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Screening of rice (*Oryza sativa* L.) genotypes for physiological characters contributing to salinity resistance, and their relationship to overall performance

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Summary. Phenotypic resistance of salinity is expressed as the ability to survive and grow in a salinised medium. Some subjective measure of overall performance has normally been used in plant breeding programmes aimed at increasing salinity resistance, not only to evaluate progeny, but to select parents. Salinity resistance has, at least implicitly, been treated as a single trait. Physiological studies of rice suggest that a range of characteristics (such as low shoot sodium concentration, compartmentation of salt in older rather than younger leaves, tolerance to salt within leaves and plant vigour) would increase the ability of the plant to cope with salinity. We describe the screening of a large number of rice genotypes for overall performance (using an objective measure based on survival) and for the aforementioned physiological traits. There was wide variation in all the characters studied, but only vigour was strongly correlated with survival. Shoot sodium concentration, which a priori is expected to be important, accounted for only a small proportion of the variability in the survival of salinity. Tissue tolerance (the cellular component of resistance reflecting the ability to compartmentalise salt within leaves) revealed a fivefold range between genotypes in the tolerance of their leaves to salt, but this was not correlated positively with survival. On the basis of such (lack of) correlation, these traits would be rejected in normal plant breeding practice, but we discuss the fallacies involved in attempting correlation between individual traits and the overall performance of a salt-sensitive species in saline conditions. We conclude that whilst overall performance (survival) can be used to evaluate the salt resistance of a genotype, it is not the basis on which parents should be selected to construct a complex character through breeding. It was the norm for varieties which had one good characteristic affecting salt resistance to be unexceptional or poor in the others. This constitutes experimental evidence that the potential for salt resistance present in the rice genome has not been realised in genotypes currently extant. The results are discussed in relation to the use of physiological traits in plant breeding, with particular reference to environmental stresses that do not affect a significant part of a species' ecological range.

Key words: *Oryza sativa* – Salinity – Screening

Introduction

Rice is a salt-sensitive crop species, and soil salinity is the single most widespread soil toxicity problem facing rice production (Greenland 1984). Although other crops may be more tolerant of salinity, for the farmer there is often no alternative to growing rice, because it is the only major crop to tolerate the flooding that is common in the wet season in the humid tropics. Soil salinity affects 2 of the 15 million km^2 of land under cultivation and between 30% and 50% of irrigation agriculture (Flowers et al. 1986). If rice production per capita is to remain close to present levels as the population rises, then an increase in salinity resistance is necessary because good agricultural land is a limited resource (Toenniessen 1984).

The quantification of salinity resistance poses serious difficulties. (1) In the field, the stress may be experimentally uncontrollable because of climatic effects upon rainfall and water tables. (2) Field heterogeneity for salinity is notoriously high (Richards 1983; Malcolm 1983) and is liable to confound any planting plan designed for field experiments. (3) Salt uptake, and so sensitivity, is modulated by environmental conditions that may affect each variety differently: any parameter which affects the transpiration rate (such as light intensity, temperature and

humidity) can dramatically change a plant's susceptibility to salinity. Salinity damage is not a determinate quantity, but can be expected to show extremely high genotype/environment interaction. We describe and evaluate a greenhouse-based assessment of phenotypic performance based on survival.

Studies of the effects of salinity on rice have indicated that salinity damage, and consequently adaptation to salinity, is complex. No single process can account for the variation in the plant's response to salinity; the subsequent distribution of salt within the plant is as important as the uptake of salt in the first place (Yeo and Flowers 1984, 1986, 1989). Varietal variation has been observed within small (ten or fewer) samples of genotypes in characteristics that modify or mitigate the consequences of salt uptake, such as compartmentation within the plant (Yeo and Flowers 1982), within the leaf tissue (Yeo and Flowers 1983), reflecting compartmentation between apoplast and protoplast (Flowers and Yeo 1986) and water-use efficiency (Flowers et al. 1989). We have hypothesised that salinity resistance in rice might be increased through combining parents with these individual physiological characteristics, rather than treating salt resistance as if it were a single character (Yeo and Flowers 1986). The assumption is that available variation has not been combined favourably in a single genotype, but that this could be achieved through a breeding programme.

The purpose of these studies was to test experimentally whether the potential to increase salt resistance in this way exists. It is not possible to examine exhaustively all the available lines of rice since, as it is an in-breeder, there are more than a hundred thousand; but a realistic sub-sample is necessary. In this paper we report the screening of a substantial number of genotypes for overall response to salinity and for a number of physiological characters (shoot Na concentration, leaf compartmentation, tissue tolerance and vigour).

Materials and methods

Selection of plant material

Seeds of *Oryza sativa* L. cultivars, breeding lines, elite breeding lines and landraces were obtained principally from the International Rice Research Institute, Manila, The Philippines. Some seed was raised in a greenhouse at the University of Sussex.

The logistics of carrying out laboratory-based screening of highly variable (Flowers and Yeo 1981; Yeo et al. 1988) material determined a sample size of about 200 genotypes (with less for some procedures that require several hundred individual measurements per variety per assessment). A conscious selection from the more than 100,000 rice accessions was preferred to a random one. A subset of the IRRI germ plasm collection was chosen to include accessions of various reputation with regard to salt resistance, most of the cultivars released by IRRI and a range of elite breeding lines used frequently in recent crosses.

Screening for phenotypic resistance to salinity

Seeds were washed to remove excess pesticide and were soaked for 24 h in aerated water at 20°–25 °C. They were sown on nylon mesh ("Netlon") supported on "Perspex" grids (70 seeds of one genotype to an area 200 mm by 150 mm constituted a site) floated on a series of interconnected tanks of recirculating culture solution (allowing 24 sites for each trial). The tanks were of an inert plastic chosen for negligible phytotoxicity (Soil-less Cultivation Systems Ltd., Haslemere, UK) and the culture solution was that described by Yoshida (Yoshida et al. 1976).

Seven days after sowing, ungerminated and obviously retarded seed was removed, and the number of seedlings was recorded (usually 65–70 and never less than 50). At 8 days, the solution was salinised with NaCl to a concentration of 70 mol m^{-3} (a total electrolyte concentration resulting in an electrical conductivity of 8 dS m^{-1}) by supplying 5,000 mol m^{-3} NaCl via a drip feed such that the salinity rose smoothly over a period of 24 h. Salinisation was carried out with NaCl alone, because the resultant Na : Ca ratio had no discernable effect on salt damage in seedlings of this species (Yeo and Flowers 1985). The number of dead plants was recorded at intervals. In preliminary trials, recordings were frequent to assess the optimal times for recordings, but during the actual screening trials recordings were restricted to minimise disturbance. The proportion of individuals dead or alive was related to two control sites in each tank planted with the reference elite breeding line IR2153-26-3-5-2, at the time when approximately 50% of the reference line had died. The mortality of each variety was normalised on the check and converted to a score on the scale from 1 (the best survivor) to 9 (the worst survivor) to conform with plant breeding practice (Table 1). The data are reported in this way because it also simplifies presentation.

The greenhouse was heated to a minimum of 25 °C by day and 20 °C by night. Upper temperatures were limited in summer by automatic venting to 28° ± 3 °C. Air was stirred by two roof-mounted fans to give an average velocity of 0.5 m s^{-1} measured with a hot-wire anemometer (ELE International Ltd.). Humidification was provided by mist jets with control via a capacitance sensor (Conviron, Winnipeg). Supplementary light was supplied for 12 h per day at 400 μmol m^{-2} s^{-1} photosynthetically active radiation (measured with a Licor LI190 SB sensor) from high-pressure sodium lamps (GEC SON/T 400 W, Camplex Plantcare Ltd.). Maximum photon fluence rates were those of the solar radiation at the time, and were up to 1,000 μmol m^{-2} s^{-1} for short periods and, though humidity averaged 70%–80% RH, it was lower during venting and during low external winter temperatures. Wherever screening takes place, climatic variation will occur, and the effects of this were assessed by replicate trials over a 2-year period.

Screening for shoot sodium concentration

Plants were grown in controlled environment conditions of 12 h photoperiod of 400–500 μmol m^{-2} s^{-1} P.A.R. at 27 °C and 1.5-kPa saturation vapour pressure deficit (svpd). The dark period was 25 °C and 0.6-kPa svpd. After 7 days, the seedlings were thinned to 32 plants per variety and salinised (60 mol m^{-3} with NaCl); they were harvested 10 days later. For each trial, there were four experimental sites in each of eight plastic tanks of 10-l that were rotated daily; one site in each tank was occupied by the reference variety (IR2153-26-3-5-2), upon which the results for each tank were normalised.

At harvest, plants were weighed, dried and extracted in acetic acid (100 mol m^{-3}) at 90 °C for 2 h and Na was determined by atomic absorption spectrophotometry (Pye Unicam SP9 800).

Table 1. Scoring systems

Score	Character			
	Survival	Shoot Na	Tissue tolerance	Leaf compartmentation
1	0.00–0.05	0.00–0.35	>5.0	0.00–0.15
2	0.05–0.25	0.35–0.50	4.5	0.15–0.30
3	0.25–0.50	0.50–0.70	4.0	0.30–0.45
4	0.50–0.90	0.70–0.90	3.5	0.45–0.60
5	0.90–1.10	0.90–1.10	3.0	0.60–0.75
6	1.10–1.25	1.10–1.40	2.5	0.75–0.90
7	1.25–1.50	1.40–1.75	2.0	0.90–1.05
8	1.50–1.75	1.75–2.00	1.5	1.05–1.20
9	>1.75	>2.00	<1.0	>1.20

Basis: Survival. Percentage of plants alive as a proportion of the percentage of plants of the check (IR2152-26-3-5-2) alive when the check variety was about 50% dead
Na transport. The shoot sodium concentration (per unit fresh weight) divided by that of the check variety
Tissue tolerance. The LC_{50} (mmol Na g^{-1} ethanol-insoluble dry wt) corresponding to 50% loss chlorophyll, estimated to the nearest 0.5
Leaf-to-leaf compartmentation. The ratio of the increase in quantitiy of sodium in leaf 4 between two harvests to the increase in quantity of sodium in leaf 3 between the same two harvests, for an interval during which the quantity of sodium in leaf 3 increased from approximately 1 to 3 µmol
Plant vigour. The average of shoot fresh and dry weights, and leaf length, relative to a reference variety (IR28)
Where scores were normalised on the check variety, the score boundaries were arbitrarily set such that the frequency of different scores was reasonably uniform

Screening for tissue tolerance

Tissue tolerance describes the cellular component of salinity resistance, i.e. varietal differences in tissue damage for the same concentration of salt in the leaf tissue. We have interpreted this as reflecting differences in apoplastic salt load in accordance with the Oertli (1968) hypothesis. The screening procedure is based on correlating leaf chlorophyll concentration (as a marker for metabolic damage) with leaf Na concentration, as described by Yeo and Flowers (1983).

Seedlings were transplanted at 7 days and salinised at 14 days in the controlled environment conditions described above. The individual variation in salt uptake within any accession (Flowers and Yeo 1981; Yeo et al. 1988) was exploited to generate a range of individual leaf Na concentrations at a single time, which varied according to the overall sensitivity of the accession to salt. At harvest, for each accession, the third leaf from 100 salinised individuals, plus 20 unsalinised controls, was excised and chlorophyll was extracted at 80 °C in 80% ethanol for 15 min. Chlorophyll was measured in this extract spectrophotometrically (Pye Unicam SP1800 or PU8620). The same ethanolic extract was then acidified with acetic acid to a final concentration of 100 mol m^{-3}, and the leaf was re-extracted for Na measurement by atomic absorption spectrophotometry. Finally, the leaf was dried to give its ethanol-insoluble dry weight.

The concentration of sodium in the leaf tissue, which corresponded to a 50% loss of chlorophyll (the LC_{50}), was calculated from the inverse correlation between sodium and chlorophyll concentrations (Yeo and Flowers 1983), and converted to a 1 (tissue-tolerant) to 9 (tissue-sensitive) scale (Table 1).

Screening for leaf-to-leaf compartmentation

The gradient of sodium from leaf to leaf is indicative of the ability of a variety to sustain at least the younger leaves at a low salt concentration; the oldest leaf fills up with Na first and then, after differing lag periods, the Na concentration increases in progressively younger leaves (Yeo and Flowers 1982). Seed was germinated and transplanted at 7 days, and plants were salinised with 100 mol m^{-3} NaCl at 22 days. Four harvests were made after 1, 2, 3 and 4 days.

At each harvest, the first four leaves of each of 30 individual plants were excised and extracted in acetic acid for Na determination by atomic absorption spectrometry, and the quantity (Q_{Na}) of sodium in each leaf blade was calculated. The parameter evaluated was the increse in Q_{Na} in leaf 4 between two harvests, divided by the increase in Q_{Na} in leaf 3 during the same period. Leaves are numbered in order of intiation; leaf 3 is the older and 4 the younger. The pair of harvests was chosen over which Q_{Na} in leaf 3 increased from approximately 1 to 3 µmol, so that varieties were compared on a common basis; taking four harvests increased the chances of producing these conditions in material covering a range of salt resistance. Scores from 1 to 9 were assigned; varieties with the lowest increase in salt content in leaf 4 relative to leaf 3 were given the highest score (1), indicating a high degree of compartment (Table 1).

Screening for plant vigour

Seed (50 per accession) was direct-sown onto sites on floating perspex grids in the same greenhouse system described for survival screening. After 7 and 14 days, shoot fresh and dry weights and average shoot length were recorded, and were divided by the mean value for the replicate reference (IR28) sites in each tank. The data for each parameter was divided into nine equal class intervals and assigned a score on the 1–9 range (1 being the largest or longest); this division is arbitrary and the data are not shown. The three scores for each variety were averaged, and this average is the vigour score assigned to each accession.

Results

Survival was used as a quantification of overall phenotypic performance; this provides a link with field reputation and with the varietal assessment during mass screening trials (which is by the subjective evaluation of salinity damage to leaves; IRRI 1976). As a measure of reproducibility, a survival trial was repeated three times with the same group of varieties. The replications were, firstly, at opposite ends of temperate year and, secondly, at the same time of year, but 2 years apart. The scores (derived as Table 1) show good uniformity (Table 2) and so the system appears to cope well with external fluctuations; over the three trials, varietal differences accounted for 88% of the variation in assigned scores. Survival screening has now been carried out for over 250 accessions.

Some 150 genotypes, which had been assessed for survival, were screened for shoot Na concentration. There was a substantial varietal variability in both attributes. Although it is the excessive transport of NaCl to the shoot that underlies salt damage in rice, the correlation between varietal shoot Na concentration and survival scores is poor (Fig. 1, Table 3), and the partial re-

Table 2. Reproducibility of survival screening at different times of the year. Survival scores were obtained at three different trials begun on 6 August 1984, 18 February 1986 and 6 August 1986. Analysis of variance: varieties SS = 214.8 (88%); residual SS = 30.0; $F = 15.2$; $P \ll 0.001$; $LSD_{P=0.05} = 1.46$

Acc [a]	Designation	Scores			Mean
041	IR14581-52	4	5	4	4.3
042	IR14581-66	9	9	9	9.0
043	IR14632-2-3	3	5	4	4.0
044	IR14753-49-2	4	3	3	3.3
045	IR15314-30-3-1-3	5	5	5	5.0
046	IR15324-117-3-2-2	3	4	3	3.3
047	IR15527-3-2-2	9	5	7	7.0
048	IR17494-32-3-1-1-3-3	4	2	3	3.0
049	IR17521-2-7-2-2-2-2	4	3	4	3.7
050	IR17525-7-20-3-2	4	3	3	3.3
051	IR18272-27-3-1	7	6	6	6.3
052	IR19392-113-2	9	7	8	8.0
053	IR19660-11-2-2-3-2	2	5	3	3.3
054	IR19660-23-2-1	3	5	4	4.0
055	IR19672-195-2-2	4	6	6	5.3
056	IR20	8	6	7	7.0
057	IR2058-85-3-3	3	4	4	3.7

[a] Sussex accession number

Table 3. Correlation matrix for survival (overall performance) and individual traits based upon all available data. Correlation (r^2), sign (+ or −) of the regression coefficient and number of paired observations. Scatter diagrams of survival/leaf Na and survival/vigour scores are illustrated in Figs. 1 and 2

	Vigour	Shoot Na	Tissue tolerance	Leaf-to-leaf
Survival	0.392 (+) ($n = 142$)	0.252 (+) ($n = 148$)	0.048 (−) ($n = 47$)	0.175 (−) ($n = 21$)
Vigour		0.177 (+) ($n = 162$)	0.284 (−) ($n = 40$)	0.213 (−) ($n = 21$)
Shoot Na			0.003 (−) ($n = 21$)	0.026 (−) ($n = 21$)
Tissue tolerance				0.038 (+) ($n = 21$)

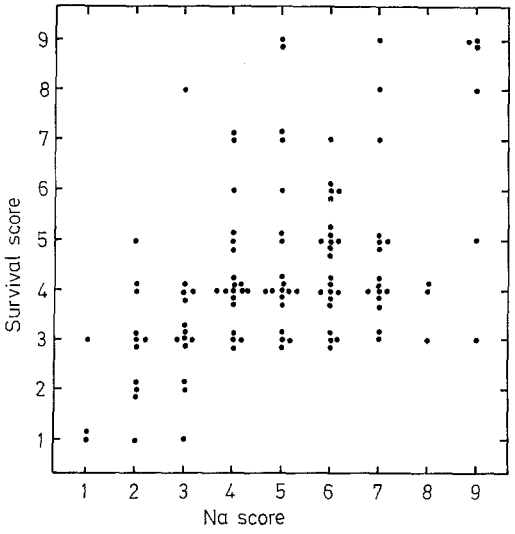

Fig. 1. Scatter diagram for scores for survival and shoot Na concentration. Each point represents a genotype. Scores range from 1 (good survival or low shoot Na concentration) to 9 (poor survival or high shoot Na concentration)

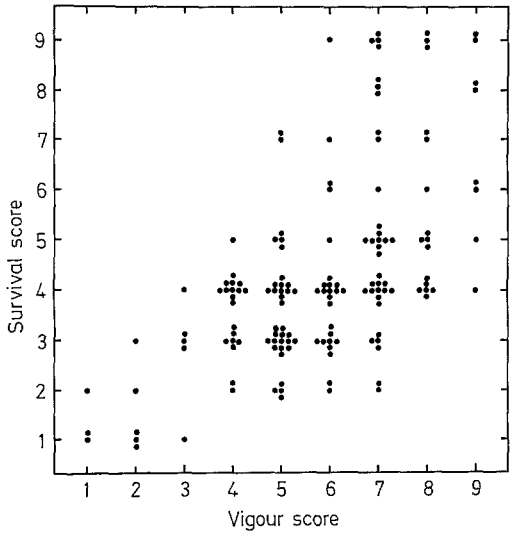

Fig. 2. Scatter diagram for scores for survival and plant vigour. Each point represents a genotype. Scores range from 1 (good survival or high vigour) to 9 (poor survival or low vigour)

gression coefficient was small and insignificant. Varieties with low Na concentrations survived well whilst those with very high Na uptake survived poorly (Fig. 1), but there remains a large range of shoot Na concentration for similar survival and, conversely, a large range of survival for similar shoot Na concentration. This could arise if the initial phenomenon of Na uptake is modulated by factors such as vigour, leaf-to-leaf compartmentation and tissue tolerance. The data for the genotypes that have been assessed with all five screening procedures is shown in Table 4. It is worth noting that, amongst this sample of 21 genotypes, the correlation between survival and vigour is stronger, and between survival and shoot Na concentration weaker, than when a much larger sample size is considered (Table 3).

Plant vigour reflects, to a large extent, the degree of dwarfing; modern rice varieties generally include dwarfing genes to increase harvest index and reduce lodging. These genes have major effects on morphology, which are expressed clearly at early seedling developmental stages (Flowers et al. 1989; Fig. 1). Of the parameters measured, vigour had the highest correlation with survival and the largest partial regression coefficient (Fig. 2, Table 3).

Table 4. Scores for overall performance (survival), and individual traits, in a range of genotype used in the studies. Genotypes are ranked according to their overall performance. The regression equation is: survival = −2.04 + 0.772 (vigour) + 0.074 (shoot Na) + 0.252 (tissue tolerance) −0.040 (leaf to leaf)

Designation	Overall (survival)	Individual traits				Average
		Vigour	Shoot Na	Tissue tolerance	Leaf to leaf	
Cheriviruppu	1	1	2	7	8	4.50
Nona Bokra	1	2	1	7	6	4.00
Pokkali	1	1	3	9	8	5.25
IR4630-22-2-5-1-3	2	7	2	1	4	3.50
IR8236-B-B-71-1-3	2	5	3	5	4	4.25
Damodar	3	4	2	7	8	5.25
IR10198-83-4-2	3	6	3	7	6	5.50
IR15324-117-3-2-2	3	6	9	7	6	7.00
IR4595-4-1-13	3	6	3	5	7	5.25
IR5	3	4	6	3	9	5.50
IR58	3	5	7	5	5	5.50
IR8241-B-B-86-2	3	5	7	4	8	6.00
Pelita I-1	4	6	5	6	6	5.75
IR22	4	6	3	5	9	5.75
IR10167-129-3-4	5	8	6	2	3	4.75
IR10168-93-4	5	6	2	4	5	4.25
IR24	5	7	4	5	2	4.50
IR36	5	7	4	5	9	6.25
IR20	6	9	4	1	6	5.00
IR40	6	9	6	6	6	6.75
IR26	9	8	7	7	2	6.00
Relationship to survival						
correlation (r^2)		0.645	0.177	0.048	0.175	0.100
partial regression coefficient		0.772	0.074	0.252	0.040	
and its probability		0.001	0.587	0.113	0.772	

A very wide range of tissue tolerance was found with LC_{50} values ranging from less than 1.0 to more than 5.0 mmol of Na per gram of ethanol-insoluble dry weight. There was no simple correlation with survival, and the partial regression coefficient was only one-third of that for vigour. Interaction with other characters confounded the issue. There was an inverse correlation between tissue tolerance and vigour; the tall traditional varieties have leaf tissue that is salt sensitive, whilst the most tissue-tolerant lines come from the dwarf category, often from lines that have very poor survival and/or shoot Na scores. While a variety could be an important source of the genes for tissue tolerance, it would be rejected in any mass screening trial, because this factor alone is not enough to dominate its overall performance (Tables 3 and 4).

There was also substantial varietal variation (at least fivefold) in the ability of leaf-to-leaf compartmentation of sodium. This trait also revealed a negative correlation with vigour, marginally significant at $P = 0.05$ and, hence, there was a trend ($P = 0.059$) for leaf compartmentation to be negatively associated with survival. The partial regression coefficient was small and insignificant.

Discussion

There has been considerable effort directed at selection for salinity resistance in rice, based on extensive germ plasm screening; more than 90,000 varieties and lines have been assessed at the International Rice Research Institute alone (Akbar 1986). As a result of mass screening trials, the cultivars identified as donors of salt resistance have been landraces of the traditional, non-dwarf plant type. Although some lines derived from these donors have a degree of resistance, a continuing problem is that the level of salt tolerance so far found in rice is inadequate (Ponnamperuma 1984). We argue from the data presented that the potential for salinity resistance in rice is not limited to the level of phenotypic expression discovered through screening.

Knowledge of the physiological effects of salinity on plants, both crop species and those species which are native to saline environments, has shown that there are no simple answers to the questions of how plants are damaged by salt and of how they survive it (Flowers et al. 1986; Greenway and Munns 1983; Munns and Termaat 1986). Salinity affects almost all plant processes because of effects on soil properties, because of the osmotic ef-

fects of high ionic concentrations, because of competitive interference with nutrient uptake and because of toxic effects within the plant tissue (Yeo and Flowers 1989). We may, therefore, expect that a range of adaptive changes will be required if a salinity-sensitive species is to be grown satisfactorily in saline conditions. A minimal requirement is to maintain ion concentrations that are compatible with metabolism in the cytoplasm of the cells of at least the younger leaves; low shoot salt concentrations, vigour to provide dilution of salt concentrations by growth, compartmentation of salt in older rather than younger leaves and tolerance to salt within the tissue are all traits that are expected to be additive in achieving this (Yeo and Flowers 1986).

As far as rice is concerned, the general observation to be made from the screening results is that there was no grouping of these resistance traits in single genotypes. It is apparent from Table 4 that there is useful varietal variation for each of the characters investigated, and that these characters are scattered amongst different varieties, many of which do not have good overall resistance to salinity. It is also apparent that the varieties that are classed as the most resistant (those with a survival score of 1 in Table 4) do not have high scores in more than two of the four traits considered. Since these varieties (such as Nona Bokra) are still the standard by which the salt resistance of others is judged, we may conclude with some confidence that no extant rice genotype possesses a more favourable combination of salt resistance traits. These observations support the view that the salinity resistance of rice could be increased above the present level of observed phenotypic expression through the crossing of existing genotypes, provided that linkage is not a bar. We have argued that, because salinity is marginal to the ecological range of rice, natural selection pressure favouring a genotype with a combination of salinity resistance traits has been limited (Yeo and Flowers 1989). In the absence of such a genotype, vigour has been the one factor with the largest influence on survival, and the presence or absence of dwarfing genes is superimposed upon this.

The results have far-reaching implications for the methods used to select parental material in plant breeding programmes aimed at improving the resistance to salinity and, perhaps, to other environmental stresses to which a species is not widely exposed in the natural environment. Some overall measure of plant performance, be it survival or visual assessment of salt damage, is the criterion in general use. Characteristics would normally be chosen on their correlation with overall performance. In our data, survival and some physiological traits were at best weakly, and at worst negatively, correlated. For example: tissue tolerance represents the amount of salt which can accumulate in the leaf tissue for a given degree of damage; some varieties could tolerate five time as much salt in their leaves as others. It is difficult to see how this could be a disadvantage in resisting salinity, yet tissue tolerance has a weak negative correlation with survival and an insignificant coefficient in multiple regression analysis. Either the traits are not, after all, relevant to salt resistance (possibly because of adverse genetic linkages), or survival is not a useful criterion by which to evaluate potential parents.

There are three fallacies associated with the use of correlation and multiple regression to analyse this data, as a result of which we question whether overall performance is the appropriate basis on which to select parents.

(1) Plant vigour is not a single variable. Our sample of rice genotypes (and this is likely to be true of any collection of genotypes in any rice breeding programme) consists of two distinct populations: those which have and those which have not been developed by incorporation of dwarfing genes. The effects of the dwarfing gene(s) are very far-reaching: the dwarf and non-dwarf plant types differ not only in growth rate, they grow in very different ways. The dwarf plant type is characterised by large numbers of small, erect leaves, a high lamina-to-sheath ratio, high tillering rate and rapid leaf development. In contrast, the non-dwarf plant type has fewer tillers and typically develops more slowly into a plant with a smaller number of large, arching leaves with large sheaths. As a consequence of the dwarfing gene (whether direct or expressed through resultant differences in morphology), the two types also differ physiologically, e.g., in water use efficiency (Flowers et al. 1989). The different numbers, sizes and growth rates of leaves, and the different balance between leaves and sheaths will inevitably affect the distribution of ions within the plant. Thus, vigour is confounded by the accompanying changes in morphology caused by dwarfing genes.

(2) The variables are not independent. Shoot Na concentration is the result of the rate of Na transport and the growth of the shoot; if the shoot grows faster, then the concentration resulting from the same amount of transport will be lower than if the shoot grows more slowly. Shoot Na concentration is dependent upon both Na transport and vigour. It is not, therefore, valid to allocate a proportion of the variation in survival to either shoot Na concentration or vigour in a single or multiple regression. Some characteristics are certainly associated with the two different plant types (whether they are linked genetically or are consequences of differences in morphology): both leaf-to-leaf compartmentation and tissue tolerance show some inverse correlation with the non-dwarf plant type. Since plant type dominates the vigour, these characteristics are not, then, independent of vigour.

(3) There is no expectation of a positive correlation between any one independent character and survival. Good performance in one character is as likely as not to be accompanied by bad performance in another.

A distinction must be made between using screening based on overall performance (1) to locate resistant varieties among a large collection of germplasm, and (2) to select parental material from which to breed resistant varieties. In the absence of any genotype with a favourable combination of traits, the vigorous growth form of the non-dwarf has the greatest effect upon the chances of survival. It would have been a prediction from our data that mass screening would result in the selection of a non-dwarf with little other in its favour with respect to the resistance of salinity; it has. It is also likely that it is this vigour that has been exploited in such selection of landraces as has been made by farmers for growth in salt-affected areas. This benefit will be lost in breeding because the plant characters that contribute to the survival of salinity by the non-dwarves are agronomically unpopular, and nearly all of the elite breeding material, which is used to donate agronomic traits, is dwarfed to at least some extent.

The alternative which we advocate, breeding with physiological characters, is not a simple solution to increasing salt resistance, but its value must be viewed in relation to the potential of other available methods: cell and tissue culture, wide-crossing and mutagenesis (Ponnamperuma 1984). The success at regenerating salt-tolerant plants from salt-tolerant cell lines has been very limited (Table 4 in Yeo and Flowers 1989). As far as wide-crossing is concerned, the potential for importing salt tolerance from related species appears poor, in contrast with some other crops. Eight *Oryza* species covering the A, B, C, D, E and F genomes all survived salinity less well than the *O. sativa* landrace (Nona Bokra) used as the resistant check (Akbar et al. 1987). We are aware of only one salt-tolerant species related sufficiently closely to *O. sativa* to support some hope of wide crossing, *Porteresia coarctata* Tateoka. However, *P. coarctata* is a perennial and depends also upon the anatomical adaptation of salt glands to remove excess salt from the leaves (Bal and Dutt 1986; Flowers et al. 1990). Although this does not disqualify *P. coarctata* as a donor of genetic information for salt tolerance, it does suggest that incorporation of such information into cultivated rice will not be a straightforward matter. Finally, some variants of *O. sativa* cv Taichung 65 produced through mutagenesis were more resistant to salt than the parent, suggesting that this is a possible route towards increasing salt tolerance (Chaudhry et al. 1987); however, these variants still did not survive salinity better than Nona Bokra.

There are problems associated with using physiological traits in plant breeding, though it should be noted that the difficulties arise in part from the logistics of using physiological markers (which could be overcome), not from the principle of selecting for physiological traits. A serious difficulty, as is apparent from many of the methods used herein, is that many individuals are needed to obtain a single assessment; consequently, selection of promising individuals, and hence lines, in the early generations, is virtually impossible. The one solution to this is the identification of biochemical markers that are linked to the physiological traits in question. This would both replace the need for routine physiological measurements and make possible the identification of the genetic potential of individuals. Possible methods include biochemical characterisation, such as been used to describe membranes that compartmentalise salt effectively (Leach et al. 1990), isozyme loci (Ranjhan et al. 1988) and restriction fragment length polymorphism, since the rice genome has recently been mapped (McCouch et al. 1988).

It is highly pertinent that donors for each physiological trait can be found from amongst varieties that are already agronomically acceptable in terms of yield potential, disease resistance and photoperiod insensitivity. The heritability and combining ability of traits contributing to salinity resistance are currently being investigated through multiple and diallel crosses.

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