Transgenical approaches to reducing glycoalkaloids in potato tubers

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Steroidal glycoalkaloids (SGAs) are ubiquitous secondary metabolites of solanaceous plants. In potato tubers, accumulation of SGAs confers bitterness and at high concentrations represents a food safety issue due to their toxic properties. High SGA levels can impede the advancement of breeding programs trying to introduce beneficial phenotypes from wild relatives. Potential safety issues surrounding SGA concentrations have led to the establishment of guidelines for maximum SGA levels expected in commercial potato cultivars. Some potentially valuable cultivars, such as Lenape in the USA, have been withdrawn from the market due to a tendency to accumulate undesirable levels of SGAs (in excess of established limits – 20 mg/g fresh weight). *In planta*, SGAs are believed to play a role in pest resistance and can also contribute to potato flavour. SGAs can accumulate in potato leaves and tubers naturally, and their deposition is increased in response to wounding, light and cold storage.

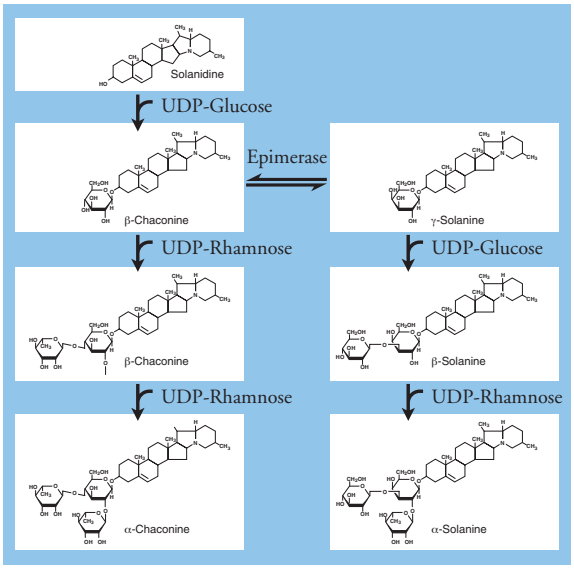

Figure 1 Proposed SGA biosynthetic pathway for the two predominant potato glycoalkaloids derived from the aglycone, solanidine.

Potatoes contain two major SGAs, α-chaconine and α-solanine. Both are triglycosylated steroidal alkaloids derived from the aglycone solanidine, to which either glucose (α-chaconine) or galactose (α-solanine) is added as the primary glycosyl residue. In collaboration with Drs Bill Belknap and Kent McCue at the USDA-ARS (Albany, California) we are studying the impact of down-regulating specific glucosyl transferase genes on tuber glycoalkaloid content and balance using two varieties, viz. Desirée (SCRI) and Lenape (USDA-ARS).

The enzyme solanidine UDP-glucose glucosyltransferase (SGT) catalyses the biosynthesis of γ-chaconine from UDP-glucose and solanidine. While biosynthetic processes leading to mature triglycosylated SGAs are not fully established, the conversion of γ-chaconine to γ-solanine by a specific epimerase would imply that SGT represents the primary pathway for biosynthesis of both major potato SGAs (Fig. 1). At the USDA-ARS, a putative potato gene encoding SGT (SGT1) was expressed in yeast growing on solanidine, and SGT activity of the protein encoded by the gene confirmed *in vitro*. At the SCRI, transgenic tubers of cv. Desiree were produced containing an antisense SGT1 gene driven by the tuber-specific granule-bound starch synthase (GBSS) promoter. This resulted in an almost complete inhibition (*ca.* 90%) of α-solanine accumulation, as quantified by HPLC. However, due to an elevated level of α-chaconine, total glycoalkaloid content was unaffected (Fig. 2), indicating metabolic compensation. The data indicates that whilst SGT1 encodes an enzyme capable of solanidine glucosyltransferase activity *in vitro*, its role *in vivo* is glucosylation of γ-solanine. The tubers are undergoing metabolic profiling using GC-MS and LC-MS to identify the nature of the compensatory mechanism. In parallel (at the USDA-ARS), the same construct was introduced in Lenape cv. with similar results (data not shown). Transgenes containing other putative SGTs are undergoing analysis.

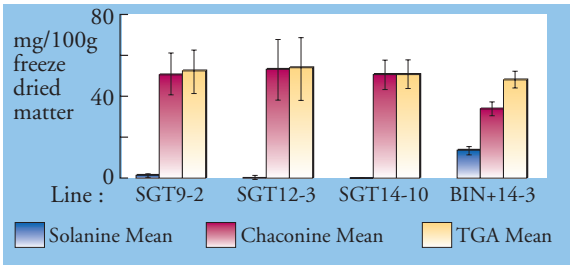

Figure 2 Levels of specific SGAs in tubers of selected second generation SGT1 lines, and an empty vector control (BIN+), as quantified by HPLC.