Development of Low-Tannin Sorghums Resistant to Birds and to Microbial Seed Deterioration

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Background

High tannin sorghums are relatively resistant to grain-eating birds and to grain molds, but are nutritionally inferior to low-tannin sorghums. We have evidence that within the diverse group of tannins and chemically related flavonoids which are characteristic of high tannin sorghums, different components are responsible for the harmful (antinutritional) effects and from the beneficial effects of bird and mold resistance. Thus it should be possible to improve the nutritional value of high tannin sorghums by elimination of antinutritional factors without loss of resistance to birds and molds.

Objectives

a). The identification and characterization of the phenols of sorghum which are responsible for resistance to bird depredation and seed deterioration. The antinutritional activity is apparently associated with condensed tannins with an average of 5 to 6 flavan-3-ol units per molecule.

b). Develop rapid, single, and specific chemical assays for the agronomically beneficial phenolic components. We will especially concentrate on developing technologically simple assays suitable for use in developing nations.

c). Identification or development of sorghum cultivars with high levels of beneficial phenols and low levels of antinutritional condensed tannins.

d). Elucidation of the site and mode of synthesis of tannins and related phenols, their regulation, and the influence of environmental factors such as light, wounding, and infection by pathogens on phenol synthesis.
Results

We had suggested that flavan-4-ols might play a role in bird resistance of sorghums, particularly in those which do not contain tannin (Subramanian et al., 1983). Before this phase of the project (Objective a) got underway, our collaborators in India showed rather conclusively that flavan-4-ols do not confer bird resistance. The non-tannin components responsible for bird resistances have not yet been identified. Our field trials of sorghum cultivars for bird resistance continue, with emphasis on analysis of polyphenol composition of bird-resistant and bird susceptible cultivars throughout seed development (Objective c).

Recently strong bird resistance has been reported in two sources of sorghums which do not contain tannin. Characterization of these sorghums with respect to components responsible for bird resistance is the focus of a preproposal recently submitted to PSTC/USAID.

The focus in the present project shifted toward seed deterioration (also in Objective a). With our Indian collaborators, we were able to show a clear correlation of mold-resistance with levels of flavan-4-ols (Jambunathan et al., 1986) in sorghum grain, leaves, and callus tissue. As a result of this work flavan-4-ols have been recognized as the most useful parameter available for predicting mold resistance of sorghum. In brown sorghums, tannins also contribute, and in white sorghums seed hardness seems also to contribute to resistance. But flavan-4-ols remain the best-characterized factor in sorghum grain mold resistance. Our flavan-4-ol assay (Watterson and Butler, 1983) is simple and seems to be likely to widely used. In subsequent work after this research project was completed, with our sorghum breeder collaborator G. Ejeta we have obtained evidence that mold resistance may not be due to the flavan-4-ols directly, but to other flavonoïd components metabolically close to the flavan-4-
ols. This work is still being pursued.

With respect to assay development (Objective b), the major development was the Blue BSA assay (Asquith and Butler, 1985) for protein precipitation by tannin. This is becoming a well accepted assay for tannins. In a related matter, we used a similar assay to compare the relative affinities of several tannins for several proteins and found that the concept of specificity applies to both the protein and the tannin components of tannin-protein interactions (Asquith and Butler, 1985). These observations suggested that differences in affinity due to specificity are functionally significant.

Assay of the activities of digestive enzymes under conditions simulating those of the digestive tract showed that dietary tannins are unlikely to significantly inhibit digestion in vivo (Blytt et al., 1998). This supports our contention that the antinutritional effects of tannin are not due to inhibition of digestion, as commonly assumed, but are due to inhibition of metabolism of digested and absorbed foodstuffs. We have recent (unpublished) data which clearly shows inhibition of post-digestive metabolism is the major mode of the antinutritional effects of tannins.

We have been using in vitro tissue culture of sorghum in an attempt to develop cultivars with high levels of beneficial phenols and low levels of antinutritional condensed tannins (Objective c). In vitro plant tissue culture is slow, laborious work. At the time this PSTC grant ended, we were just learning how to regenerate cultured callus tissue into intact plants (Cai et al., 1987). We now have regenerated about 6,500 plants and are doing selections on them to identify variants with the desired properties (unpublished).

With respect to tannin synthesis (Objective d), Lesley Putman's thesis work (not yet published) involved identification of the enzyme and substrate for the final step in tannin biosynthesis in developing sorghum seeds. Lesley identified
the enzyme as polyphenol oxidase (Putman, 1987). The substrate which she identified was a C-glucosylflavone; this gave the maximum rate of reaction with polyphenoloxidase from the seeds, but the product did not have properties corresponding to tannin (Putman, 1987). On the basis of this work and that of Haslam (), we propose the following scenario for tannin biosynthesis. The chemical defense of the sorghum seed is established before deposit of protein and starch in the endosperm. Synthesis of oligomeric dimer and trimer precursors of tannin is essentially complete within 2 weeks of pollination. During grain development, damage to the seed by wounding or possibly infection results in rapid polymerization of the precursors to tannin by polyphenol oxidase as a defense measure. If no damage occurs, polymerization to tannin occurs later, during desiccation of fully developed seed, producing the tannin characteristic of dry mature grain.
References


