Theor Appl Genet (1996) 93: 494–502

R. Cheng · A. Saito · Y. Takano · Y. Ukai

Estimation of the position and effect of a lethal factor locus on a molecular marker linkage map

Received: 14 August 1995 / Accepted: 1 December 1995

Abstract In the mapping of DNA markers the distortion of segregation of marker genotypes is often observed, which may be caused by a lethal factor acting in filial generations derived from distant crosses. A method is presented for estimating the recombination values between a lethal factor locus and neighboring molecular markers, and the relative viability or fertilization ability of gametes or zygotes affected by the lethal factor in an F_2 population using the maximum likelihood method and the expectation conditional maximization (ECM) algorithm. Three selection models of gamete or zygote were considered, and the most likely one was determined by goodness of fit of the observed frequency of the phenotypes to the expected ones under the models. The method was applied to segregation data of molecular markers of an F_2 population consisting of 144 individuals derived from a cross between an *Indica* and a *Japonica* rice variety. The presence of a lethal factor locus (*L*) located on chromosome III that caused partial gametic selection in both the male and female sides was suggested. The locus *L* was tightly linked to RFLP marker number 23 of the RFLP linkage map of Saito et al. (1991a), and the fertilization chance of a male or female gamete possessing the lethal factor was, on average, 41.5% of that of the normal gamete.

Key words Mapping · Segregation distortion · Molecular marker · Recombination value · Maximum likelihood method

Communicated by G. Wenzel

R. Cheng (✉) · Y. Takano · Y. Ukai
Laboratory of Biometrics, Division of Agriculture and Agricultural Life Sciences, The University of Tokyo, Yayoi 1-1-1, Bunkyo, Tokyo 113, Japan

A. Saito
Kyushu Agricultural Experimental Station MAFF, Nishigoshi, Kikuchi, Kumamoto 861-11, Japan

Introduction

During the last decade the molecular biology techniques of restriction fragment length polymorphism (RFLP) and randomly amplified polymorphic DNA (RAPD) have been widely used to construct linkage maps of the genomes of numerous crop species. In order to obtain higher polymorphisms, crosses between wild species and cultivars, between subspecies, and even between species have been made for linkage analysis (McCouch et al. 1988; Paterson et al. 1988, 1990, 1991; Saito et al. 1991a; Schon et al. 1991; Causse et al. 1994). Segregation distortion of marker genotypes is often encountered in such distant crosses (Wendel and Parks 1984; Torres et al. 1985; McCouch et al. 1988; Paterson et al. 1988; Saito et al. 1991b; Lyttle 1991; Schon et al. 1991; Paterson et al. 1991; Zivy et al. 1992; Causse et al. 1994). One of the major reasons of this distortion may be the elimination of gametes or zygotes, which is controlled by a lethal factor located in the neighboring region of the marker.

In rice, Iwata et al. (1964) were the first to report linkage between a lethal factor (they called it gametophyte gene) and two morphological markers. They estimated recombination values and the differential fertilization ability of the male gametes using F_2 and F_3 segregation data. Similar methods have been adopted by other researchers for analyzing linkage between lethal factors and morphological or isozyme markers in rice (Nakagahra 1972; Nakagahra et al. 1972; Mori et al. 1973; Nakagahra et al. 1974; Maekawa et al. 1981; Maekawa 1982; Kinoshita and Takamura 1984; Maekawa and Kita 1985; Lin and Ikehashi 1993).

Lorieux et al. (1995a, b) recently presented a maximum-likelihood method for mapping genetic markers showing aberrant segregation in a backcross and an F_2 population. They compared three cases, i.e., codominant versus codominant, dominant versus dominant, and dominant versus codominant markers. However, for simplicity, they assumed that the lethal factor is located just at the position of the markers under study.

In this report we deal with a method for mapping a lethal factor as well as with estimating true recombination values between markers that show an aberrant segregation by using cosegregation data of two codominant markers in the F_2. We consider three models of gametic or zygotic selection affected by a lethal factor: (1) either male or female gamete selection; (2) both male and female gamete selection; and (3) zygotic selection. The relative viability or fertilization ability of a gamete or zygote carrying a lethal allele as compared with that of a normal one was also estimated. The expectation conditional maximization (ECM) algorithm (Meng and Rubin 1993), which is a special version of EM-algorithm (Dempster et al. 1977), was used to estimate the parameters and their standard errors.

Among the segregation data of molecular markers in F_2 which were used to construct a rice RFLP linkage map by Saito et al. (1991a), data for a group of loci exhibiting distorted segregation on chromosome III were used as a numerical example to illustrate our method. The most likely model for gametic or zygotic selection could be determined by goodness of fit in testing the observed frequencies of the phenotypes in F_2 against the ones expected under the model supposed.

Theory

Consider a mating of *AALLBB* (P_1)×*aallbb* (P_2), where (1) *A-a* and *B-b* are two flanking codominant molecular markers, and (2) *L-l* is a lethal factor locus that is assumed to be located between markers *A* and *B* on the same chromosome, the order of the three loci is then *A-L-B*. Let the recombination value between *A* and *L* be expressed as r_1, and that between *L* and *B* as r_2 . No chiasma interference is assumed. When the gametic or zygotic selection on *L* caused by the lethal factor was present, the differential viability of gametes with the genotype *l* or zygotes *ll* was expressed as t ($0<t<1$) relative to that of the normal gametes, *L*, or zygotes, *LL* and *Ll*. We suppose three models of gametic or zygotic selection on *L* affected by a lethal factor: (1) either male or female gametic selection, where a viability ratio of the three genotypes *LL*, *Ll*, and *ll* is $1:1+t:t$ ($0<t<1$); (2) both male and female gametic selection with equal differential viability, where the ratio is $1:2t:t^2$, assuming equal depression of viability for both sexes; and (3) zygotic selection, where the ratio is $1:2:t$, assuming the selection results from the interaction between the cytoplasm of P_1 and a recessive lethal gene in the nucleus derived from P_2. Such interaction was reported in rice by Sato et al. (1990) and Morishima et al. (1992).

If *L-l* were the normal molecular marker locus with no lethal effect for the recessive, the expected gametic frequencies of eight genotypes, *ALB, ALb, AlB, Alb, aLB, aLb, alB*, and *alb*, derived from the F_1 (*ALB/alb*) individuals would be $(1-r_1)(1-r_2)/2$, $(1-r_1)r_2/2$, $r_1r_2/2$, $r_1(1-r_2)/2$, $r_1(1-r_2)/2$, $r_1r_2/2$, $(1-r_1)r_2/2$ and $(1-r_1)(1-r_2)/2$, respectively. Table 1 shows the expected frequencies f_{ij} of the 27 classes of F_2 genotypes with no differential viability. The *LL*, *Ll*, and *ll* genotypes cannot be observed directly, and only the segregation data of the nine phenotypic classes with respect to molecular markers *A* and *B* can be obtained. Corresponding expected frequencies, f_i (i=1,2,...,9), can be expressed as the sum of products of functions, $f_{ij}(r_1,r_2)$ and $g_j(t)$,

$$\boldsymbol{f} = \boldsymbol{A}\boldsymbol{g} \tag{1}$$

where $\boldsymbol{f}'=(f_1, f_2,\ldots, f_9)$, $\boldsymbol{g}'=(g_1, g_2, g_3)$, and $\boldsymbol{A}=(f_{ij})$ is a 9×3 matrix (A prime shows a transposed matrix), and where (g_1, g_2, g_3) can be expressed as follows

$$\left(\frac{1}{2(1+t)}\ \frac{1}{4}\ \frac{t}{2(1+t)}\right) \quad \text{for model 1} \tag{2}$$

$$\left(\frac{1}{(1+t)^2}\ \frac{t}{(1+t)^2}\ \frac{t^2}{(1+t)^2}\right) \quad \text{for model 2} \tag{3}$$

$$\text{and}\ \left(\frac{1}{3+t}\ \frac{1}{3+t}\ \frac{t}{3+t}\right) \quad \text{for model 3.} \tag{4}$$

Theoretically, the observed frequency a_i (i=1,2,...,9) of a phenotype with respect to *A* and *B* is the sum of three quan-

Table 1 Expected genotype frequencies $f_{i,j}$ in an F_2 population[a,b]

Genotype	$LL(f_{i1})$[c]	$Ll(f_{i2})$	$ll(f_{i3})$
AABB	$(1-r_1)^2(1-r_2)^2$	$2r_1(1-r_1)\,r_2(1-r_2)$	$r_1^2\,r_2^2$
AABb	$2(1-r_1)^2\,r_2(1-r_2)$	$2r_1(1-r_1)\,[r_2^2+(1-r_2)^2]$	$2r_1^2\,r_2(1-r_2)$
AAbb	$(1-r_1)^2\,r_2^2$	$2r_1(1-r_1)\,r_2(1-r_2)$	$r_1^2(1-r_2)^2$
AaBB	$2r_1(1-r_1)(1-r_2)^2$	$2[r_1^2+(1-r_1)^2]\,r_2(1-r_2)$	$2r_1(1-r_1)\,r_2^2$
AaBb	$4r_1(1-r_1)\,r_2(1-r_2)$	$2[r_1^2+(1-r_1)^2]\,[r_2^2+(1-r_2)^2]$	$4r_1(1-r_1)\,r_2(1-r_2)$
Aabb	$2r_1(1-r_1)\,r_2^2$	$2[r_1^2+(1-r_1)^2]\,r_2(1-r_2)$	$2r_1(1-r_1)\,(1-r_2)^2$
aaBB	$r_1^2(1-r_2)^2$	$2r_1(1-r_1)\,r_2(1-r_2)$	$(1-r_1)^2\,r_2^2$
aaBb	$2r_1^2\,r_2(1-r_2)$	$2r_1(1-r_1)\,[r_2^2+(1-r_2)^2]$	$2(1-r_1)^2\,r_2(1-r_2)$
aabb	$r_1^2\,r_2^2$	$2r_1(1-r_1)\,r_2(1-r_2)$	$(1-r_1)^2(1-r_2)^2$

[a] The recombination values between *A* and *L*, and between *L* and *B* are r_1 and r_2, respectively
[b] All frequencies are multiplied by 1/4
[c] i=1, 2,..., 9

tities, i.e., $a_i=a_{i1}+a_{i2}+a_{i3}$, (i=1,2,...,9), and the a_{ij} could be expressed as

$$a_{ij}=a_i\times\frac{f_{ij}\times g_j}{f_i}\qquad(i=1,2,\ldots,9;\ j=1,2,3). \tag{5}$$

The likelihood could be obtained as follows:

$$\mathbf{e}^{\mathbf{L}}\infty\prod_{i=1}^{9}\prod_{j=1}^{3}\{f_{ij}(r_1,r_2)\times g_j(t)\}^{a_{ij}}, \tag{6}$$

so that the log-likelihood is

$$\mathbf{L}=\sum_{i=1}^{9}\sum_{j=1}^{3}(a_{ij}\log f_{ij}(r_1,r_2)+a_{ij}\log g_j(t)), \tag{7}$$

then three equations for score are

$$S_{r_1}=\frac{\partial L}{\partial r_1}=\sum_{i=1}^{9}\sum_{j=1}^{3}\left[\frac{a_{ij}}{f_{ij}}\times\frac{\partial f_{ij}}{\partial r_1}\right]=0 \tag{8}$$

$$S_{r_2}=\frac{\partial L}{\partial r_2}=\sum_{i=1}^{9}\sum_{j=1}^{3}\left[\frac{a_{ij}}{f_{ij}}\times\frac{\partial f_{ij}}{\partial r_2}\right]=0 \tag{9}$$

$$S_t=\frac{\partial L}{\partial t}=\sum_{j=1}^{3}\left[\frac{\partial\log g_j}{\partial t}\times\sum_{i=1}^{9}a_{ij}\right]=0. \tag{10}$$

It should be noted that Eqs. 8 and 9 do not include t, indicating that recombination values r_1 and r_2 can be estimated independently of the estimate of t in any of the three models. The maximum likelihood estimates can be obtained by solving these equations. S_t=0 could be algebraically solved

for model 1	for model 2	for model 3
$t=\dfrac{\sum_{i=1}^{9}a_{i3}}{\sum_{i=1}^{9}a_{i1}}$	$t=\dfrac{\sum_{i=1}^{9}a_{i2}+2\sum_{i=1}^{9}a_{i3}}{\sum_{i=1}^{9}a_{i2}+2\sum_{i=1}^{9}a_{i1}}$	$t=\dfrac{3\sum_{i=1}^{9}a_{i3}}{\sum_{i=1}^{9}a_{i1}+\sum_{i=1}^{9}a_{i2}}$

(11)

As S_{r_1}=0 and S_{r_2}=0 cannot be solved by conventional algebraic methods, iterative calculation is needed to obtain the estimates of r_1 and r_2. In each iteration cycle the estimates were obtained by the bisection method. For finding the maximum likelihood estimates of r_1, r_2 and t simultaneously, we first give the r_1, r_2 and t arbitrary initial values within the intervals (0, 0.5), (0, 0.5), and (0, 1). In the (k+1)st expectation step (E-step) of the ECM-algorithm, $a_{ij}^{(k+1)}$ can be obtained by Eq. 5, with observed frequency a_i and the current estimates of the parameters $r_1^{(k)}$, $r_2^{(k)}$ and $t^{(k)}$. The (k+1)st conditional maximization step (CM-step) then finds the maximum likelihood estimates of $r_1^{(k+1)}$, $r_2^{(k+1)}$ and $t^{(k+1)}$. The practical procedures of the CM step are

1) By formula (11), estimating $t^{(k+1)}$ with $a_{ij}^{(k+1)}$;
   By solving Eq. 8, $S_{r_1}(r_1 : r_2^{(k)}, a_{ij}^{(k+1)})=0$, to obtain the $r_1^{(k+1)}$;
2) By solving Eq. 9, $S_{r_2}(r_2{:}r_1^{(k+1)}, a_{ij}^{(k+1)})=0$, to obtain the $r_2^{(k+1)}$.

Thus, one cycle of iteration of ECM consists of one E-step and two CM-steps. The three estimates would converge to their respective limits simultaneously (Wu 1983). When the differences between the value of an estimate in the previous CM-step and current one become very small and less than the pre-determined quantity, then the iteration is stopped and the final estimates ($\hat{r}_1$,$\hat{r}_2$ and $\hat{t}$) are obtained.

Let $\theta_1=r_1$, $\theta_2=r_2$, and $\theta_3=t$, and n be the total number of F_2 individuals, then the Fisher's information matrix $\mathbf{I}$ is given by

$$\mathbf{I}=\begin{bmatrix}\mathrm{I}_{11} & \mathrm{I}_{12} & 0\\ \mathrm{I}_{21} & \mathrm{I}_{22} & 0\\ 0 & 0 & \mathrm{I}_{33}\end{bmatrix}.$$

The elements of the matrix will be obtained as follows (Bailey 1961; Mather 1963)

$$\mathrm{I}_{\mathrm{lm}}=-E\frac{\partial^2 L}{\partial\theta_l\,\partial\theta_m}\qquad(\mathrm{l,m}=1,2,3). \tag{12}$$

$\mathbf{I}$ is a symmetrical matrix ($\mathrm{I}_{\mathrm{lm}}=\mathrm{I}_{\mathrm{ml}}$), and the diagonal elements of $\mathbf{I}$ are given by

$$\mathrm{I}_{11}=\mathrm{I}_{r_1}=-E\frac{\partial^2 L}{\partial r_1^2}=n\sum_{i=1}^{9}\sum_{j=1}^{3}\frac{g_j}{f_{ij}}\left(\frac{\partial f_{ij}}{\partial r_1}\right)^2 \tag{13}$$

$$\mathrm{I}_{22}=\mathrm{I}_{r_2}=-E\frac{\partial^2 L}{\partial r_2^2}=n\sum_{i=1}^{9}\sum_{j=1}^{3}\frac{g_j}{f_{ij}}\left(\frac{\partial f_{ij}}{\partial r_2}\right)^2 \tag{14}$$

$$\mathrm{I}_{33}=\mathrm{I}_t=-E\frac{\partial^2 L}{\partial t^2}=\frac{n}{2t(1+t)^2}\qquad\text{for model 1} \tag{15.1}$$

$$\mathrm{I}_{33}=\mathrm{I}_t=-E\frac{\partial^2 L}{\partial t^2}=\frac{2n}{t(1+t)^2}\qquad\text{for model 2} \tag{15.2}$$

$$\mathrm{I}_{33}=\mathrm{I}_t=-E\frac{\partial^2 L}{\partial t^2}=\frac{3n}{t(3+t)^2}\qquad\text{for model 3.} \tag{15.3}$$

Only I_{12} and I_{21} are non-zeros in the off-diagonal elements and given by

$$\mathrm{I}_{12}=\mathrm{I}_{21}=-E\frac{\partial^2 L}{\partial r_1\,\partial r_2}=n\sum_{i=1}^{9}\sum_{j=1}^{3}\frac{g_j}{f_{ij}}\left(\frac{\partial f_{ij}}{\partial r_1}\right)\left(\frac{\partial f_{ij}}{\partial r_2}\right). \tag{16}$$

The covariance matrix for the three estimates is given by the inverse of the information matrix as follows

$$\mathbf{I}^{-1}=\begin{bmatrix}\mathrm{I}^{11} & \mathrm{I}^{12} & 0\\ \mathrm{I}^{21} & \mathrm{I}^{22} & 0\\ 0 & 0 & \mathrm{I}^{33}\end{bmatrix},$$

where the non-zero elements are

$$\mathrm{I}^{11}=\frac{\mathrm{I}_{22}}{\mathrm{I}_{11}\mathrm{I}_{22}-\mathrm{I}_{12}^2} \tag{17}$$

$$\mathrm{I}^{22}=\frac{\mathrm{I}_{11}}{\mathrm{I}_{11}\mathrm{I}_{22}-\mathrm{I}_{12}^2} \tag{18}$$

$$I^{33} = \frac{1}{I_{33}} \tag{19}$$

$$I^{12} = I^{21}\frac{-I_{12}}{I_{11}I_{22} - I_{12}^2} \quad \text{or} \quad \frac{-I_{21}}{I_{11}I_{22} - I_{12}^2}. \tag{20}$$

Since the maximum likelihood estimates $\hat{\Theta}=(\hat{\theta}_1, \hat{\theta}_2, \hat{\theta}_3)'$ follow multivariate normal distribution in a large sample with mean Θ and covariance matrix I^{-1}, the variances of the three estimates of parameters, $\hat{r}_1$, $\hat{r}_2$ and $\hat{t}$ would be I^{11}, I^{22}, and I^{33}, respectively, and the corresponding standard error is given by I^{ii}(i=1,2,3).

As described above, in each of the three models the recombination values (r_1, r_2) between a lethal factor locus and a pair of molecular markers (*A* and *B*), and the differential viability, *t*, as well as their standard errors can be estimated. But it has to be determined which of the three models is the best fitted for the data. We used the formula $E_i=n\times f_i(\hat{r}_1, \hat{r}_2, \hat{t})$ to calculate the expected number of individuals, E_i, in each phenotype with markers *A* and *B*, then calculate

$$\chi^2 = \sum_{i=1}^{9}\left[\frac{(E_i - a_i)^2}{E_i}\right] \qquad (df = 5)$$

for testing the goodness of fit. The model showing the smallest χ^2 value was chosen as the best fitted one. The map distances between markers *A*, *B* and the lethal factor locus, *L*, can then be calculated by a mapping function (Haldane 1919) from the estimates of recombination values obtained under the best fitted model. Finally, the lethal factor locus can be located on the linkage map.

Application

Molecular marker data

In mapping rice chromosomes, Saito et al. (1991b) reported that there was a group of molecular markers showing distorted segregation on chromosome III in a F_2 derived from a cross of 'Kasalath' (*Indica*) × 'Fl134' (*Japonica*). A distorted segregation of markers in the same region of chromosome III was also reported by McCouch et al. (1988) and Causse et al. (1994) in backcross populations. In this paper we used the data from Saito et al. (1991b) to analyze the linkage between the molecular markers and a possible lethal factor locus (*L*) in the neighboring region of chromosome III.

Table 2 shows the distorted segregations of 21 molecular markers which are located in a region of chromosome III and the results of χ^2 testing for the fitness of the observed segregation ratio to the expected ratio 1:2:1 for codominance. χ^2 values were larger than 9.21 ($\chi^2_{2,0.01}$) in all of the cases shown in the table, the threshold value was at the 1% significant level with 2 degrees of freedom. The possibility of gametic or zygotic selection affected by a lethal factor was highly suggested in this region. When the χ^2 values were plotted against the location of the marker, a clear relationship between the locations and values was observed (Fig. 1), suggesting that the lethal factor locus was located close to RFLP marker no. 23 at the peak.

Table 2 Distorted segregation of a group of RFLP markers on rice chromosome III and the results of χ^2 testing for the segregation ratio (Data from Saito et al. 1991a)

Marker	Segregation (total)	χ^2(1:2:1)
144	62 : 63 : 19 (144)	27.93
51	67 : 59 : 18 (144)	38.04
355	54 : 47 : 9 (110)	39.15
23	72 : 58 : 14 (144)	52.17
15	67 : 63 : 14 (144)	41.26
394	59 : 48 : 9 (117)	45.82
62	68 : 56 : 12 (136)	50.35
326	60 : 49 : 9 (118)	47.47
74	68 : 63 : 13 (144)	44.26
285	68 : 62 : 13 (143)	44.83
238	62 : 67 : 15 (144)	31.38
234	61 : 66 : 13 (140)	33.37
182	61 : 69 : 14 (144)	30.93
391	61 : 68 : 14 (143)	31.24
129	61 : 65 : 18 (144)	27.04
392	61 : 64 : 18 (143)	27.43
256	62 : 63 : 18 (143)	29.10
373	60 : 67 : 17 (144)	26.38
55	55 : 71 : 18 (144)	19.04
348	53 : 71 : 20 (144)	15.15
184	52 : 70 : 22 (144)	12.61

Linkage analysis

Let the genotypes of 'Kasalath' (P_1) and 'Fl134' (P_2) be *AALLBB* and *aallbb*, respectively, where *A-a* is flanking marker locus no. 23, *B-b* is one of other 17 molecular marker loci, and *L-l* is a lethal factor locus. Tentatively, let *l* be a lethal allele and *L* a normal one. Using the procedures described in the Theory section, we estimated the recombination values between the lethal factor locus (*L*) and molecular markers *A* (no. 23) and each *B*, (r_1, r_2) and the differential viabilities of gametes or zygotes (*t*) in each of the three supposed models. The estimates and their standard errors for the models 1, 2, and 3 are shown in Tables 3, 4 and 5, respectively.

It is clear that all of the estimated recombination values (r_1) between RFLP marker no. 23 (*A*) and the lethal factor locus (*L*) in model 1 and model 2 were close to zero (Tables 3 and 4). The estimates of r_1 and the standard errors of *t* were larger in model 3 than those in the other two models (Table 5). In order to determine the best fitted model, the results of χ^2 testing for the goodness of fit are shown in Table 6. All of the χ^2 values in model 3 (Table 6) were larger than $\chi^2_{5,0.01}$(>15.09), indicating that the zygotic selection model (model 3) was the least fitted. While all of the χ^2 values in both models 1 and 2 were smaller than $\chi^2_{5,0.05}$ (<11.07), χ^2 values were always smaller in model 2 than in model 1. In particular, when RFLP marker no. 326 was used as the locus *B* in model 2, the χ^2 was

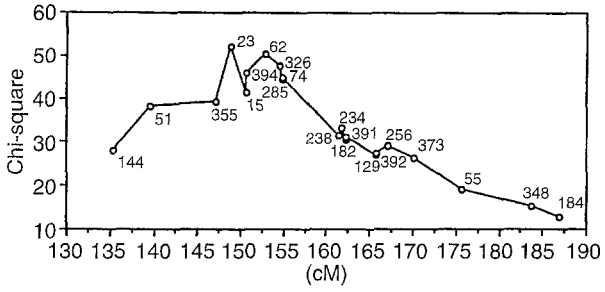

Fig. 1 The region of the distorted segregation group of 21 RFLP molecular markers on rice chromosome III (Saito et al. 1991a) and chi-square values

Table 3 The maximum likelihood estimates of recombination values (r_1, r_2) and differential viabilities (t) by model 1

A-L-B	$r_1 \pm SE^a$	$r_2 \pm SE$	$t \pm SE$
23-*L*-15	0.000 ± 0.000	0.017 ± 0.008	0.194 ± 0.062
394	0.009 ± 0.006	0.044 ± 0.014	0.136 ± 0.055
62	0.021 ± 0.009	0.021 ± 0.009	0.166 ± 0.058
326	0.011 ± 0.007	0.037 ± 0.013	0.134 ± 0.054
74	0.010 ± 0.006	0.029 ± 0.010	0.187 ± 0.061
285	0.010 ± 0.006	0.030 ± 0.010	0.187 ± 0.061
238	0.000 ± 0.001	0.075 ± 0.016	0.194 ± 0.062
234	0.001 ± 0.002	0.073 ± 0.016	0.182 ± 0.060
182	0.000 ± 0.001	0.075 ± 0.016	0.194 ± 0.062
391	0.000 ± 0.001	0.076 ± 0.016	0.194 ± 0.062
129	0.000 ± 0.001	0.098 ± 0.019	0.194 ± 0.062
392	0.000 ± 0.001	0.098 ± 0.019	0.194 ± 0.062
256	0.000 ± 0.001	0.113 ± 0.020	0.197 ± 0.063
373	0.009 ± 0.006	0.114 ± 0.020	0.184 ± 0.060
55	0.000 ± 0.001	0.136 ± 0.022	0.194 ± 0.062
348	0.000 ± 0.001	0.176 ± 0.025	0.194 ± 0.062
184	0.000 ± 0.001	0.205 ± 0.027	0.194 ± 0.062

[a] SE is the standard error of estimate

Table 4 The maximum likelihood estimates of recombination values (r_1, r_2) and differential viabilities (t) by model 2

A-L-B	$r_1 \pm SE^a$	$r_2 \pm SE$	$t \pm SE$
23-*L*-15	0.000 ± 0.000	0.017 ± 0.008	0.426 ± 0.055
394	0.009 ± 0.006	0.045 ± 0.014	0.361 ± 0.053
62	0.017 ± 0.009	0.025 ± 0.010	0.395 ± 0.053
326	0.011 ± 0.007	0.038 ± 0.013	0.363 ± 0.053
74	0.007 ± 0.005	0.032 ± 0.011	0.420 ± 0.054
285	0.007 ± 0.005	0.032 ± 0.011	0.417 ± 0.054
238	0.000 ± 0.001	0.075 ± 0.016	0.426 ± 0.055
234	0.000 ± 0.001	0.074 ± 0.016	0.414 ± 0.054
182	0.000 ± 0.001	0.075 ± 0.016	0.426 ± 0.055
391	0.000 ± 0.001	0.076 ± 0.016	0.423 ± 0.055
129	0.000 ± 0.001	0.098 ± 0.018	0.426 ± 0.055
392	0.000 ± 0.001	0.098 ± 0.018	0.423 ± 0.055
256	0.001 ± 0.002	0.113 ± 0.020	0.429 ± 0.055
373	0.000 ± 0.001	0.120 ± 0.020	0.425 ± 0.055
55	0.000 ± 0.001	0.137 ± 0.022	0.426 ± 0.055
348	0.000 ± 0.001	0.176 ± 0.025	0.426 ± 0.055
184	0.000 ± 0.001	0.205 ± 0.027	0.426 ± 0.055

[a] SE is the standard error of estimate

Table 5 The maximum likelihood estimates of recombination values (r_1, r_2) and differential viabilities (t) by model 3

A-L-B	$r_1 \pm SE^a$	$r_2 \pm SE$	$t \pm SE$
23-*L*-15	0.005 ± 0.004	0.014 ± 0.007	0.323 ± 0.091
394	0.027 ± 0.011	0.027 ± 0.011	0.230 ± 0.083
62	0.033 ± 0.011	0.009 ± 0.006	0.281 ± 0.086
326	0.024 ± 0.010	0.024 ± 0.010	0.228 ± 0.082
74	0.039 ± 0.012	0.000 ± 0.001	0.298 ± 0.087
285	0.039 ± 0.012	0.000 ± 0.001	0.300 ± 0.087
238	0.024 ± 0.009	0.053 ± 0.014	0.300 ± 0.087
234	0.038 ± 0.012	0.039 ± 0.012	0.276 ± 0.084
182	0.039 ± 0.012	0.039 ± 0.012	0.293 ± 0.086
391	0.039 ± 0.012	0.039 ± 0.012	0.296 ± 0.087
129	0.007 ± 0.005	0.092 ± 0.018	0.316 ± 0.090
392	0.007 ± 0.005	0.092 ± 0.018	0.318 ± 0.090
256	0.007 ± 0.005	0.107 ± 0.020	0.318 ± 0.090
373	0.038 ± 0.012	0.089 ± 0.018	0.269 ± 0.081
55	0.038 ± 0.012	0.106 ± 0.022	0.276 ± 0.083
348	0.036 ± 0.011	0.150 ± 0.024	0.275 ± 0.083
184	0.044 ± 0.012	0.176 ± 0.026	0.265 ± 0.081

[a] SE is the standard error of estimate

Table 6 The results of the χ^2 test for model fitting

Markers *A-L-B*	χ^2-value		
	Model 1	Model 2	Model 3
23-*L*-15	10.92	5.48	34.81**
394	7.30	1.09	30.76**
62	6.97	1.05	30.14**
326	6.54	0.75	29.54**
74	8.99	3.40	31.18**
285	8.60	3.36	30.34**
238	7.72	2.32	30.62**
234	8.68	3.11	31.62**
182	9.36	3.96	32.15**
391	9.68	3.92	32.92**
129	6.36	1.09	29.32**
392	6.75	1.13	30.15**
256	6.06	1.13	28.09**
373	9.08	4.53	29.02**
55	8.07	2.91	29.83**
348	8.52	3.16	30.65**
184	6.66	1.49	28.28**

** Significant at the 1% level

found to be the smallest and only 0.75. Therefore, model 2 was considered to be the best fitted model. We calculated the weighted averages of estimated r_1 and t in model 2 over the 17 cases shown in the table and obtained $\bar{r}_1$=0.008, and $\bar{t}$=0.415. By the Haldane's mapping function (Haldane 1919) the map distance between RFLP marker no. 23 and the lethal factor locus (*L*) was calculated to be 0.8 cM. The weighted average of estimated differential viabilities of the male and female gametes of genotype *l* was 41.5% as compared with the normal gamete. In Fig. 2, we located the lethal factor locus (*L*) on chromosome III of the rice RFLP linkage map constructed by Saito et al. (1991a). The fact that the estimated value of t was less than 1 shows that

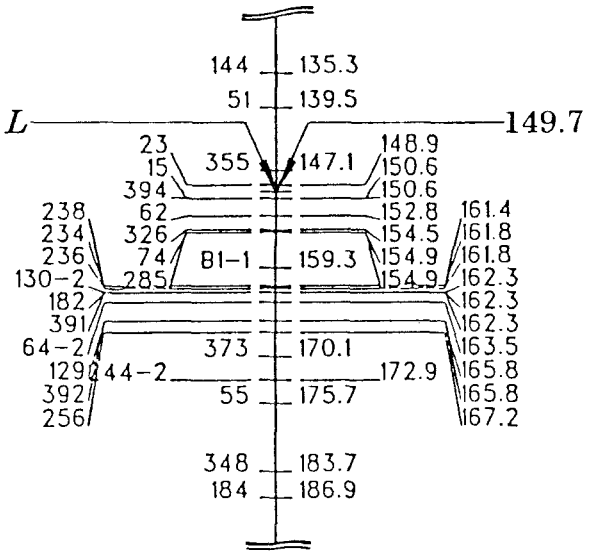

Fig. 2 The location of the lethal factor locus (*L*) on rice chromosome III. The *numbers* and *letters* on the *left* are the marker names, and the *numbers* on the *right* indicate the locations of the markers (in cM)

the donor of the lethal factor was the *Japonica* parent 'Fl134'. If it were otherwise, the estimated t would be larger than 1.

Discussion

Aberrant segregation has been repeatedly observed for different plant species in plant breeding studies in which distant crosses, i.e., crosses between different species, and subspecies and between wild and cultivated species, were made. When molecular linkage maps are constructed, crosses are usually made between varieties that are distantly related in order to get higher degrees of DNA polymorphism, and the consequence of this is that aberrant segregations are often encountered (Wendel and Parks 1984; Torres et al. 1985; Paterson et al. 1988; Pham et al. 1990; Lyttle 1991; Schon et al. 1991; Paterson et al. 1991; Zivy et al. 1992). McCouch et al. (1988) and Saito et al. (1991b) found conspicuously distorted segregation at a group of marker loci in a localized region on chromosome III of rice. Causse et al. (1994) also found aberrant segregations of markers in the same region of chromosome III. They suggested the presence of a lethal factor linked with those markers and gave a position of the factor based on the scatter diagram of allele frequency against map distance. The lethal factor, which was found to be located at a position near to RFLP marker no. 23 and 149.7 cM from a trait marker *chl-1* on chromosome III of the current map, is thought to be identical to the one found by Causse et al. (1994), although the latter did not give the exact position of the lethal factor and the mechanism underlying the phenomenon. The closer the linkage between a marker and a lethal factor, the more conspicuous is the expected distortion of segregation. Therefore, with respect to allele frequency as adopted by Causse et al. (1994), the magnitude of the chi-square values in testing the observed ratio against the expected one or upper probability of the chi-square distribution can be used for guessing the position of the lethal factor. However, it is only the interval between flanking markers of the lethal factor and not the position that can be determined by such methods. If the interval is not sufficiently close we cannot locate the factor on a linkage map. In the present study we showed that by using the cosegregation data of two codominant markers with distorted segregation we can estimate the location, viability, or relative fertilization ability and the model for the action of the lethal factor. The action of the lethal factor was found to be effective on both male and female gametes with a fertilization ability 41.5% of that of the normal gamete.

Distorted segregation has been reported in rice as early as 1928 by Chao, followed by Ramiah et al. (1931), Oka (1953a, b), and Mizushima and Kondo (1960). It is now believed that distorted segregation at a specific marker locus in rice is mainly due to the linkage of this marker to a gametophyte gene (denoted as *ga*) located nearby on the same chromosome (Iwata et al. 1964; Nakagahra 1972; Mori et al. 1973), although other factors such as complementary genes, duplicate genes, chromosomal abnormality, and competitive fertilization between marker genotypes have been suggested as the cause of distortion in some reports.

An attempt at locating of a gametic lethal factor in rice was first made by Iwata et al. (1964) using F_2, F_3 and F_4 populations derived from a cross between a mutant and a tester line. They found three types (low, normal, and high) with respect to the segregation ratio of the recessive of a marker (*wx* or *C*) among F_3 lines derived from F_2 heterozygous plants and were able to estimate the recombination value between the lethal factor and the marker from the relative proportion of these types. Since no clear sterility of pollen or seed was detected, the effect of the *ga* gene, they supposed, may be in lowering the ability of the *ga* pollens to take part in fertilization. They were also able to estimate the relative ability of the *ga* pollen, which was only 0.05 of that of the normal pollen. Nakagahra et al. (1972) found another gametophyte gene (*ga2*) and were able to locate it to the interval between markers *bc* (brittle culm) and *dl* (drooping leaf) on the chromosome III. So far, 12 gametophyte genes have been identified and located on the linkage map of rice (Iwata et al. 1964; Nakagahra 1972; Nakagahra et al. 1972; Mori et al. 1973; Nakagahra et al. 1974; Maekawa et al. 1981; Maekawa 1982; Kinoshita and Takamura 1984; Maekawa and Kita 1985; Lin and Ikehashi 1993). In all of these studies recombination values between the marker and gametophyte gene were estimated by adopting the method of Iwata (1964). Compared with this, our method has some advantages. Firstly, segregation data in an F_2 generation is sufficient for the analysis, and F_3 or later generations are not necessary. Secondly, the model of the action of the lethal factor can be determined

from the data analysis. We can judge which of the models is the best by chi-square values after fitting the observed frequencies to the ones expected under the supposed model. Thirdly, in Iwata's method discrimination of F_3 lines with a higher, lower, and normal segregation ratio of the recessive genotype would be difficult unless the linkage between the marker and the lethal factor is close and/or the gamete or zygote with the lethal allele has a sufficiently lower fertilization ability as compared with the normal one.

All of the gametophyte genes (*ga1* to *ga12*) so far reported in rice are believed to act only in the male gamete. On the other hand, three genes (*S-5, S-7* and *S-8*) have been reported to be related to hybrid sterility, which results from female gamete abortion (Yanagihara et al. 1992a, b, 1995; Wan et al. 1993). Ikehashi and Araki (1984, 1986, 1988) investigated the fertility of F_1 plants from many crosses between subspecies *Japonica*, *Indica*, and *Javanica* and found that different selection models are likely for different crosses, i.e., the selection of either the male or female gamete for some crosses and the selection of both gametes for others.

Of the gametophyte genes so far reported *ga2* and *ga3* are known to be located on chromosome III. Nakagahra et al. (1974) obtained varying estimates of recombination values between *ga2* and a marker, *bl*, for different crosses, the average being 11.2% which corresponds to a map distance of 12.7 cM by the Haldane map function. If *ga2* is located downward of *bl* on the linkage map, it would be in the region where the lethal factor described in the present report was found, although the action of *ga* is only on the male gamete while that of the latter is on both gametes. In the case of selection for both gametes we assumed for simplicity that the fertilization ability of the gametes with the lethal allele is equal for the male and female sides. The model of selection for both gametes with unequal fertilization abilities was also investigated, but nearly the same result as in equal ability was obtained. Saito et al. (1991b) reported that the fertility of seed set in the F_1s derived from the cross 'Kasalath' × 'Fl134' was 85%. In spite of the high seed fertility they observed, our estimate of t for both male and female gametes with genotype l was as low as 0.415, which is equivalent to a gamete fertility of $0.7075(=(1+t)/2)$. Low pollen fertility associated with high seed fertility is not an uncommon phenomenon, but it is difficult to explain how high seed fertility can be expected under the condition of low female gamete fertility. One explanation for this may be that partial elimination of egg mother cells with the lethal allele occurs in the course of oogenesis and that the fertility of the resulting egg cells itself is not so low. Further study on this point is needed.

We also observed two other regions of distorted segregation – at a group of loci in a localized region of chromosomes IV and IX. Two lethal factors corresponding to these regions were found, the actions of which were in both cases on both the male and female gametes. The estimated value of t was 0.73 and 0.54 for chromosome IV and IX, respectively. The donor of the lethal factor was from 'Kasalath' for the former and from Fl134 for the latter. The position of the lethal factor on chromosome IV was 1.3 cM upstream of RFLP marker no. 282, which is 46.8 cM upstream of the *lg* gene. The region was different from the locations of *ga6* or *ga10* which are known to be on the same chromosome. On chromosome IX a lethal factor was found to be located 0.8 cM upstream of the marker no. 404. No gametophytic or sterility gene has been reported on this chromosome.

In this paper, we only presented the method in the case of codominance for both markers. It can easily be extended to the cases of codominance versus dominance and dominance versus dominance for markers A versus B. The vector $(g_1, g_2, g_3)'$ is identical in all of the three models, and expected frequencies, $f_{ij}(r_1,r_2)$ (i=1,2,...,6 for the case of codominance versus dominance, and i=1,2,...,4 for dominance versus dominance), can be easily obtained. In the present method two flanking markers with distorted segregation are required and at least one of them must be codominant. Since the three parameters, i.e., r_1 (between the lethal factor and neighboring marker A), r_2 (between the lethal factor and another marker B), and t, relative fertilization ability of gamete or zygote are to be estimated, 4 or more degrees of freedom (*df*) in the segregation data are required for determination of the best model with respect to the action of the lethal gene. If both markers are dominant, the number of classes in the segregation data in the F_2 generation is only 4 and hence the *df* is *3*, leaving no extra *df* after fitting the observed frequencies to the expected ones. Although r_1, r_2, and t can be calculated even in this case, the observed and expected frequencies are identical for all classes of segregation whatever model may be adopted, and so we cannot determine which of the three models is the best. In this respect RFLP markers which are mostly codominant are suitable for the present method.

Recently, Lorieux et al. (1995a,b) reported how estimation of the recombination value between markers is affected by differential viability at the gamete or zygote level in an F_2 and a backcross population. They considered two cases of selection, male gamete and zygote selection, but not both male and female gamete selection. They showed how to estimate the recombination value and relative viability of the gametes or zygotes by maximum likelihood. The effect of distortion on the estimation of recombination value for three cases where the two markers are dominant-dominant, codominant-codominant, and dominant-codominant were compared and it was concluded that estimation of the recombination value is less affected by selection in codominant markers than in dominant markers. They assumed in their analysis for simplicity that the markers showing distortion are exactly located on genes affected by gametic or zygotic selection. Contrary to their method, we assumed that the distortion of segregation of markers is due to linkage of the marker with a lethal gene, the effect of which is partial or entire selection of the gamete or zygote with the gene. Our results proved theoretically that the estimation of recombination value is not affected by distortion at all in the case of codominance versus codominance as shown in the Eps. 8 and 9, in which estimation of recombination values (r_1, r_2) can be made independently of the estimation of fertilization ability (t) in any of the three models adopted. In our method, in which

aberrant segregation of a marker is due to the linkage of the marker with a lethal factor, the same is true even for codominant versus dominant and dominant versus dominant markers irrespectively of the models for the action of the lethal factor.

Acknowledgments We gratefully acknowledge all of the members of the group which constructed the rice RFLP linkage map (Saito et al. 1991a) for providing us the valuable data. This study was partly supported by the Japan Society for the Promotion of Science (JSPS) in the form of a postdoctoral fellowship for foreign researchers.

Appendix

Information values for estimators r_1 and r_2

$$I_{lm} = -E\left(\frac{\partial^2 L}{\partial r_l\, \partial r_m}\right) = -E\left[\frac{\partial}{\partial r_m}\left(\frac{\partial L}{\partial r_l}\right)\right]$$

$$= -E\left\{\frac{\partial}{\partial r_m}\left[\sum_{j=1}^{3}\sum_{i=1}^{9}\left(\frac{a_{ij}}{f_{ij}(r_l, r_m)} \times \frac{\partial f_{ij}(r_l, r_m)}{\partial r_l}\right)\right]\right\}$$

$$= E\sum_{j=1}^{3}\sum_{i=1}^{9}\left\{\frac{a_{ij}}{f_{ij}^2} \times \left(\frac{\partial f_{ij}}{\partial r_l}\right)\left(\frac{\partial f_{ij}}{\partial r_m}\right) - \frac{a_{ij}}{f_{ij}} \times \left(\frac{\partial^2 f_{ij}}{\partial r_l\, \partial r_m}\right)\right\}$$

in which we substitute the expected values for the observed ones, i.e., nf_i for a_i; then by formula 5, we obtain

$$a_{ij} = a_i \times \frac{f_{ij} \times g_j}{f_i} = nf_{ij}\, g_j .$$

This gives

$$-E\left(\frac{\partial^2 L}{\partial r_l\, \partial r_m}\right) = n\sum_{j=1}^{3}\sum_{i=1}^{9}\frac{g_j}{f_{ij}}\left(\frac{\partial f_{ij}}{\partial r_l}\right)\left(\frac{\partial f_{ij}}{\partial r_m}\right) - n\sum_{j=1}^{3}\left(g_j\left(\sum_{i=1}^{9}\frac{\partial^2 f_{ij}}{\partial r_l\, \partial r_m}\right)\right),$$

where

$$\sum_{i=1}^{9}\frac{\partial^2 f_{ij}}{\partial r_l\, \partial r_m} = \frac{\partial^2}{\partial r_l\, \partial r_m}\left(\sum_{i=1}^{9} f_{ij}\right) = 0 \qquad \text{for } j = 1,2,3.$$

Finally, we obtain

$$I_{lm} = -E\left(\frac{\partial^2 L}{\partial r_l\, \partial r_m}\right) = n\sum_{j=1}^{3}\sum_{i=1}^{9}\frac{g_j}{f_{ij}}\left(\frac{\partial f_{ij}}{\partial r_l}\right)\left(\frac{\partial f_{ij}}{\partial r_m}\right)$$

$$= n\sum_{i=1}^{9}\sum_{j=1}^{3}\frac{g_j}{f_{ij}}\left(\frac{\partial f_{ij}}{\partial r_l}\right)\left(\frac{\partial f_{ij}}{\partial r_m}\right).$$

In particular, when l = m = 1,

$$I_{11} = n\sum_{i=1}^{9}\sum_{j=1}^{3}\frac{g_j}{f_{ij}}\left(\frac{\partial f_{ij}}{\partial r_1}\right)^2,$$

l = m = 2,

$$I_{22} = n\sum_{i=1}^{9}\sum_{j=1}^{3}\frac{g_j}{f_{ij}}\left(\frac{\partial f_{ij}}{\partial r_2}\right)^2,$$

and l = 1, m = 2 or l = 2, m = 1,

$$I_{12} = I_{21} = n\sum_{i=1}^{9}\sum_{j=1}^{3}\frac{g_j}{f_{ij}}\left(\frac{\partial f_{ij}}{\partial r_1}\right)\left(\frac{\partial f_{ij}}{\partial r_2}\right).$$

References

Bailey NTJ (1961) Introduction to the mathematical theory of genetic linkage. Oxford University Press, London

Causse M, Fulton TM, Cho YG, Ahn S, Chunwongse J, Wu KS, Xiao JH, Ronald PC, Harrington SE, Second G, McCouch SR, Tanksley SD (1994) Saturated molecular map of the rice genome based on an interspecific backcross population. Genetics 138:1251–1274

Chao LF (1928) The disturbing effect of the glutinous gene in rice on a Mendelian ratio. Genetics 13: 191–225

Dempster AP, Laird NM, Rubin DB (1977) Maximum likelihood from incomplete data via the EM-algorithm. J R Stat Soc Ser B 39:1–38

Haldane JBS (1919) The combination of linkage values, and the calculation of distance between the loci of linked factors. J Genet 8:299–309

Ikehashi H, Araki H (1984) Varietal screening for compatibility types revealed in F_1 fertility of distant crosses in rice. Jpn J Breed 34:304–312

Ikehashi H, Araki H (1986) Genetics of F_1 sterility in remote crosses of rice. In: Rice genetics. *IRRI*, Manila, Philippines, pp 119–130

Ikehashi H, Araki H (1988) Multiple alleles controlling F_1 sterility in remote crosses of rice. Jpn J Breed 38:283–291

Iwata N, Nagamatsu T, Omura J (1964) Abnormal segregation of waxy and apiculus coloration by a gametophyte gene belonging to the first linkage group in rice. Jpn J Breed 14:33–39

Kinoshita T, Takamura I (1984) Genetical studies on rice plant. Inheritance and linkage relationship on zebra chlorosis and zebra necrosis in rice. LXXXVIII: J Fac Agric Hokkaido Univ 61:445–455

Lin SY, Ikehashi H (1993) A gamete abortion locus detected by segregation distortion of isozyme locus *Est-9* in wide crosses of rice (*Oryza sativa* L.). Euphytica 67:35–40

Lorieux M, Goffinet B, Perrier X, Gonzalez de Leon D, Lanaud C (1995a) Maximum-likelihood models for mapping genetic markers showing segregation distortion. 1. Backcross populations. Theor Appl Genet 90:73–80

Lorieux M, Perrier X, Goffinet B, Lanaud C, Gonzalez de Leon D (1995b) Maximum-likelihood models for mapping genetic markers showing segregation distortion. 2. F_2 populations. Theor Appl Genet 90:81–89

Lyttle TW (1991) Segregation distorters. Annu Rev Genet 25:511–557

Maekawa M (1982) Studies on genetical differentiation in distant crosses of rice. Mem Fac Agric Hokkaido Univ 13:147–177

Maekawa M, Kita F (1985) New gametophyte genes located in the third linkage group (chromosome 3) of rice. Jpn J Breed 35:25–31

Maekawa M, Kinoshita T, Takahashi M (1981) Genetical studies on rice plant . LXXVI: A new gametophyte gene in the second linkage group of rice. J Fac Agric Hokkaido Univ 60:107–114

Mather K (1963) Measurement of linkage in heredity, 2nd edn. Methuen & Co, London

McCouch SR, Kochert G, Yu ZH, Wang ZY, Khush GS, Coffman WR, Tanksley SD (1988) Molecular mapping of rice chromosomes. Theor Appl Genet 76:815–829

Meng XL, Rubin DB (1993) Maximum likelihood estimation via the ECM algorithm: general framework. Biometrika 80:267–278

Mizushima U, Kondo A (1960) Fundamental studies on rice breeding through hybridization between Japanese and foreign varieties. II. Structural difference of chromosomes between a Japanese and an Indian variety proves by anomalous mode of segregation in apiculus anthocyanin pigmentation in their hybrid. Jpn J Breed 10:1–9

Mori K, Kinoshita T, Takahashi M (1973) Genetical studies on rice plant. LVIII: Segregation distortion and its causation of an endosperm character in crosses of distantly related rice varieties. Mem Fac Agric Hokkaido Univ 9:74–86

Morishima H, Sano Y, Oka HI (1992) Evolutionary studies in cultivated rice and its wild relatives. Oxford Surv Biol 8:135–184

Nakagahra M (1972) Genetic mechanism on the distorted segregation of marker genes belonging to the eleventh linkage group in cultivated rice. Jpn J Breed 22:232–238

Nakagahra M, Omura T, Iwata N (1972) Gametophyte genes and their loci on the eleventh linkage group of cultivated rice. Jpn J Breed 22:305–312

Nakagahra M, Omura T, Iwata N (1974) New certation gene on the first linkage group found by inter-subspecific hybridization of cultivated rice. J Fac Agric Kyushu Univ 18:157–167

Oka HI (1953a) The mechanism of sterility in the intervarietal hybrid. Phylogenetic differentiation of the cultivated rice plant. VI. Jpn J Breed 2:217–224

Oka HI (1953b) Gene analysis of intervarietal hybrid sterility and certation due to certain combination of gamete-development-genes in rice. Phylogenetic differentiation of the cultivated rice plant. VIII. Jpn J Breed 3:23–30

Paterson AH, Lander ES, Hewitt JD, Peterson S, Lincoln SE, Tanksley SD (1988) Resolution of quantitative traits into Mendelian factors by using a complete linkage map of restriction fragment length polymorphisms Nature 335:721–726

Paterson AH, DeVerna JW, Lanini B, Tanksley SD (1990) Fine mapping of quantitative trait loci using selected over-lapping recombinant chromosome, in an interspecies cross of tomato. Genetics 124:735–742

Paterson AH, Damon S, Hewitt JD, Zamir D, Rabinowitch HD, Lincoln SE, Lander ES, Tanksley SD (1991) Mendelian factors underlying quantitative traits in tomato: Comparison across species, generations, and environments. Genetics 127:181–197

Pham JL, Glaszmann JC, Sano R, Barbier P, Ghesquiere A, Second G (1990) Isozyme markers in rice: genetic analysis and linkage relationships. Genome 33:348–359

Ramiah K, Jabithara S, Mudaliar SD (1931) The inheritance of characters in rice IV. Mem Dept Agric India Bot Ser 18:159–229

Saito A, Yano M, Kishimoto N, Nakagahra M, Yoshimura A, Saito K, Kuhara S, Ukai Y, Kawase M, Magamine T, Yoshimura S, Ideta O, Ohsawa R, Hayano Y, Iwata N, Sugiura M (1991a) Linkage map of restriction fragment length polymorphism loci in rice. Jpn J Breed 41:665–670

Saito A, Shimosaka E, Hayano Y, Saito K, Matuura S, Yano M (1991b) Molecular mapping on chromosome 3 of rice. Jpn J Breed 41 [Suppl. 1]: 158–159

Sato YI, Sato T, Morishima H (1990) Changing segregation ratios for *gl-1* due to unequal chances of fertilization of female gametes. Rice Genet Newsl 7:97–98

Schon CG, Hayes PM, Blake TK, Knapp SJ (1991) Gametophytic selection in a winter spring barley cross. Genome 34:918–922

Torres AM, Mau-Lastovicka T, Williams TE, Soost RK (1985) Segregation distortion and linkage of Citrus and Poncirus isozyme genes. J Hered 76:289–294

Wan J, Yanagihara S, Kato H, Ikehashi H (1993) Multiple alleles at a new locus causing hybrid sterility between a Korean Indica variety and a Japonica variety in rice (*Oryza sativa* L.). Jpn J Breed 43:507–516

Wendel JF, Parks CR (1984) Distorted segregation and linkage of alcohol dehydrogenase genes in *Camellia japonica* (theaceae). Biochem Genet 22:739–748

Wu CFJ (1983) On the convergence properties of EM algorithm. Ann Stat 11:95–103

Yanagihara S, Ikehashi H, Araki H, Maruyama K (1992a) Difference in recombination frequency between Chromogen (*C*) and Waxy (*wx*) genes in Japonica and Indica rice (*Oryza sativa* L.) by using Japonica lines with a wide compatibility ($S\text{-}5^n$). Jpn J Breed 42:15–21

Yanagihara S, Kato H, Ikehashi H (1992b)A new locus for multiple alleles causing hybrid sterility between an Aus variety and Javanica varieties in rice (*Oryza sativa* L.). Jpn J Breed 42:793–810

Yanagihara S, McCouch SR, Ishikawa K, Ogi Y, Maruyama K, Ikehashi H (1995) Molecular analysis of the inheritance of the *S-5* locus, conferring wide compatibility in Indica/Japonica hybrids of rice (*O. sativa* L.). Theor Appl Genet 90:182–188

Zivy M, Devaux P, Blaisonneaux J, Jean R, Thiellement H (1992) Segregation distortion and linkage studies in microspore-derived double haploid lines of *Hordeum vulgare* L. Theor Appl Genet 83:919–924