## Different polyphenolic components of soft fruits inhibit $\alpha$ -amylase and $\alpha$ -glucosidase

GORDON J. MCDOUGALL, FAINA SHPIRO, PATRICIA DOBSON, PAULINE SMITH, ALISON BLAKE AND DEREK STEWART Quality Health and Nutrition, Scottish Crop Research Institute Invergowrie, Dundee DD2 5DA, United Kingdom - contact gmcdou@scri.sari.ac.uk

# Scottish Crop

#### Introduction

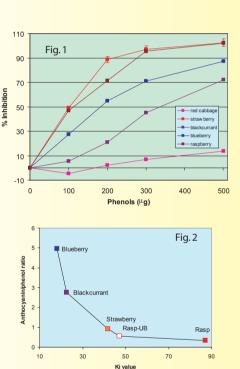
A diet rich in fruits and vegetables is associated with a reduced risk of cancer and cardiovascular disease. This protective effect has been attributed to the activity of antioxidants in preventing oxidative damage to biomolecules caused by free radicals generated during aerobic metabolism (1). Soft fruits are a particularly palatable and rich source of dietary polyphenol antioxidants and berry intake has been correlated with reductions in cardiovascular disease (2). However, it is becoming clear that polyphenols have bioactivities independent of their antioxidant capacities. Polyphenolic fractions from plants can cause insulin-like effects in glucose utilisation and some compounds are as effective inhibitors of intestinal  $\alpha$ -glucosidase activity as drugs used to treat non-insulin dependent diabetes mellitus (NIDDM) (3). This study reports on the bioeffectiveness of polyphenols from soft fruits in inhibiting  $\alpha$ -amylase and  $\alpha$ -glucosidase, crucial enzymes of starch digestion.



### Results

Polyphenol-rich extracts were effective inhibitors of α-amylase (Fig. 1). Strawberry and raspberry extracts were almost as effective as green tea, a known inhibitor (4). The extracts also inhibited rat intestinal  $\alpha$ -glucosidase but the order of effectiveness was altered. The degree of inhibition was related to the anthocyanin content of the extracts (Fig. 2). This finding agrees with (3). previous reports Conversely those extracts most effective against amylase (green tea strawberry, raspberry, red grape and red wine) contained appreciable amounts of tannin-like substances.

Inhibition of  $\alpha$ -amylase and  $\alpha$ -glucosidase



Removing tannins by "fining" with gelatin removed the amylase inhibitors but they were recovered from the gelatin by washing in methanol. This strongly suggests that the amylase inhibitors were tannin-like polyphenols. To test this further, raspberry extracts were subfractionated by chromatography on Sephadex LH-20 using a procedure known to enrich tannins.

The tannin-rich LH bound fraction contained the majority of the amylase inhibitory activity whereas the glucosidase inhibitors were retained in the anthocyanin-enriched unbound fraction (Fig. 3).

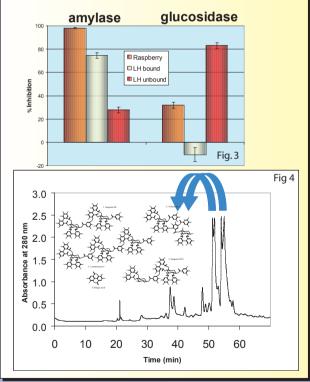
#### Conclusions

Different components of polyphenol-rich extracts from soft fruits inhibit  $\alpha$ -glucosidase and  $\alpha$ -amylase. Green teas are known to inhibit salivary  $\alpha$ -amylase and reduce the oral concentration of fermentable sugars after eating starchy foods (4). The inhibition of  $\alpha$ -amylase by strawberry (and other soft fruit) extracts may provide a more palatable means of protecting against dental caries, especially in young children. In addition, similar inhibition of pancreatic  $\alpha$ -amylase by soft fruit extracts may provide a means of controlling post-meal blood glucose levels in patients with non-insulin dependent diabetes mellitus (NIDDM) without recourse to artificial  $\alpha$ -amylase inhibitors (3). The nature of the inhibition is under study.

SCRI recieves gift-in-aid from the Scottish Executive Environment and Rural Affairs Department.

#### Identification of $\alpha$ -amylase inhibitors

The LH-20 bound fraction was separated by reverse phase chromatography on a preparative C18 column (Fig. 4). All of the amylase inhibitory activity eluted in two fractions near the end of the gradient (Fig. 4, arrows). The amylase inhibitors were analysed by LC-MS and were found to consist of a mixture of ellagitannin compounds already reported in raspberry (5). The main constituents were Sanguiin H6, Sanguiin H10 and Lambertianin C (see Fig. 4, insert) with smaller amounts of ellagic acid and Nobotannin A.



1. Hertog, et al., Potentially anticarcinogenic secondary metabolites from fruit and vegetables. In Phytochemistry of Fruit and Vegetables; Tomas-Barberan, F. A., Robins, R. J., Eds.; Clarendon Press: Oxford, U.K., 1997; pp 13-329. 2.. Knekt, P. et al., Flavonoid intake and coronary mortality in Finland: a cohort study. Brit. Med. J. 1996, 312, 478-481. 3. Matsui, T t al., α-glucosidase inhibitory action of natural acylated anthocyanins. 2.  $\alpha$ -glucosidase inhibition by isolated acylated anthocyanins. J. Agric. Food Chem. 2001, 49, 1952-195. 4. Zhang J. and Kashket. S (1998) Inhibition of salivary amylase by black and green teas. Caries Research 32, 233-238. 5. Mullen W et al., Ellagitannins, flavanoids, and other phenolics in red raspberries and their contribution to antioxidant capacity and vasorelaxation properties. J. Agric. Food Chem. 2002; 50: 5191-5196