Genotypic Influence on L-Ascorbic Acid Accumulation in Blackcurrant (Ribes nigrum) Fruit

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Materials and Methods

Plant Material and Growth Conditions
Ribes nigrum cultivars Hedda and Baldwin and genotype 8982-6 were grown in the field at Invergowrie, Dundee and subjected to standard commercial fertiliser and pesticide regimes. In the three years 2002/04 inclusive, ripe fruit AsA content was 71 ± 21, 196 ± 9 and 268 ± 25 mg g⁻¹ FW for Hedda, Baldwin and 8982-6, respectively.

Extraction and Measurement of AsA
AsA was extracted from fresh or lyophilised tissues in 5% metaphosphoric acid containing 5 mM tris[2-carboxyethyl]phosphine hydrochloride (TCEP). After centrifugation, total AsA was quantified in the supernatant by HPLC with diode array detection at 245 nm. Radioactive AsA was extracted in 5% perchloric acid containing 5 mM TCEP. [¹⁴C]AsA was partially purified on SAX cartridges prior to quantification by HPLC with radioflow detection.

Results

Changes in tissue AsA throughout growth cycle
- Phenotypic differences primarily confined to fruit
- Insufficient AsA storage to account for annual fruit accumulation

HPLC chromatogram of blackcurrant leaf exudates
- Potentially of value to quantify transport contributions

Effect of potential precursors on fruit AsA content
- Only precursors of the L-Gal pathway enhance fruit AsA content
- No evidence for operation of salvage (L-GalUA) pathway in immature fruit

Biosynthesis of [¹⁴C]AsA from [U-¹⁴C]mannose by blackcurrant fruit
- Decline in AsA biosynthesis throughout fruit maturation
- Correlation between fruit AsA content and AsA biosynthetic capacity in different genotypes

Conclusions
- Fruit AsA must be synthesised each year
- Potential contribution from AsA synthesised in leaves and imported via the phloem
- Low rates of fruit AsA turnover
- Correlation between fruit biosynthetic capacity and AsA contents of individual genotypes

Table 1
Effect of potential precursors on fruit AsA content

Table 2
Turnover of fruit AsA pools

Figure 1
Potential mechanisms affecting AsA concentration of blackcurrant fruit

Figure 2
Changes in tissue AsA throughout growth cycle

Figure 3
HPLC chromatogram of blackcurrant leaf exudates

Figure 4
Schematic of proposed AsA biosynthetic pathways

Figure 5
Biosynthesis of [¹⁴C]AsA from [U-¹⁴C]mannose by blackcurrant fruit

Figure 6
Turnover of fruit AsA pools

Acknowledgements
We wish to thank the following for funding this work: Blackcurrant Growers Association and SEERAD. Work was funded by the BBSRC Horticulture Link programme MEG244/02 with contributions from MIBRIC, MIBRIC, Blackcurrant Growers Association and SEERAD.