NOTE

# *Amaranthus hypochondriacus*: Starch Isolation and Partial Characterization[1]

G. A. YANEZ, J. K. MESSINGER, C. E. WALKER, and J. H. RUPNOW[2]

Amaranth is a nongrass, broad-leaved plant that produces small seeds on a sorghum-like head. The main species are *Amaranthus cruentus*, *A. caudatus*, and *A. hypochondriacus* (NRC 1984). It constitutes an important part of the diet in areas of Latin America, Africa, and Asia (Teutonico and Knorr 1985). The grain has relatively high protein, lysine, fat, and mineral contents. The leaves are also edible, being a good source of carotene, iron, calcium, ascorbic acid, and protein. The nutrients and caloric content of amaranth grain provide a theoretical nutritional value comparable or slightly superior to the common cereal grains (NRC 1984).

Starch is the most abundant carbohydrate component of amaranth (Saunders and Becker 1984), and it is mainly stored in the perisperm (Okuno and Sakaguchi 1981). Amaranth starch content is reported to range from 48 to 69% depending on the species (Saunders and Becker 1984). The starch granule varies from 1 to 3 $\mu$m in diameter and is spherical or polygonal in shape (Tomita et al 1981, Saunders and Becker 1984). Okuno and Sakaguchi (1981) reported both glutinous and nonglutinous starches in the seeds of *A. hypochondriacus*. The occurrence of both forms of starch has also been reported in such cereals as corn, rice, barley, and sorghum.

Starch properties of *A. hypochondriacus* and its potential uses in bread were studied by Lorenz (1981), who reported that bread made from blends of wheat and over 10% amaranth flours has reduced volume and acceptability. Further research is needed on amaranth to determine its functional properties and potential food uses. The purpose of this study was to isolate the starch from amaranth and to measure its pasting properties, resistance to freezing and thawing, and digestibility.

[1]Published as paper no. 7851, Journal Series, Nebraska Agricultural Experiment Station.

[2]Department of Food Science and Technology, University of Nebraska, Lincoln 68583-0919.



## MATERIALS AND METHODS

### Sample Identification

Starch was isolated from the seeds of gold *A. hypochondriacus* R103 obtained from Post Rock Natural Grains, Rte. 1 Box 24A Lauray, KS 67649. Argo corn starch (CPC International Inc., Englewood Cliffs, NJ 07632), potato starch (Sigma Chemical Co., St. Louis, MO 63178), and whole hard red winter wheat flour (Gooch Mills, Lincoln, NE) were included, where appropiate, for comparison.

### Chemical Analysis

The grain was milled in a cyclone sample mill (Udy Corp., Boulder, CO 80301), with a 1-mm screen. The proximate composition of the whole grain was determined according to standard methods of the AACC (44-19, 46-12; AACC 1983) and AOAC (7.010, 7.045, and 7.054; AOAC 1980).

The same methods were used to determine the proximate composition of the isolated starch.

### Starch Isolation

Starch isolation was performed according to the alkaline method described by Yanez and Walker (1986). The total steeping time was 24 hr instead of 48 hr.

Starch content of the whole grain was determined according to the Yellow Springs Instrument (YSI) glucose analyzer procedure (Blake 1981) and using an enzymatic external hydrolysis. An amaranth flour suspension (2 g/75 ml water) was boiled for 30 min with occasional stirring and cooled to 60° C. After adding 10 ml of 1 *M* sodium citrate buffer (pH 4.8), the sample was incubated with 1 ml of glucoamylase (Diazyme D.L. 200, Miles Lab. Inc., Elkhart, IN) for 2 hr at 60° C. The suspension was brought to a final volume of 250 ml with water. A dilution of 1:10 was made, and glucose released was measured on a YSI industrial immobilized enzyme analyzer (model 27, Yellow Springs Instrument Co., Yellow Springs, OH 45387). Potato starch (Sigma Chemical Co.), at the same concentration, was used as a reference.

Amylose content was determined according to Juliano et al (1981), using known amylose/amylopectin (Sigma Chemical Co., lots 121F-3778 and 102F3922, respectively) solutions to plot the standard curve.

**Starch Functional Characteristics**

A Brabender Visco/amylo/Graph was used to analyze amaranth starch pasting properties (C.W. Brabender Instruments, South Hackensack, NJ 07606). The procedure given in the *Amylograph Handbook* (Shuey and Tipples 1980) was followed for the preparation of the starch samples, and AACC method 22-10 was used to obtain an amylogram of amaranth whole flour (AACC 1983).

Stability of amaranth starch paste to freezing and thawing was tested according to the method of Schoch (1968), using distilled water to prepare a 5% (w/v) starch paste, and subjecting it to 10 freeze/thaw cycles. A change in the method to improve separation was centrifugation for 20 min at 940 × *g* instead of 900 × *g*.

**Starch Digestibility**

In vitro digestibility was determined according to a modification of the method of Bernfeld (1955) as described by Frels and Rupnow (1984), using porcine pancreatic alpha-amylase (Sigma Chemical Co.). Gelatinized starch was not reduced with sodium borohydride and no inhibitor was added.

**Endogenous Amylase Activity**

Flour suspensions (10% w/v in distilled water) were prepared from amaranth flour, wheat flour, and amaranth/wheat blends (10:90, 20:80) and incubated at 25° C. Aliquots were taken at various intervals up to 8 hr, centrifuged at 8,000 × *g* for 2 min, then analyzed for glucose on the YSI analyzer.

## RESULTS AND DISCUSSION

**Chemical Analysis**

The chemical composition of the corn starch, whole amaranth flour, and isolated starch are given in Table I. The protein content of the flour was in the range given by several authors (Wolf et al 1950, Becker et el 1981, Lorenz 1981, NRC 1984).

Iodine binding indicated that grain of this amaranth species contained a glutinous waxy starch with low amylose content. The percentage of starch in the whole grain was about the same as reported previously by Wolf et al (1950), Becker et al (1981), and Saunders and Becker (1984).

**Functional Properties**

The size of the starch granules was similar to that reported by others (Tomita et al 1981, Saunders and Becker 1984), with a predominant size of 1 $\mu$m and polygonal shape. The granule size as well as the amylose content appeared to have influenced the performance of the amaranth starch under the tests studied. Amaranth starch had a peak amylograph viscosity of 400 compared to 420 Brabender units (BU) for corn starch, with pasting and peak temperatures of 67 and 74° C as compared to 76 and 93° C for the corn starch (Fig. 1). Corn starch had a high setback viscosity caused by aggregated structures developed from the high amylose content (Becker et al 1981). Conversely, the low setback observed in amaranth starch is probably caused by its low amylose content. Amaranth starch viscosity did not drop when held at 95° C. The starch was apparently resistant to mechanical breakdown. An amylogram of amaranth flour gave the same pasting and peak viscosity temperatures as the starch.

Lorenz (1981) and Becker et al (1981) reported a peak viscosity of only 320 BU for *A. hypochondriacus*, with a starch amylose content of 7.2%. Modi and Kulkarni (1976) reported pasting and peak temperatures of 70 and 78° C respectively, and 900 BU peak viscosity for *A. panniculatus*, compared to corn starch with a 79° C pasting temperature and 90° C peak temperature, and 520 BU peak

**TABLE I**
**Chemical Analyses of Amaranth Flour and Amaranth and Corn Starches**[a]

| Component (%) | Amaranth Flour | Amaranth Starch | Corn Starch |
|---|---|---|---|
| Moisture | 9.67 | 6.16 | 8.72 |
| Nitrogen | 2.69 | 0.02 | 0.07 |
| Ash | 2.80 | 1.39 | 0.05 |
| Fat | 7.15 | 0.39 | 0.12 |
| Fiber | 3.34 | ··· | ··· |
| Amylose | ··· | 4.90 | 26.30 |
| Starch | 62.30 | ··· | ··· |

[a]Values are an average of three determinations calculated based on dry matter.

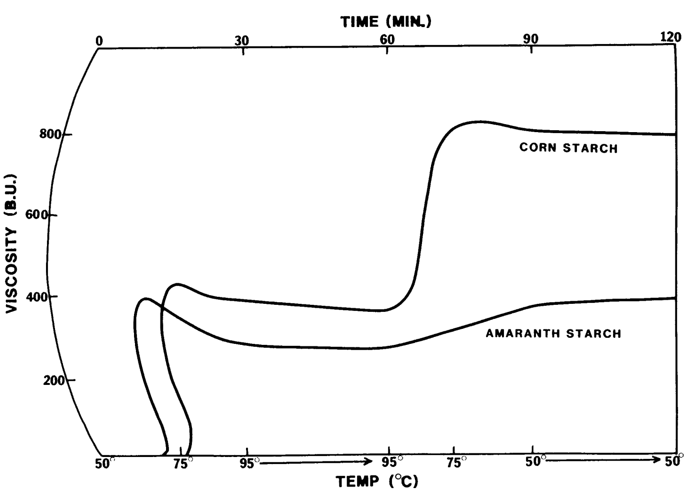


**Fig. 1.** Comparison of amylograms of amaranth and corn starch.

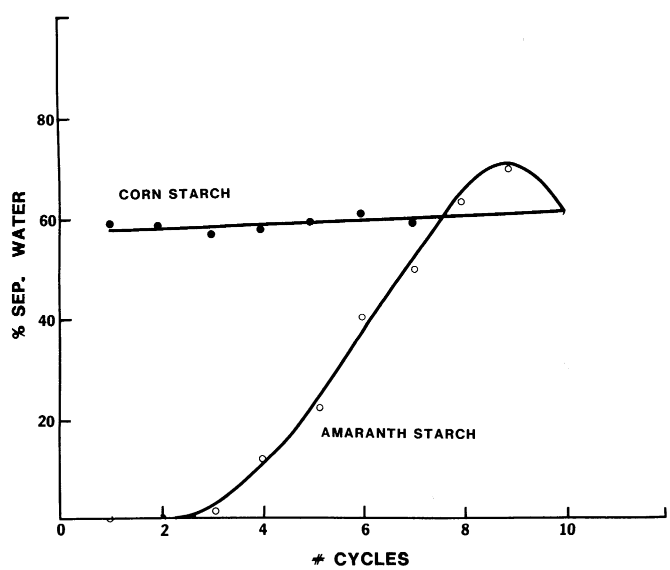


**Fig. 2** Stability of amaranth and corn starch through freezing and thawing cycles (average of two determinations).

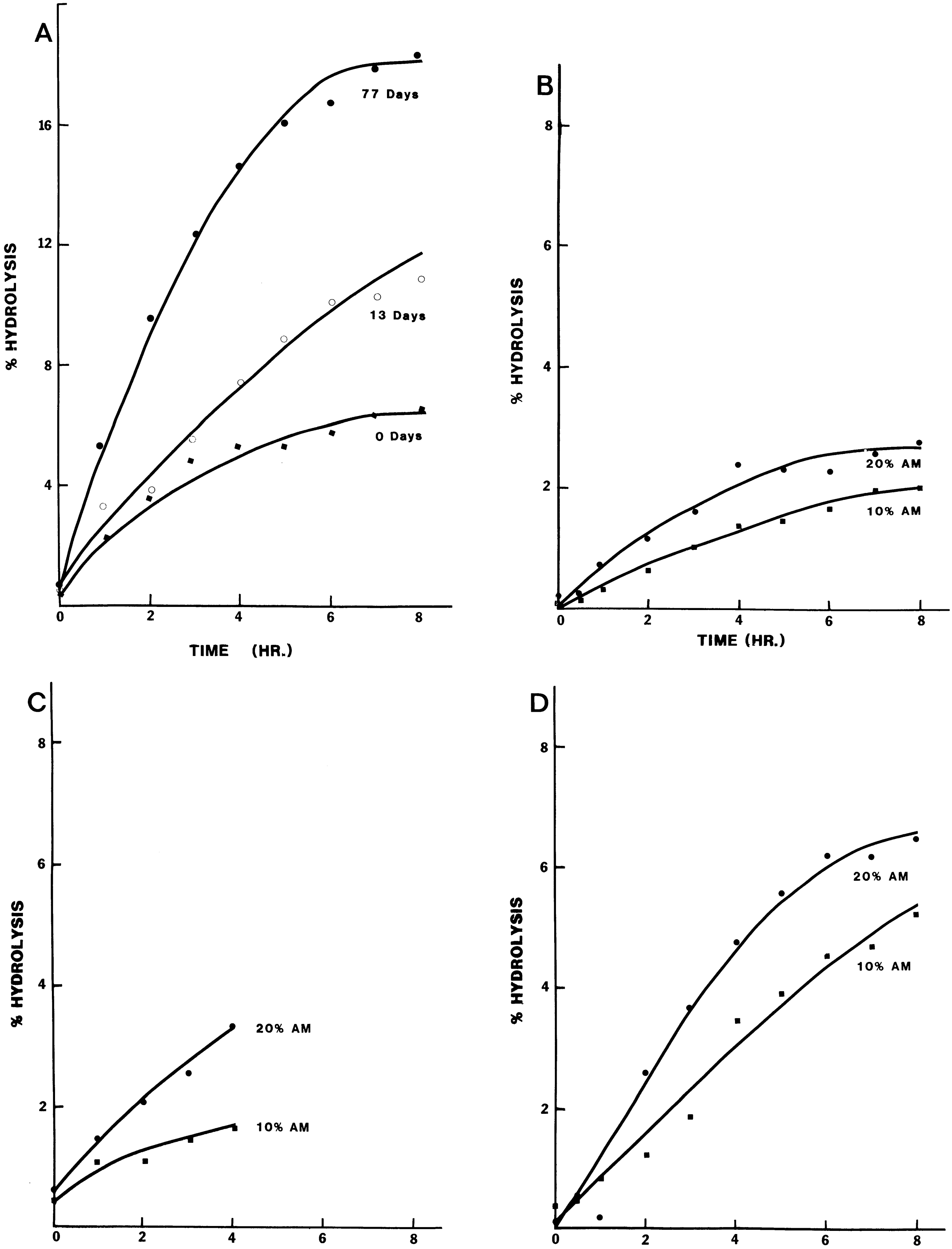


**Fig. 3.** Endogenous amylase activity of amaranth flour and amaranth/wheat blends. (All curves fit a second order regression with $r > 0.93$; **AM** = % amaranth flour). **A,** Response of 100% amaranth flour after different storage times; **B,** Response of blends at 0 days of storage; **C,** after 13 days of storage; **D,** after 77 days of storage.

viscosity. Both temperatures were close to the results obtained in this study with only the peak viscosities being different; the discrepancies could be due to the differences in starch concentration, pH, or amylose content of the samples.

The results of freezing and thawing tests (Fig. 2) indicated that amaranth starch possessed good stability by withstanding four cycles before marked syneresis occurred. This slow progressive deterioration is also characteristic of waxy sorghum starch (Schoch 1968). Corn starch showed the rapid deterioration typical of starch containing higher concentrations of amylose.

In vitro digestibility tests of amaranth starch resulted in 63.5% hydrolysis after 60 min whereas corn starch had 35.0%. Most digestion occurred in the first 10 min of incubation, and then both curves flattened out. Tomita et al (1981) reported that amylases tend to digest waxy starch faster than normal starch. Furthermore, Sugimoto et al (1981) reported that normal and waxy starch were digested 2.4 and five times faster than normal maize starch by pancreatin and crude glucoamylase. The present study supported their results.

Amaranth flour showed endogenous amylase activity (Fig. 3). After 8 hr there was 6.5% starch hydrolysis, whereas wheat flour showed consistantly less than 1% hydrolysis. This finding could be important because, in general, cereal grains do not show major activity until germination. Blends of amaranth and wheat flour were also tested, where it was observed that increasing the amount of amaranth in the blend increased starch hydrolysis. The degree of starch hydrolysis by the endogenous amylase also was related to the age of the flour. Amaranth flour stored for 13 days at 25° C showed 10.6% hydrolysis after 8 hr, and flour stored 77 days was 18.4% hydrolyzed. Apparently, starch in the amaranth flour that was stored for 77 days was more susceptible to the action of the native amylase. Comparable results were also obtained with the blends.

## CONCLUSION

Because of its interesting properties, there are several potential applications for amaranth starch. It could be useful in instant soup because of its low pasting temperature and peak viscosity. Because it does not form a solid gel and is stable after freezing and thawing, it could be beneficial in gravies or sauces. In canned goods such as cream-style corn it could form a protective colloid to reduce or prevent protein denaturation. It would also be applicable in vegetable soups, where a starch with low gelatinization temperature is needed to maintain a complete dispersion of the product during filling.

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[Received August 19, 1985. Accepted January 16, 1986.]