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PHENOLICS OF *PYRUS* INTERSPECIFIC HYBRIDS*

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Key Word Index—*Pyrus*; Rosaceae; leaf; phenolic compounds; flavonoids; phenolic acids; cinnamic acids; interspecific crosses; inheritance.

Abstract—The leaf phenolics of a number of *Pyrus* interspecific hybrids (from controlled hand-crosses) and the parental individuals have been examined. In most instances it was found that the presence of certain phenolics in the hybrids is of diagnostic value in predicting the identities of one or both of the original parents. The flavone and flavonol glycosides appear to be inherited as simple dominant characters (whether inherited from male or female parents) but phenolics such as epicatechin, catechin, caffeoylcalleryanin and *p*-hydroxybenzoic acid did not always appear to show such simple dominance.

INTRODUCTION

MUCH of the *Pyrus* material present in the major botanic gardens and arboreta is considered to be of uncertain hybrid origin[1-3] and in some instances the investigations of the present author[4-7] on the leaf phenolics of authentic wild *Pyrus* species have enabled diagnoses to be made of the possible parentage of these hybrids. All wild *Pyrus* species are diploid with $2n = 34$ and there is a high degree of interspecific fertility within the genus.[8] Westwood and Bjornstad[9] have examined the fruit characteristics of a number of authentic *Pyrus* interspecific hybrids which proved fertile. They found that persistent calyx is dominant over deciduous, russet skin is dominant over smooth, stoneless flesh is dominant over stony and that the carpel number is intermediate between the two parent species. The investigations of Catlin and Olsson[10] have shown that phenolics can sometimes be of use in the identification of *Pyrus* interspecific hybrids but their work did not extend to the identification of the phenolics detected on their chromatograms. The object of the present investigations is to examine the known leaf phenolics of a number of *Pyrus* interspecific hybrids (from controlled hand-crosses) together with the actual parental individuals. By these investigations it should

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[1] M. N. WESTWOOD, *Hort. Sci.* **1,** 85 (1966).
[2] M. N. WESTWOOD, M. M. THOMPSON and P. B. LOMBARD, *Amer. Pom. Soc. Fruit Var. Hort. Digest* **25,** 87 (1971).
[3] M. N. WESTWOOD, private communications.
[4] J. S. CHALLICE and A. H. WILLIAMS, *Phytochem.* **7,** 119 (1968).
[5] J. S. CHALLICE and A. H. WILLIAMS, *Phytochem.* **7,** 1781 (1968).
[6] J. S. CHALLICE and A. H. WILLIAMS, *Phytochem.* **9,** 1271 (1970).
[7] J. S. CHALLICE and M. N. WESTWOOD, *Phytochem.* **11,** 37 (1972).
[8] Q. B. ZIELINSKI and M. M. THOMPSON, *Bot. Gaz.* **128,** 109 (1967).
[9] M. N. WESTWOOD and H. I. BJORNSTAD, *Bull. Torrey Bot. Club* **98,** 22 (1971).
[10] P. B. CATLIN and E. A. OLSSON, *Proc. Am. Soc. Hort. Sci.* **93,** 88 (1968).

be possible to determine whether the phenolics concerned are inherited in a simple dominant manner in the hybrid material. Ideally one should examine large numbers of seedling progeny from the various crosses in order to arrive at the Mendelian ratios; unfortunately this was not possible owing to the limited material available.

RESULTS AND DISCUSSION

Table 1 presents the phenolics distribution data for nine authentic interspecific hybrids, together with the actual parental individuals (female parent listed first). In most instances it was found that the presence of certain phenolics in the hybrids is of diagnostic value in predicting the identities of one or both of the original parents. The flavone and flavonol glycosides appear to be inherited as simple dominant characters (whether inherited from

TABLE 1. LEAF PHENOLICS OF *Pyrus* INTERSPECIFIC HYBRIDS FROM CONTROLLED CROSSES (female parent listed first, male parent second)

Specimen	Flavone glycosides							
	F3	F2	F1	F4B	F4A	FO	FZ	FS
Dimorphophylla W6	+(+)	++	+(+)	o	o	o	+	o
Fauriei W6	o	o	o	o	o	o	o	o
Dimorph. W6 × faur. W6	+	++	+(+)	o	o	o	+	o
Communis cv. old home	o	o	o	o	o	o	o	o
Calleryana 2	+	++	+	o	o	o	t	o
Comm. cv. old home × call. 2	+	+(+)	+	o	o	o	t	o
Betulaefolia 1	o	o	o	o	o	o	o	o
Communis cv. farmingdale	o	o	o	o	o	o	o	o
Betul. 1 × comm. cv. farmingdale	o	o	o	o	o	o	o	o
Fauriei W6	o	o	o	o	o	o	o	o
Betulaefolia 1	o	o	o	o	o	o	o	o
Faur. W6 × betul. 1	o	o	o	o	o	o	o	o
Betulaefolia 2	o	o	o	o	o	o	o	o
Calleryana 5	t	++	+	o	o	o	t	o
Betul. 2 × call. 5	t	++	+	o	o	o	t	o
Amygdaliformis var. persica	o	o	o	o	o	o	o	o
Betulaefolia EW6	o	o	o	o	o	o	o	o
Amyg. var. persica × betul. EW6	o	o	o	o	o	o	o	o
Betulaefolia 2	o	o	o	o	o	o	o	o
Communis cv. farmingdale	o	o	o	o	o	o	o	o
Betul. 2 × comm. cv. farmingdale	o	o	o	o	o	o	o	o
Dimorphophylla W6	+(+)	++	+(+)	o	o	o	+	o
Calleryana W6/QBZ	t	+(+)	t	o	o	o	o	o
Dimorph. W6 × call. W6/QBZ	t	+(+)	t	o	o	o	+	o
Ussuriensis W6	o	o	o	+(+)	+ +	|	o	+
Calleryana W6/QBZ	t	+(+)	t	o	o	o	o	o
Ussur. W6 × call. W6/QBZ	+	+(+)	o	+	+(+)	(+)	o	+

Specimen	F7	U1,2	CPl,2	C3,4	A2	A3	L1-5
Dimorphophylla W6	+(+)	o	++	o	o	t	o
Fauriei W6	o	o	o	o	o	o	o
Dimorph. W6 × faur. W6	++	o	o	o	o	o	o
Communis cv. old home	o	o	o	+	+(+)	o	o
Calleryana 2	+	o	++	o	o	o	o
Comm. cv. old home × call. 2	+(+)	o	++	o	(t)	o	o
Betulaefolia 1	+(+)	+	o	o	o	o	+ +
Communis cv. farmingdale	o	o	o	t	+	(t)	o
Betul. 1 × comm. cv. farmingdale	++	o	o	+	t	o	+ +
Fauriei W6	o	o	o	o	o	o	o
Betulaefolia 1	+(+)	+	o	o	o	o	+ +
Faur. W6 × betul. 1	++	+	o	o	o	o	+(+)
Betulaefolia 2	+	+(+)	o	t	o	o	t
Calleryana 5	+	o	+++	t	o	o	o
Betul. 2 × call. 5	+	o	++	t	o	o	o
Amygdaliformis var. persica	o	t	o	t	t	(t)	o?
Betulaefolia EW6	(+)	+(+)	o	+	o	o	t
Amyg. var. persica × betul. EW6	++	+	o	t	o	o	t
Betulaefolia 2	+	+(+)	o	t	o	o	t
Communis cv. farmingdale	++	o	o	+	+	(t)	o
Betul. 2 × comm. cv. farmingdale	++	+	o	t		(t)	o
Dimorphophylla W6	+(+)	o	++	o	o	t	o
Calleryana W6/QBZ	(+)	o	++(+)	o	o	o	o
Dimorph. W6 × call. W6/QBZ	(+)	o	++	o	o	+	o
Ussuriensis W6	+	o	o	o	o	t	o
Calleryana W6/QBZ	(+)	o	++(+)	o	o	o	o
Ussur. W6 × Call. W6/QBZ	o	o	+(+)	o	o	o	o

TABLE 1—*cont.*

Specimen	Flavone glycosides									
	P1	P2	P4	P6	P5	P10	C1, 2	A1	F5	F6
Dimorphophylla W6	o	o	o	o	+	+	++	+	++	++
Fauriei W6	o	o	o	o	o	o	+	+	++	+++
Dimorph. W6 × Faur. W6	o	o	o	o	o	o	+(+)	+(+)	++	++
Communis cv. old home	o	o	o	o	o	o	++	++	++	++
Calleryana 2	+++	+	++	++	+	o	++	o	+	+(+)
Comm. cv. old home × call. 2	++	+	++	+(+)	+	o	++	++	++	++
Betulaefolia 1	o	o	o	o	o	o	+(+)	++	++	++
Communis cv. farmingdale	o	o	o	o	o	o	++	++	++	++
Betul. 1 × comm. cv. farmingdale	o	o	o	o	o	o	+++	++	++	++
Fauriei W6	o	o	o	o	o	o	+	+	++	+++
Betulaefolia 1	o	o	o	o	o	o	+(+)	++	++	++
Faur. W6 × betul. 1	o	o	o	o	o	o	++	++	+++	++(+)
Betulaefolia 2	o	o	o	o	o	o	t	+	++	++
Calleryana 5	+++	+(+)	++	++	+(+)	o	+	o	t	(+)
Betul. 2 × call. 5	++	o	++	+(+)	+	o	+	+(+)	++	++
Amygdaliformis var. persica	o	o	o	o	o	o	+(+)	+	++	++
Betulaefolia EW6	o	o	o	o	o	o	++	++	++	++
Amyg. var. persica × betul. EW6	o	o	o	o	o	o	++	+(+)	++	++
Betulaefolia 2	o	o	o	o	o	o	(t)	+	++	++
Communis cv. farmingdale	o	o	o	o	o	o	+++	++	++	++
Betul. 2 × comm. cv. farmingdale	o	o	o	o	o	o	+(+)	++	+(+)	++
Dimorphophylla W6	o	o	o	o	+	+	++	+	++	++
Calleryana W6/QBZ	+++	+	++	++	+	o	+(+)	o	t	+
Dimorph. W6 × call. W6/QBZ	+++	+	++	++	+	o	+(+)	o	t	+
Ussuriensis W6	o	o	o	o	o	o	++	++	++	+(+)
Calleryana W6/QBZ	+++	+	++	++	+	o	+(+)	o	t	+
Ussur. W6 × call. W6/QBZ	++	+	+(+)	+(+)	+	o	++	t	++	+(+)

Scoring code: o = absent, t = trace amount only (diazo/Gibbs colour only just visible), + = small amount (weak but noticeable diazo/Gibbs colour), ++ = moderate amount (strong diazo/Gibbs colour), +++ = large amount (exceptionally strong diazo/Gibbs colour), ( ) = reservations regarding enclosed symbol—score on low side. Scoring codes are based upon strengths of two dimensional chromatogram spots (SBA or TPW and 2% HAc solvents run *ca.* 35 cm in each direction). Chromatograms loaded with the extract of 50 mg fresh leaf tissue.

Phenolic codes: F3 = apigenin 7-glucoside, F2 = luteolin 7-glucoside, F1 = luteolin 7-rhamnosylglucoside, F4B = apigenin 4′-glucoside, F4A = luteolin 4′-glucoside, FO = suspected apigenin 7,4′-diglucoside, FZ = chrysoeriol 7-glucoside, FS = suspected luteolin 5-methyl ether, F7 = flavonol 3-triglycosides?, U1, 2 = epicatechin and catechin, CP1, 2 = caffeoylcalleryanin, C3, 4 = caffeoylarbutin, A2 = acetylarbutin (pyroside), A3 = 4-allylphenol? L1-5 = leucoanthocyanidins, P1 = protocatechuoylcalleryanin, P2 = vanilloylcalleryanin, P4 = *p*-hydroxybenzoylcalleryanin, P6 = protocatechuic acid 3-glucoside, P5 = *p*-hydroxybenzoic acid, P10 = glucose ester of latter?, C1, 2 = isochlorogenic acid, A1 = arbutin, F5 = flavonol 3-monoglycosides, F6 = flavonol 3-diglycosides.

Chlorogenic and neochlorogenic acids were present in all leaf samples.

male or female parents) but phenolics such as epicatechin and catechin (U1, 2), caffeoylcalleryanin (CP1, 2) and *p*-hydroxybenzoic acid (P5) do not always appear to show such simple dominance. For example, caffeoylcalleryanin is absent from a *P. dimorphophylla* × *P. faurieri* cross and the *P. fauriei* male parent but present in the *P. dimorphophylla* female parent. In four crosses involving *P. calleryana* in each case as the male parent, the entire range of caffeoyl, protocatechuoyl, vanilloyl and *p*-hydroxybenzoyl esters of calleryanin, together with protocatechuic acid 3-glucoside, was inherited from *P. calleryana* by the respective hybrids as simple dominant characters. It is of interest to note here that a suspected *Pyrus* hybrid (in which the phenolic esters of calleryanin and protocatechuic acid 3-glucoside could not be detected), upon self-fertilization gave rise to three seedlings one of which was found to contain abundant amounts of the latter phenolic acid derivatives.[11] It is evident that the genes for these phenolics were hidden in a recessive condition in the original hybrid. Complex leucoanthocyanidins, as leaf constituents, showed species differences and were shown in one hybrid to originate from the female parent and in another

[11] J. S. CHALLICE, *Rep. Long Ashton Res. Stn* (1971).

hybrid to originate from the male parent. In practice the use of phenolics in the identification of *Pyrus* hybrids has proved to be a valuable adjunct to the use of hybrid fruit characters.[9]

EXPERIMENTAL

Full details, including two dimensional chromatographic maps of the phenolics and details of identification studies, will be found in previous papers of this series.[4–7] The fresh leaf material was obtained from Professor M. N. Westwood of Oregon State University, U.S.A.

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