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SHORT COMMUNICATION

METABOLIC PATHWAYS AND SYNTHETIC ABILITY OF LEAVES OF SEVERAL *PYRUS* SPECIES

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Abstract—Relationship between type of metabolism and synthetic ability of leaves of several pear species was investigated. Species incorporating high quantities of labelled glucose into the alcohol insoluble fraction of the leaf respired through the Embden–Meyerhof–Parnas glycolitic pathway and citric-acid-cycle sequence. Species incorporating small amounts of glucose respired mostly through the pentose phosphate pathway. As leaves aged, their synthetic ability decreased, and respiration changed toward the pentose phosphate pathway. When species (having leaves of the same age) were compared, respiration through the pentose phosphate pathway seemed to be associated with small leaf size. It was concluded that the type of respiration in leaves was determined by the energy requirement for synthetic processes. When the energy requirement was low, metabolism of leaves proceeded through the pentose pathway. The synthetic ability in turn was influenced genetically, by environment, or by the age of the leaf.

INTRODUCTION

ATTEMPTS have been made in the past to establish changes in the metabolic pathways in tissues of higher plants initiated by various physiological conditions. The change from the Embden-Meyerhof-Parnas glycolitic pathway to the pentose phosphate pathway (PPP) can be caused by severed water uptake into the tissues,[1,2] by infection of plants by an obligate parasite,[3,4] or by subjecting plants to a high concentration of growth regulators.[3] Changes in metabolic pathways caused by ontogenetic changes, age of the leaf,[5] ripening of fruits,[6,7] and maturing of seeds[8] also have been reported. Data concerning types of metabolic pathways associated with genetic charecters, however, are not available. Contribution of PPP to the total respiration is estimated by the so called C_6/C_1 ratio. Although the estimation of PPP activity by this method may not be precise, it is a good indication of the metabolic status of the leaf and therefore it is used in this paper.

RESULTS AND DISCUSSION

During the investigation of resistance in pears to fire blight (a bacterial disease), we discovered that the type of metabolic pathway found in different species and cultivars was

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associated with incorporation of ^{14}C of labelled glucose into the alcohol insoluble fraction (AIF) of the leaf (Table 1). Incorporation of either carbon 1 or 6 of glucose into the AIF of the leaf was low for *Pyrus salicifolia* Balb., *P. betulaefolia* Bunge, and *P. communis* Linn. The relative contribution of the PPP to the total CO_2 production, as it was measured by the C_6/C_1 ratio, was relatively high in the leaves of the above species. The ratio seemed to correspond to the degree of incorporation. *P. salicifolia* had the lowest rate of incorporation into the AIF (2 per cent of total uptake) and also the lowest C_6/C_1 ratio (0·32). *P. betulaefolia* and *P. communis* had slightly higher rates of incorporation and also somewhat higher respiratory ratios. All the other species tested had a high rate of incorporation into the AIF and higher C_6/C_1 ratios.

TABLE 1. RESPIRATORY MECHANISM IN LEAVES OF SEVERAL *Pyrus* SPECIES

Species	Type of respiration C_6/C_1 ratio	Incorporation‡ of ^{14}C into alc. insol. fraction % of uptake
With large leaves		
P. ussuriensis Maxim.	0·86 (0·06)*	6·8 (0·32)*
P. calleryana Decne. cv. Bradford	0·75 (0·05)	5·4 (0·43)
P. serotina Rehd.	0·64 (0·02)	5·1 (0·24)
P. bretschneideri Rehd.	0·57 (0·03)	3·9 (0·36)
P. pashia Buch-Ham.	0·53 (0·03)	5·1 (0·58)
With small leaves		
P. betulaefolia Bunge	0·47 (0·02)	2·8 (0·36)
P. communis Linn.†	0·43 (0·06)	2·5 (0·27)
P. salicifolia Balb.	0·32 (0·05)	2·0 (0·60)

* Standard deviation.

† *P. communis* Linn. used in this experiment is a small leaf type collected from Bulgaria.

‡ Data given for glucose-6-^{14}C, incorporation of glucose-1-^{14}C was similar but slightly less.

If the relationship between the synthetic ability, as measured by incorporation of label into AIF of the leaf, and the type of metabolic pathway is genetically determined, it should be susceptible to confirmation under altered physiological conditions. To confirm such a relationship, leaves of 3 cultivars of *P. communis* (Bartlett, Old Home, and Magness) were used. Leaves were harvested at 3 stages of development: in a very young stage; at the time when they reached full expansion; and at maturity. As the leaves advanced in age, the incorporation of label from ^{14}C glucose into the AIF of leaves decreased and the C_6/C_1 respiratory ratio was lowered markedly (Table 2). An inverse relationship was found between synthetic ability and metabolism through the PPP (Fig. 1). One might expect an accelerated respiration through the Embden–Meyerhof–Parnas glycolitic pathway and citric-acid-cycle sequence at times when energy is required for synthetic processes. The fact that in certain cases respiratory ratios as low as 0·2 were obtained indicate that leaves can easily change the type of respiration when less energy is required, so that a high portion of total CO_2 is released through the PPP. When leaves of different species of the same physiological age were compared, the low incorporation into the AIF and the high respiration through the PPP were also expressed in small leaf size (Table 1). Comparisons between species having small

and large leaves, however, have to be made under well controlled conditions since environment as well as leaf age may affect the type of respiration.

TABLE 2. EFFECT OF AGE ON RESPIRATORY PATHWAYS OF PEAR CULTIVARS

Age of leaf	Bartlett	Old Home	Magness
	C_6/C_1 ratio in leaves		
Young	0·56	0·52	0·63
Fully expanded	0·33	0·32	0·47
Mature	0·19	0·16	0·27
	Incorporation of label into alc. insol. fraction % of uptake		
Young	6·40	7·60	8·70
Fully expanded	5·10	5·20	6·70
Mature	1·80	1·60	1·30

Distribution of data is shown in Fig. 1.

From the above investigations we concluded that the type of metabolism is influenced markedly by the synthetic processes in the leaf, which in turn can be influenced by the genetic makeup of the plant as well as by the environment or by the age of the leaf. We found no relationship between the type of metabolism and fire blight resistance of pears.

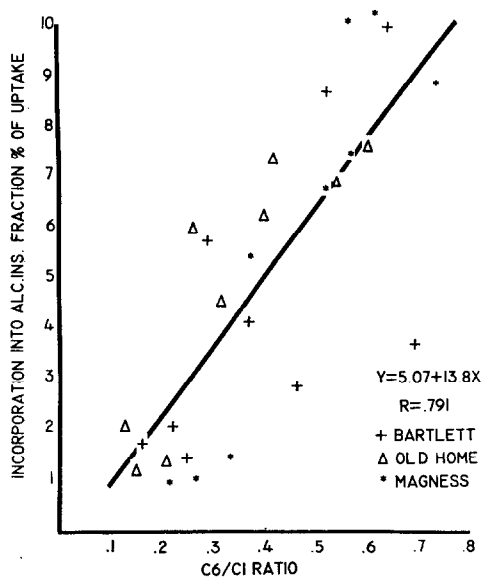

FIG. 1. RELATIONSHIP BETWEEN INCORPORATION OF GLUCOSE INTO THE ALCOHOL INSOLUBLE FRACTION OF LEAF AND TYPE OF RESPIRATION.

EXPERIMENTAL

Leaves of *Pyrus* species were collected from the orchard during the spring at the time when new leaves were fully expanded. Determinations were arranged so that the C_6/C_1 ratios within one replication were determined for all species on the same day. *P. communis* cultivars were grown in the greenhouse. Leaves from all three stages of leaf development were harvested at the same time.

Type of metabolism was determined by using the C_6/C_1 ratio. This method was originally designed by Bloom, Stetten and Stetten[9] and was reviewed by Gibbs.[10] For our determination 0·5 g of leaf disks of 8 mm diameter was used. Disks were suspended in 10 ml of 0·03 M phosphate buffer at pH 5·2 and 1 μc of ^{14}C-labelled glucose, labelled in either C_1 or C_6 position, was added to the flask. Specific activities of the sugars were 2·96 mc/mole for G-1-^{14}C and 4·9 mc/mole for G-6-^{14}C. CO_2 was collected in a multiple unit radiometric respirometer described elsewhere.[11] Immediately following 1 hr incubation, the tissue was extracted with 10 ml of 80 per cent (v/v) ethanol, filtered and both the ethanol soluble fraction and AIF counted. CO_2 was counted as $BaCO_3$. Activity obtained by counting $BaCO_3$, ethanol extract and AIF were added and the total was used as the total uptake by the tissue. The respiratory CO_2 and the AIF were expressed as per cent of uptake. C_6/C_1 ratios were determined by dividing the per cent of activity recovered as CO_2 for G-6-^{14}C by that of G-1-^{14}C.

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