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# The pattern of inheritance in apple (*Malus* × *domestica* Borkh.): further results from leaf isozyme analysis

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**Summary.** Eight progenies from controlled crosses and one self-progeny of apple were analysed by electrophoresis for six leaf enzymes. Based on a polyploid origin for this species, three hypotheses were tested: monogenic disomic, bigenic disomic and tetrasomic inheritance. Three enzymes exhibited monogenic inheritance; two exhibited bigenic disomic inheritance specified by two homoeologous genes; and one exhibited bigenic disomic inheritance due to two linked genes. In all cases tetrasomic inheritance was disproved. These results agreed with previous data obtained from pollen isozyme analysis. They indicated a probable allopolyploid origin of the apple genome and the loss of duplicated gene expression in some cases.

**Key words:** Isozymes – Inheritance – Polyploidy – *Malus*

## Introduction

The cultivated apple (*Malus*×*domestica* Borkh.) belongs to the *Maloideae* subfamily of the *Rosaceae*. Its high basic chromosome number (x = 17) is generally attributed to a polyploid origin prior to the cretaceous period. Autopolyploidy as well as allopolyploidy have been postulated according to different methodologies. Cytological observations supported the autopolyploid hypothesis of Darlington and Moffett (1930), with a basic number of x = 7. Their arguments were criticized by Sax (1931), who reported pure diploid meiotic behavior and proposed an allopolyploid hypothesis. On the basis of morphological and anatomical comparisons, Stebbins (1950) postulated hybridization between primitive members of the subfamilies *Spiraeoideae* (x = 9) and *Prunoideae* (x = 8). The analysis of a wide range of phenolic compounds by Challice (1981) led to the same conclusion.

Isozyme electrophoresis provides a new approach to this question. Electrophoretic variants are usually controlled by codominant alleles at one or several loci. Isozyme analysis can be used to distinguish between two patterns of inheritance (Breese and Thomas 1976): (1) disomic inheritance, which connotes to homoeologous genomes with preferential pairing between homologous chromosomes in meiosis, and (2) polysomic inheritance, which connotes to homologous genomes with random chromosome pairing in meiosis.

Very few isozyme inheritance studies have been reported in apple: Misic et al. 1980, working with peroxidases from bark, demonstrated disomic inheritance involving null alleles and a closed linkage between two loci. The present work continues a previous study of the inheritance of seven pollen enzymes in apple (Chevreau et al. 1985); the results provided evidence of bigenic disomic inheritance for five enzymes; the two others had a simple monogenic control.

In this study, isozymes present in leaves were selected so that large progenies of seedlings from controlled crosses could be analysed, and the patterns of inheritance of several enzymes present in both pollen and leaves could be compared. Nine progenies were analysed for six enzymes and the observed segregations tested with the different hypotheses.

## Material and methods

Leaves were collected from young seedlings growing in a greenhouse. Extracts were prepared by homogeneization of 100 mg lyophilised young leaves in 1 ml Tris HCl 0.01 M pH = 8.0 and 100 mg PVP, to prevent phenol oxidation, followed by centrifugation at 35,000 g. Electrophoresis was performed in horizontal starch gel for phosphoglucoisomerase

(PGI), and in vertical polyacrylamide gel for esterase (EST), acid phosphatase (AP), endopeptidase (ENP), peroxidase (PER), and superoxide dismutase (SOD). A discontinuous system (Histidine NaCl/Citrate NaOH at pH=7.0) was used for starch gels (13% starch), and electrophoresis was conducted at 130 V. Polyacrylamide gels were composed of a Tris borate EDTA running gel at pH=8.3 (9% acrylamide) and a Tris HCl stacking gel at pH=6.8 (4% acrylamide); electrophoresis was performed at 150 V during 1 h, then at 300 V. The staining conditions were adapted from Vallejos (1983). Precise gel composition and staining solutions are described in Laurens (1986).

Nine progenies, each of about 40 plants, were analysed. Five were progenies from controlled crosses made in an isolation cage, and had not been subjected to any selection.

Self progeny from 'Golden Delicious': G. ♀
'Golden Delicious' × 'T.N.R.': G. × T.N.R.
'Golden Delicious' × 'White Angel': G. × W.A.
'Early Red One' × 'T.N.R.': E. × T.N.R.
'Early Red One' × 'White Angel': E. × W.A.

For the other progenies, pollinations were made without isolation cage and all the analysed plants had been selected for resistance to scab (*Venturia inaequalis*). Most parents are hybrids, still undergoing selection:

'Liberty' × 'X 3191': L. × X 91
'Liberty' × 'X 3174': L. × X 74
'P20 R18 A71' × 'X 3174': P20 × X 74
'P22 R25 A32' × 'X 3174': P22 × X 74

Grafted plants of the parents of each progeny were grown in the greenhouse, and their leaves were analysed.

Two different hypotheses were tested to interpret each enzyme segregation: (1) disomic inheritance, involving one or two genes, and (2) tetrasomic inheritance; because only bivalent pairing can be seen at meiosis (Lespinasse 1973), chromosomal segregation was postulated.

## Results

### *Acid phosphatase*

Only the region most distant from the origin was interpreted in acid phosphatase zymograms. Three bands were seen; the corresponding alleles are named $Ap^a$, $Ap^b$ and $Ap^c$ (Fig. 1). Results are shown in Tables 1 and 6.

Heterozygous phenotypes have only 2 bands, thus indicating a monomeric structure of these isozymes. A monogenic disomic inheritance is in agreement with all the observed segregations.

Seven segregations involving only one or two alleles can also be explained by tetrasomic inheritance. However in P20 × X 74 and P22 × X 74 progenies, a phenotypic class [a b c] would be expected under a tetrasomic hypothesis. Since this class was never observed, tetrasomic inheritance can be ruled out. In the progeny from P20 × X 74, five [a] phenotypes cannot be explained by any of the hypotheses. These plants may have arisen from either accidental selfing or outcrossing.

### *Endopeptidase*

Only one zone was seen in endopeptidase zymograms. Four band positions governed by the alleles $Enp^a$, $Enp^b$, $Enp^{b'}$ and $Enp^c$ were identified (Fig. 2). Based on the observed phenotypes, endopeptidases seem to have a monomeric structure. Segregation data are shown in Tables 2 and 6. Monogenic inheritance accounts for all the observed segregations. Tetrasomic inheritance does

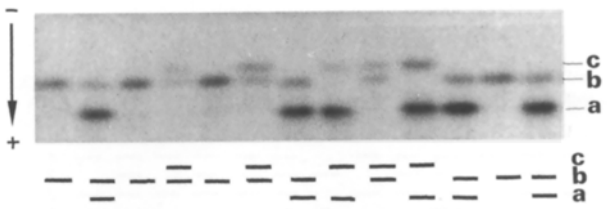


**Fig. 1.** Acid phosphatase zymograms of the progeny P22 × X 74

**Table 1.** Segregation data for AP

| Parental phenotypes | | Phenotypes in progeny | | | | | | $\chi^2$ | |
|---|---|---|---|---|---|---|---|---|---|
| ♀ | ♂ | [a] | [b] | [c] | [ab] | [bc] | [ac] | 1:1 | 1:1:1:1 |
| [b] | × [b] | | | | | | | | |
| G. | × T.N.R. | – | 40 | – | – | – | – | – | – |
| E. | × T.N.R. | – | 40 | – | – | – | – | – | – |
| L. | × X 91 | – | 37 | – | – | – | – | – | – |
| G♂ | | – | 34 | – | – | – | – | – | – |
| [b] | × [a] | | | | | | | | |
| G. | × W.A. | – | – | – | 44 | – | – | – | – |
| E. | × W.A. | – | – | – | 40 | – | – | – | – |
| L. [b] | × X 74 [bc] | – | 21 | – | – | 16 | – | 0.68 | – |
| P22 [ab] | × X 74 [bc] | – | 11 | – | 10 | 10 | 7 | – | 0.95 |
| P20 [ac] | × X 74 [bc] | [5] | – | 4 | 9 | 13 | 7 | – | 5.18 |

All $\chi^2$ tests are nonsignificant $P(\alpha) > 0.05$

**Table 2.** Segregation data for ENP

| Parental phenotypes | | Phenotypes in progeny | | | | | | | $\chi^2$ | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| ♀ | ♂ | [a] | [b] | [ab] | [bb′] | [ab′] | [bc] | [ac] | 1:1 | 2:1:1 | 1:1:1:1 |
| G. ♀ [a] | | 34 | – | [4] | – | – | – | [1] | – | – | – |
| E. [b] | × T.N.R. [b] | – | 39 | – | – | – | – | – | – | – | – |
| G. [a] | × T.N.R. [b] | – | – | 40 | – | – | – | – | – | – | – |
| E. [b] | × W.A. [bb′] | – | 20 | – | 19 | – | – | – | 0.03 | – | – |
| P22 [b] | × X 74 [ab] | – | 21 | 17 | – | – | – | – | 0.42 | – | – |
| P20 [ab] | × X 74 [ab] | 10 | 11 | 17 | – | – | – | – | – | 0.47 | – |
| G. [a] | × W.A. [bb′] | – | – | 22 | – | 17 | – | – | 0.64 | – | – |
| [bc] | × [ab] | | | | | | | | | | |
| L. | × X 91 | – | 12 | 11 | – | – | 6 | 8 | – | – | 2.46 |
| L. | × X 74 | – | 13 | 7 | – | – | 10 | 8 | – | – | 2.21 |

All $\chi^2$ tests are nonsignificant $P(\alpha) > 0.05$

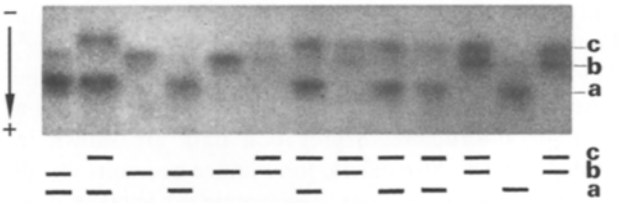


**Fig. 2.** Endopeptidase zymograms of the progeny L. × X 74

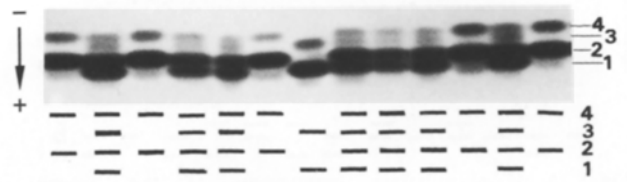


**Fig. 3.** Esterase zymograms of the progeny L. × X 91

**Table 3.** Segregation data for EST

| Parental phenotypes | | Phenotypes in progeny | | | $\chi^2$ |
|---|---|---|---|---|---|
| ♀ | ♂ | [1.3] | [2.4] | [1.2.3.4] | 1:1 |
| G. ♀ [2.4] | | – | 34 | – | – |
| P22 [1.3] × X 74 [2.4] | | [1] | – | 37 | – |
| [2.4] × [1.2.3.4] | | | | | |
| G. | × T.N.R. | – | 15 | 20 | 0.71 |
| G. | × W.A. | – | 20 | 20 | – |
| E. | × W.A. | – | 17 | 21 | 0.42 |
| E. | × T.N.R. | – | 19 | 20 | 0.02 |
| [1.2.3.4] × [2.4] | | | | | |
| L. | × X 91 | [1] | 19 | 17 | 0.11 |
| L. | × X 74 | – | 20 | 17 | 0.24 |
| P20 | × X 74 | [2] | 17 | 19 | 0.11 |

All $\chi^2$ tests are nonsignificant $P(\alpha) > 0.05$

not fit the observed segregation pattern of P20 × X 74, G. × W.A., L. × X 91 and L. × X 74 progenies, because of the lack of several phenotypic classes.

Five plants from G. ♀ progeny exhibited extra bands, which must have originated from outcrossing.

*Esterase*

Esterase zymograms can be divided into three zones; only the fastest one is interpreted. Four bands were seen (Fig. 3) and can be grouped in pairs; the phenotype [1.3] is associated with the allele $Est^a$ and the phenotype [2.4] with the allele $Est^b$. The four banded heterozygous phenotype indicates a monomeric structure for these isozymes. Results can be seen in Tables 3 and 6.

The observed segregations are very simple; they can be explained easily by monogenic disomic inheritance, and also by tetrasomic inheritance. Among all the progenies, four plants exhibited an unexpected [1.3] phenotype. As the bands one and three are absent from their male parent zymograms, those plants certainly came from accidental outcrossing or selfing of the female parent.

*Phosphoglucoisomerase*

All the observed plants (parents and progenies) exhibited the same three banded phenotype (Fig. 4). A dimeric structure of this enzyme can be postulated, in agreement with numerous reports in other plant species. This unique pattern is controlled by the two alleles $Pgi^a$ and $Pgi^b$, the central band being the heterodimer.

The absence of segregation in progenies, either by crossing or selfing, can be explained only by the hypothesis of a duplicated gene with a disomic inheritance. Each of the genes being homozygous for one al-

lele, a fixed heterozygosity is created. Tetrasomic inheritance is incompatible with the observed segregations.

*Superoxide dismutase*

Superoxide dismutase zymograms are complex and only one zone can be interpreted. Two phenotypes can be seen (Fig. 4); the three banded one [1.3.5] is supposed to be controlled by the allele $Sod^a$; the five banded one [1.2.3.4.5] is supposed to be heterozygous, the allele $Sod^b$ coding for the bands 2.4. In spite of the symmetry of the diagrams, these superoxide dismutases seem monomeric. Results are shown in Tables 4 and 6.

Because of the absence of segregation in the progenies P20×X 74, P22×X 74 and L.×X 74, a monogenic disomic interpretation is impossible. The lack of [2.4] phenotypes can be explained by a bigenic disomic interpretation, one of the genes being fixed for the allele $Sod^a$. Tetrasomic inheritance is also compatible with the very simple observed segregations.

Two unexpected [1.3.5] phenotypes were observed, one in the P20×X 74 progeny, the other in the P22×X 74 progeny. Those two plants probably do not belong to the progeny.

*Peroxidase*

The ten band peroxidase zymograms can be simplified by grouping bands that are always present simultaneously into four groups (Fig. 5), each under the control of one allele. This indicates a monomeric structure for these peroxidases. Results are presented in Tables 5 and 6.

A monogenic disomic hypothesis is incompatible with the presence of phenotypes controlled by three or four alleles in several progenies (G.×T.N.R., E.×T.N.R., and L.×X 91). Bigenic disomic inheritance would provide the following explanation:

Group A controlled by $Per\ 1^a$
Group B controlled by $Per\ 1^b$ locus *Per 1*
Group C controlled by $Per\ 2^c$
Group D controlled by $Per\ 2^d$ locus *Per 2*

**Table 4.** Segregation data for SOD

| Parental phenotypes ♀ | ♂ | Phenotypes in progeny [1.3] | [1.2.3.4.5] | $\chi^2$ 1:1 | 3:1 |
|---|---|---|---|---|---|
| [1.3]×[1.3] | | | | | |
| E. | ×W.A. | 38 | – | – | – |
| E. | ×T.N.R. | 39 | – | – | – |
| [1.3]×[1.2.3.4.5] | | | | | |
| P20 | ×X 74 | [1] | 37 | – | – |
| P22 | ×X 74 | [1] | 37 | – | – |
| [1.2.3.4.5]×[1.3] | | | | | |
| G. | ×T.N.R. | 22 | 18 | 0.40 | – |
| G. | ×W.A. | 16 | 23 | 1.25 | – |
| [1.2.3.4.5]×[1.2.3.4.5] | | | | | |
| L. | ×X 74 | – | 37 | – | – |
| L. | ×X 91 | 7 | 37 | – | 0.73 |
| G♀ | ♂ | 12 | 22 | – | 1.92 |

All $\chi^2$ tests are nonsignificant $P(\alpha) > 0.05$

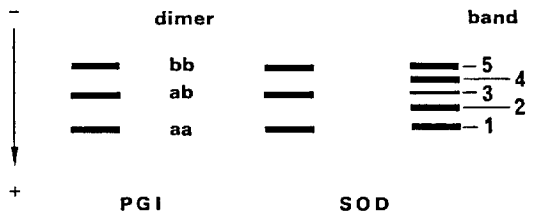


**Fig. 4.** Schematic representation of phosphoglucoisomerase zymograms: only one phenotype was observed, and superoxide dismutase: two phenotypes were observed

**Table 5.** Segregation data for PER

| Parental phenotypes ♀ | ♂ | Phenotypes in progeny [B] | [BC] | [CD] | [BCD] | [ABD] | [ABCD] | [ACD] | $\chi^2$ 1:1 | 3:1 | 2:1:1 |
|---|---|---|---|---|---|---|---|---|---|---|---|
| G. | ♂ [BC] | 12 | 22 | – | – | – | – | – | – | 1.92 | – |
| E. [B] | × W.A. [B] | 40 | – | – | – | – | – | – | – | – | – |
| [BC] | × [B] | | | | | | | | | | |
| G. | × W.A. | 27 | 15 | – | – | – | – | – | 3.43 | – | – |
| P20 | × X 74 | 20 | 18 | – | – | – | – | – | 0.10 | – | – |
| P22 | × X 74 | 17 | 21 | – | – | – | – | – | 0.42 | – | – |
| G. [BC] | × T.N.R. [AD] | – | – | – | – | 25 | 15 | – | 2.50 | – | – |
| E. [B] | × T.N.R. [AD] | – | – | – | – | 39 | – | – | – | – | – |
| L. [BC] | × X 74 [B] | – | 36 | [1] | – | – | – | – | – | – | – |
| L. [BC] | × X 91 [BD] | – | 25 | 7 | 5 | – | – | – | – | – | 4.78 |

All $\chi^2$ tests are nonsignificant $P(\alpha) > 0.05$

The existence of a null allele at the locus *Per 2* is deduced from the presence of [B] phenotypes and from several segregations. All the observed segregations are in agreement with this bigenic interpretation, except L.×X 91. The hypothesis of a linkage between the two loci and the presence of a null allele *Per 1*$^n$ in the L. genotype can explain the 3 observed classes in this progeny, and is not incompatible with the other results.

Tetrasomic inheritance was also tested: this hypothesis is incompatible with two segregations (G.×T.N.R., E.×T.N.R.), because of the lack of several phenotypic classes. Tetrasomic pattern is very unlikely with segregation of L.×X 91.

Only one plant from the progeny L.×X 91 exhibited an extra band indicating an outcrossing origin.

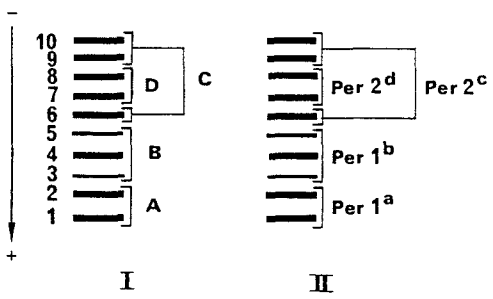


**Fig. 5.** Schematic representation of peroxidase zymograms: I: position of bands and groups of bands, II: designation of alleles according to bigenic disomic hypothesis

**Table 6.** Genotypes of the parents, according to disomic hypothesis

| Locus | *Ap* | *Enp* | *Est* | *Sod 1, Sod 2* | *Per 1, Per 2* |
|---|---|---|---|---|---|
| G. | *bb* | *aa* | *bb* | *aa, ab* | *bb, cn* |
| E. | *bb* | *bb* | *bb* | *aa, aa* | *bb, nn* |
| T.N.R. | *bb* | *bb* | *ab* | *aa, aa* | *aa, dd* |
| W.A. | *aa* | *bb'* | *ab* | *aa, aa* | *bb, nn* |
| L. | *bb* | *bc* | *ab* | *aa, ab* | *bn, cc* |
| X 74 | *bc* | *ab* | *bb* | *aa, ab* | *bb, nn* |
| X 91 | *bb* | *ab* | *bb* | *aa, ab* | *bn, nd* |
| P20 | *ac* | *ab* | *ab* | *aa, aa* | *bb, cn* |
| P22 | *ab* | *bb* | *aa* | *aa, aa* | *bb, cn* |

## Discussion

By analysing leaf samples, young seedlings from five unselected progenies could be studied. In the four other progenies, no significant deviation from expected ratios was observed, although they had been previously selected for scab resistance. The total number of plants analysed per progeny is quite limited (40), but the small number of phenotypic classes per progeny (1 to 4) permitted interpretation of the segregations.

Among 360 analysed plants, 15 had an unexpected phenotype which can be explained either by outcrossing or by accidental self-pollination. Two types are distinguished: among the five progenies from strictly controlled pollinations, only one contains five "out-cross" plants which remain difficult to explain. Each of the other four progenies with a low level of pollination control, contain some plants with an unexpected phenotype, indicating a ratio of 6% "out cross" plants.

The results of this study are interpreted in conjunction with the results obtained previously from pollen analysis (Chevreau et al. 1985) (Table 7). All the results are in agreement with a disomic inheritance hypothesis.

Monogenic inheritance is clearly demonstrated for two enzymes: the same gene *Est 1* is active in pollen

**Table 7.** Summary of isozyme inheritance in pollen and leaves

| Enzyme | Analysed on | | Structure | Genes | | Alleles | |
|---|---|---|---|---|---|---|---|
| | Pollen | Leaves | | No. | Type | Active | Null |
| EST 1 | × | × | monomeric | 1 | – | 2 | – |
| IDH | × | | dimeric | 1 | – | 2 | – |
| AP | × | | monomeric | 2 | duplicated | 2 | – |
| | | × | monomeric | 1 | – | 3 | – |
| ENP | × | | monomeric | 2 | duplicated | 2 | – |
| | | × | monomeric | 1 | – | 3 | – |
| PGI | × | × | dimeric | 2 | duplicated | 4 | – |
| ADH | × | | dimeric | 2 | duplicated | 2 | – |
| SOD | | × | monomeric | 2 | duplicated | 2 | – |
| EST 2 | × | | monomeric | 2 | duplicated | 2 | 1 |
| PGM | × | | monomeric | 2 | independent | 4 | – |
| PER | | × | monomeric | 2 | linked | 4 | 2 |

and leaves, each allele controlling a two banded phenotype; the faster zone of IDH zymograms is controlled by one gene in pollen.

Differences between pollen and leaf results exist for two monomeric enzymes: ENP and AP. In both cases, leaf and pollen zymograms are very similar but the genetic interpretation is different: leaf results are in agreement with a monogenic inheritance, with three alleles, whereas pollen results are more complicated and require the hypothesis of two homoeologous genes with two identical alleles. Some varieties have different patterns in leaf and pollen extracts, for ENP or AP. This confirms a difference of genetic control or gene expression between the two organs.

Bigenic inheritance is established for all of the other enzymes, with either duplicated, independent, or linked genes. PGI is a dimeric enzyme with a three banded phenotype both in pollen and leaves. This identity and the observation of a "fixed heterozygosity" can be explained only by the existence of a pair of duplicated genes. The same interpretation is proposed for ADH, in pollen. The enzymes SOD in leaves, and EST 2 in pollen are monomeric. The results support the hypothesis of duplicated genes in both cases. The existence of a null allele at the EST 2 locus is postulated. PGM is a monomeric enzyme active in pollen, whose complicated patterns can be explained with two independent genes carrying either identical or different alleles. PER analysed on leaves is also monomeric. A linkage between two different genes, each of them carrying two active alleles and a null allele is postulated. These results are very similar to those obtained by Misic et al. (1981) on apple bark peroxidases.

Tetrasomic inheritance hypothesis was tested with all the observed segregations. In some simple cases (EST 1, EST 2, IDH and SOD), this hypothesis was not incompatible with the results. However, segregations for PER, PGI, PGM, ADH, and ENP and AP in leaves, could not be explained by a tetrasomic inheritance. No evidence of typical tetrasomic segregation was obtained in either study.

### *Conclusion*

Apple pollen and leaf isozymes analysed in these studies have a disomic pattern of inheritance. This is in accordance with the diploid meiotic behavior of the species. Duplication of isozyme loci are known to occur in diploid species (Gottlieb 1982; Weeden 1983). However, the polyploid origin of apple genome is well established. In this case, the existence of several duplicated genes and also single genes is in agreement with the hypothesis of an allopolyploid origin of apple genome: some isozymes retain their duplicated control due to homoeologous genes, while some lose this duplicated expression through gene-silencing in the course of evolution. This isozymic study leads to the same conclusion as other studies using cytology (Sax 1933; Zhang et al. 1987), morphology (Stebbins 1950), or phenolic biochemistry (Challice 1981).

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## References

Breese EL, Thomas AC (1976) Monitoring chromosome segregation in *L. multiflorum* × *L. perenne* amphiploids. Rep Welsh Plant Breed Stn For, pp 29–32

Challice JS (1981) Chemotaxonomic studies in the family *Rosaceae* and the evolutionary origins of the subfamily *Maloideae*. Preslia 53:289–304

Chevreau E, Lespinasse Y, Gallet M (1985) Inheritance of pollen enzymes and polyploid origin of apple (*Malus* × *domestica* Borkh.). Theor Appl Genet 71:268–277

Darlington CD, Moffett AA (1930) Primary and secondary chromosome balance in *Pyrus*. J Genet 22:129–151

Gottlieb LD (1982) Conservation and duplication of isozymes in plants. Science 216:373–380

Laurens F (1986) Etude du déterminisme de quelques systèmes enzymatiques chez une espèce d'origine polyploïde: le pommier (*Malus* × *domestica* Borkh.) DEA Université de Rennes I, 50 pp

Lespinasse Y (1973) Chromosomic observations in *Malus* and *Pyrus*. In: Proc Eucarpia Fruit Sect Symp V. Canterbury, pp 75–83

Misic P, Vinterhalter D, Todorovic R (1980) The inheritance of isoperoxidase in apple. Jugosl Vocarstvo 14:43–47

Sax K (1931) The origin and relationships of the *Pomoideae*. J Arnold Arbor Harv Univ 12:3–22

Sax K (1933) The origin of the *Pomoideae*. J Am Soc Hortic Sci 30:147–150

Stebbins G (1950) Polyploidy and the origin of higher categories. In: Variation and evolution in plants. Columbia University Press, New York, pp 359–362

Vallejos CE (1983) Enzyme activity staining. In: Tanksley SD, Orton LJ (eds) Isozymes in plant genetics and breeding, Part A. Amsterdam, pp 469–490

Weeden NF (1983) Evolution in plant isozymes. In: Tanksley SD, Orton LJ (eds) Isozymes in plant genetics and breeding, Part A. Amsterdam, pp 177–208

Zhang YX, Salesses G, Lespinasse Y (1987) Etude cytogénétique de quelques clones diploïdes et polyloïdes de Pommier (*Malus* × *domestica* Borkh.): considérations quant à l'origine du Pommier. Cytologia (in press)