different size seedlings.
 The size of seedlings : heavy broken line ; large, dotted line ; medium, fine broken line ; small.
 For survival rate and other items, see footnotes of Table 1.

in these practices has been how to make the seedlings escape from the killing high temperature by establishing them before the commencement of the first, hot summer, and how to increase the overall survival. However, the contrary is the fundamentals of breeding, since it requires the elimination of undesirable genotypes. In the present experiment it was disclosed that a reliable selection for the heat resistance was possible, even in the one year old seedlings. Therefore, in the practical breeding the following procedures might be recommended for the efficient breeding, *i.e.*, to save money and labour and to deal with larger seedling population; (1) to sow the seeds as thinly and uniformly as possible and raise the seedlings through next one or two summers under the unavoidable selection without superfluous care, and (2) to transplant the survived seedlings, if necessary with the choice of larger seedlings, and impose further severe selections on them.

Summary

The investigation on the early selection for the heat resistance of rhododendrons was carried out, using the one year old seedlings of different sizes. Within the same cross combination the survival rate varied with the seedling size, and it increased as the size increased and *vice versa*. Some substantially same results had been obtained in the preliminary experiment¹⁾, using the three years old seedlings. Based on these results, the application to practical breeding was discussed.

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