BREEDING OF DISEASE RESISTANT TOMATOES IN JAPAN

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In 1977 Japanese seed growers listed 147 varieties as resistant to disease(s). These include 143 resistant to Fusarium wilt, 57 to leaf mold, 53 to root-knot nematode and 40 to TMV (Tobacco Mosaic Virus). The total for these four diseases is 293 and there is a score of 60 for other diseases. The grand total of 353 is more than twice the number of listed varieties and indicates that, on average, each variety possesses resistance to more than two diseases. Table 1 shows the distribution of resistance in these varieties. Of the 53 varieties with single disease resistance 50 are resistant to Fusarium wilt and 3 to leaf mold. No varieties have resistance to TMV or root-knot nematode alone.

Table 1. Number of tomato varieties with resistance to Tobacco Mosaic Virus, leaf mold, Fusarium wilt and root-knot nematode.

Resistance	Disease *		Number of varieties
Single	TMV		0
	C		3
	F		50
	M		0
		Total	53
Double	TMV.C		0
	TMV.F		12
	TMV.M		0
	C.F		19
	C.M		1
	F.M		22
		Total	54
Triple	TMV.C.F		10
	TMV.C.M		0
	TMV.F.M		6
	C.F.M		12
		Total	28
Quadruple	TMV.C.F.M		12
		Grand Total	147

* TMV : Tobacco Mosaic Virus C : leaf mold
F : Fusarium wilt M : root-knot nematode

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As parental lines with combined resistance became available, hybrid varieties with combined resistance were produced. Furthermore, by crossing two lines with combined resistance, the number of diseases controlled could be increased. At present several hybrid varieties possess resistance to six diseases though they still have yield and quality defects. These must be resolved by further breeding because the use of resistant varieties will be essential for tomato growing in the future.

One difficulty usually encountered when breeding tomatoes for Japan is that the desired characters in tomato fruit for fresh use are rather different from those in most foreign countries. The skin color must be colorless to give pink fruit rather than yellow as in red-fruited varieties. The fruit must be of the greenback type (green color remains around the calyx) without uniform ripening It seems that these requirements are mainly due to the conservatism of the wholesale market rather than the consumers'. The consequence is that resistant varieties introduced from abroad can not be used as received; they must be modified to suit the Japanese style.

1. CMV (Cucumber Mosaic Virus)

No resistant commercial varieties are available at present although breeding work is in progress. Several TMV resistant tomato varieties including 'Ohio M-R 9', 'Ohio M-R 12' and 'L-253' showed delayed appearance of symptoms or less severe symptoms compared with fully susceptible varieties. The degree of resistance was affected by many factors including inoculation methods, plant stage and environment (Kuniyasu and others, 1975). Several lines have been bred from the cross between 'L-253' and 'Ohio 690712'. However, the resistance of these breeding lines and the original resistant varieties proved subsequently to be dependent on CMV isolates inoculated from the following experiments (Yasui and Yamakawa).

Eleven CMV isolates sampled from different host crops and tested for their pathogenicity on tomato variety 'L-253' were divided into four groups. 1) 'L-253' was less affected than susceptible varieties which were severely affected. 2) both 'L-253' and susceptible varieties were severely affected, although 'L-253' showed somewhat less symptoms. 3) 'L-253' was equally or more severely affected than susceptible varieties. 4) either 'L-253' and susceptible varieties were less affected. The number of isolates in each group was 4, 2, 2 and 3 respectively. In order to clarify whether pathogenicity of an isolate was altered by its passage through a particular host, CMV isolates were used to inoculate the tomato cultivars, 'Ohio M-R 12' which is resistant to some CMV isolates and 'Kyoryoku Beiju' which is fully susceptible, after separate passage through both cultivars and CMV transfer to 'KY-57' tobacco plant affected leaves which were used as the inoculum for the tests. The results showed clear effects of the passage. The isolates which affected 'Ohio M-R 12' to a much lesser extent than 'Kyoryoku Beiju' before passage or after the passage through 'Kyoryoku Beiju' affected 'Ohio M-R 12' even more severely than 'Kyoryoku Beiju' after passage through 'Ohio M-R 12'. Thus the resistance of 'Ohio M-R 12' was completely negated by the passage. Although we must admit the diversity and dynamic status of CMV populations indicated by these experiments and consequently the difficulties likely to be encountered in resistance-breeding work, it does not seem impossible to breed effective resistant varieties if we can assume the existence of stabilizing selection of CMV.

2. TMV (Tobacco Mosaic Virus)

In 1973 five resistant varieties with the gene Tm including 'Raiden' and 'Tukama' were released from the Kikyogahara Station. However, as early as 1974 these varieties were reported to be infected with TMV in several major tomato-producing areas like Shizuoka and Chiba which suggested the occurrence of a new strain. The use of other resistance genes, Tm-2 and Tm-2^2, became necessary. At present about one quarter of the TMV resistant varieties have Tm-2 or Tm-2^2. The gene Tm-2^2 was introduced to Japanese varieties mainly from American varieties like 'Ohio MR 9 and MR-12', and it now appears to be carried by between five and ten of the Japanese varieties recently released. The use of Tm-2 has been rather limited so far because of its tight linkage with nv. The three lines 'IRB 301-30, 31 and 32' bred by the

author from interspecific hybridization between *Lycopersicon peruvianum* P. I. 126944 and the cultivated tomato possess Tm-2 without linkage to nv. Although Tm-2 and Tm-2^2 confer much higher resistance than Tm, they tend to cause necrosis of the growing point, stem or fruit especially in heterozygous plants grown at high temperature. In order to avoid this, future TMV resistant varieties will have to be homozygous for Tm-2 or Tm-2^2 or possess gene Tm as well. Several seed growers and the Kikyogahara Station have already bred such resistant varieties.

Another difficulty may be encountered when tomato grafting is carried out. As mentioned later several resistant root-stocks are used in Japanese tomato growing to give protection against bacterial wilt, brown root rot and other diseases; they are mostly TMV susceptible. When Tm-2 or Tm-2^2 scions are grafted on to them and the root-stocks become infected with TMV, the virus easily moves to the scions and induces severe necrosis in growing point, leaf, stem and fruit. In contrast when a TMV resistant (Tm-2 or Tm-2^2) root-stock is used for a TMV-susceptible or TM type resistant scion any TMV infection of the scion moves to the stock, affects the root and causes the whole plant to wilt. This wilting of susceptible scions grafted on to TMV-resistant 'KNVF' (presumably the resistance is confered to by Tm-2^2 gene) has actually been observed in several places. Similar wilting is also observed when a susceptible scion is treated with a symptomless mutant strain of TMV, L 11 A, for cross protection. This indicates that even a symptomless strain can affect Tm-2 or Tm-2^2 type resistance, when transmitted via a graft union.

3. Bacterial canker (*Corynebacterium michiganense*)

'Okitsu Sozai 1' was selected from an interspecific hybrid between *L. hirsutum* and *L. esculentum*, and released to tomato breeders including private seed growers as a breeding stock for bacterial canker resistance (Kuriyama and others, 1974). The line is still segregating for horticultural traits such as fruit size and color, and needs to be improved further. The mode of inheritance of the resistance is not clear but it seems to be complex.

4. Bacterial wilt (*Pseudomonas solanacearum*)

No resistant cultivar is available with satisfactory marketing quality, so grafting to resistant root-stocks is carried out for heavily infested soils. 'BF Okitsu 101', a resistant line used as a root-stock, was originally introduced from North Carolina State University as 'NC 1953-64N' and was subsequently selected for Fusarium wilt (race 1) resistance (Kotani and others, 1970). #7998 introduced from the University of Hawaii in 1975 has shown higher resistance than 'BF Okitsu 101' and is now being tested as a root-stock.

5. Brown root rot (*Pyrenochaeta lycopersici*)

This disease was identified quite recently: it has increased as tomato growing under glass or plastic has been extended to the cooler season. Grafting culture using a resistant root-stock 'KNVF' is popular. 'KNVF' is a hybrid between *L. esculentum* and *L. hirsutum* var. *glabratum*, the seeds being imported from the Netherlands and the United Kingdom. 'Shinko Ichigo' released from the Takii Seed Company is considered to be a hybrid with similar parentage.

6. Fusarium wilt (*Fusarium oxysporum* f. *lycopersici*)

At present more than 130 varieties are available with resistance to race 1. Sugahara and Suzuki (1966) found race 2 (they termed it race J-2) in the Fukuoka region, but it has never caused serious damage to tomato crops in Japan. Yamamoto and others (1974) found a new soil-borne disease and identified the causal agent as *Fusarium oxysporum* f. *lycopersici* from its morphology and pathogenicity. They termed it race J-3 because varieties resistant to races 1 and 2 were susceptible to this race. However the symptoms are quite different from those caused by J-1 and J-2. J-3 causes root- and crown-rot but not much browning of vascular bundles of the stem while J-1 or J-2 cause heavy browning of the vascular bundles but not

root rot. The favorable temperatures are also different. J-3 is more severe in cooler seasons, J-1 and J-2 in warmer seasons. Thus J-3 can be seen as the cause of a new disease rather than as a new physiological race The disease is commonly called 'root-rot wilt' in Japanese. Komada found resistant plants in *L. peruvianum* P. I. 126944 which had been successfully hybridized after chronic gamma irradiation with *L. esculentum* by the author. 'IRB 301-30 and 31' selected from these hybrids have been shown to possess resistance to root-rot disease which is increasing in Ohio, U. S. A. and Canadian green-house soils (Dr. James D. Farley, personal communication). Further backcrosses are being made to Japanese varieties in order to improve the resistant cultivars

A single dominant gene confers the resistance, but several other modifiers are likely to be involved.

7. Late blight (*Phytophthora infestans*)

The Kikyogahara and Okitsu Stations have long been concerned with late blight resistance. Both stations have used the resistant line 'West Virginia 700'. 'WV36' was also used at Okitsu.

8. Leaf mold (*Cladosporium fulvum*)

Among 57 resistant varieties (almost all are hybrids) available in Japan, at least 13 have been bred using 'Okitsu' lines (No.7-No.12) as a direct or indirect parent. The 'Okitsu' lines derive their resistance from 'Improved Bay State'. At least four varieties owe their resistance to 'STEP 390'. The source of resistance in other varieties is not known because they were mostly bred by private seed growers. However, most of them seem to owe their resistance to American varieties or lines including 'Improved Bay State', 'Waltham Moldproof Forcing' and 'STEP 390'.

Recently those varieties with gene(s) from 'Improved Bay State' have frequently been reported to suffer from leaf mold in several major tomato-producing areas. Kishi and Abiko confirmed the differentiation of new races. The author has been conducting a backcross breeding programme using the interspecific hybrid previously mentioned, and has obtained several breeding lines which have not yet been released. The lines are resistant not only to Japanese races but also to races 1.2.3, 4, 1.2.4, and 2.3.4 as tested at the Institute for Horticultural Plant Breeding, the Netherlands (Ir. I. W. Boukema, personal communication, 1973). The lines are being backcrossed to Japanese varieties.

9. Leaf spot (*Stemphylium lycopersici*)

Fifteen varieties are reported to be resistant to leaf spot. They are resistant also to Fusarium wilt and several other diseases. American varieties resistant to leaf-spot and Fusarium wilt, including 'Anahu', 'Manalucie', 'Florida MH-1', 'Tropic' and 'Walter', were often used as a breeding stock in Japan. Although they were used mainly for Fusarium wilt resistance, it has been shown that the gene Sm which confers resistance to leaf spot is tightly linked with the gene I for Fusarium resistance. Sm seems to have been introduced to Japanese varieties together with Fusarium-wilt resistance.

10. Verticillium wilt (*Verticillium albo-atrum*)

The disease was found in 1972 in Tokyo and several other districts, and the Tokyo Agricultural Experiment Station started breeding work immediately. It has recently bred several 'NFVR' lines with combined resistance to root-knot nematode, Fusarium wilt and Verticillium wilt, using 'Tropi-red' and 'Loran Blood' as sources of resistance.

11. Root-knot nematode (*Meloidogyne incognita* var. *acrita*)

'NFR-1, 2 and 3' derived the resistance gene Mi from 'Anahu', and were used as parents of hybrid varieties including 'Azuma' and 'Raiden'. Many other resistant varieties have been released mainly by seed growers. Their source of resistance is not clear but it seems that there

are some differences in resistance between the varieties. The varieties currently available are resistant to *M. incognita* var. *acrita* but not to *M. hapla*. A population of *M. incognita* var. *acrita* which infected a variety with the 'Anahu' -type resistance, was reported from Chiba and a breeding source with high and stable resistance is being sought.

Discussion

G. S. Khush, The Philippines: I understand that bacterial wilt resistant gene is closely linked to small fruit size. As far as I know, all the breeding lines with resistance to bacterial wilt have small fruit. In your breeding program, have you been able to break this linkage?

Answer: It is true that most of the varieties with high resistance to bacterial wilt have small fruit. However several varieties introduced from the Vegetable Research and Development Centre have fairly large fruit in spite of having high resistance. Therefore, I do not think that it is impossible to breed large fruit resistant varieties.

G. S. Khush, The Philippines: As you know, there are many genes for male sterility in tomatoes. Some are linked to seedling marker genes. As an example, there is an anthocyanin-less gene (a) on chromosome 11 which is closely linked to a gene for male sterility. This favorable combination may be exploited in hybrid seed production using a tertiary trisomic as described by Khush and Rich in an article published in the Canadian Journal of Genetics and Cytology in 1968.

Answer: Thank you for your suggestion. I have once introduced male sterile lines linked with anthocyaninless gene and I have heard that a seed grower was using the line to produce a parent of a F1 hybrid. However a parent of a F1 hybrid must have above all good combining ability. Perhaps, for that reason, I have not yet heard that the seed grower was using the ms line at the present time. I would like to take this opportunity to mention that almost all the tomato cultivars in Japan are F1 hybrids and consequently, when a new disease resistance is introduced, the line should have a superior combining ability other than disease resistance. The breeding work is not that easy and does not consist of a simple transfer of a few resistance genes.

G. S. Khush, The Philippines: Have you used Solanum Pennelli as a source of genes for disease resistance in your program?

Answer: I introduced S. Pennelli made available to me by Professor Rick of the University of California in Davis. I checked it for resistance to IMV. The resistance seemed to be of the tolerant type but I have not yet had the opportunity of trying it for other diseases.

S. Kamimura, Japan:

1. What is the relationship between race 3 of Fusarium disease which is found in the United States and J3 which is present in Japan?

2. About ten years ago, a method of inoculation using an attenuated virus was developed by Dr. Oshima of the Institute for Plant Virus Research to control TMV virus disease. Recently it has been shown that this method was being applied in Hokkaido, Chiba and Shizuoka and that it produced good results in controlling virus diseases. I would like to have your opinion on this problem in relation to the advancement of TMV resistant breeding work.

Answer: New diseases similar to that caused by J3 have been found in Ohio and Florida, in the United States and also in Canada. In Ohio, they are considered as new entities and called Crown and Root Rot diseases. The fact that our breeding line resistant to J3 also shows resistance to the Wilt disease present in Ohio and Florida suggests that the diseases were caused by the same pathogenic agent. The use of attenuated virus for cross protection from virulent TMV is now being applied very successfully. The use of resistant varieties, however, must be pursued because in the long run varietal resistance is the cheaper and safer way to confer protection.