

IDENTIFICATION OF TWO NOVEL PTERIN GLUCOSIDES IN TOMATO FRUIT

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Introduction

This study is part of a project to genetically engineer tomato plants to overproduce folate in their fruit [1]. Folates are composed of substituted pterin moieties coupled to p-aminobenzoylglutamate. Initial strategies centered on overexpressing GTP cyclohydrolase I (GTPCHI), the enzyme responsible for the initiation of the pterin biosynthetic pathway. While fruit from these strains exhibited a two-fold increase in folate concentrations, there was a concomitant increase in pterin concentrations of between 20-100 times those of the wild type fruit. Among the pterins observed were two novel pterins (PT1 and PT2). Treatment of the unknown pterins with acid hydrolysis, or with beta-glucosidase (but not alpha-glucosidase) resulted in the liberation of, respectively, neopterin and monapterin, stereoisomers and intermediates of folate synthesis [1].

Aims

To identify the unknown sugar(s) to which neopterin and monapterin are coupled. Due to the chromatographic behavior of the unknown compounds, and the close structural resemblance of their pterin moieties, it is surmised that both compounds contain the same sugar (this will be confirmed once the sugar is identified). Therefore, all further identification was carried out on PT1, the more abundant of the two compounds.

Experimental

PT1 was purified to apparent homogeneity using liquid phase extraction, anion and cation exchange chromatography, and reverse phase HPLC. The purified sample was subjected to NMR analysis using the 750 MHz spectrometer.

Results and Discussion

The NMR characteristics of the compound were consistent with the unknown sugar being beta-D-glucose. Identification will be confirmed by LC-MS. This appears to be the first demonstration of a pterin glucoside in higher plants; such compounds have only been reported previously from prokaryotes [2].

References

- [1] Diaz de la Garza, R., *et al.*, Proc. Natl. Acad. Sci. USA, **101**, 13720-13725 (2004).
- [2] Forrest, H.S. *et al.*, Annu. Rev. Microbiol., **24**, 91-108 (1970).