

## Plasma glucose and insulin responses to traditional Pima Indian meals<sup>1,2</sup>

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**ABSTRACT** The in vivo glycemic and insulin responses and in vitro starch digestibility were determined for six staple foods (corn, lima beans, white and yellow teparies, mesquite, and acorns) traditionally consumed by Pima Indians. Equivalent carbohydrate portions (25 g) of the foods were fed to eight healthy Caucasian volunteers. The calculated glycemic indices (GIs) ( $\bar{x} \pm \text{SEM}$  with glucose as the standard) were all low, ranging from  $16 \pm 2$  for acorns to  $40 \pm 5$  for corn. Insulin responses and in vitro starch digestibilities correlated with the GI. These results provide further support for the hypothesis that the slow digestion and absorption of starch in traditional foods was a factor that helped protect susceptible populations from developing diabetes. *Am J Clin Nutr* 1990;51:416–20.

**KEY WORDS** Glycemic index, Pima Indian, traditional foods, carbohydrate

### Introduction

During this century the abandonment of traditional subsistence activities (gathering, hunting, and/or farming) among several indigenous populations has been associated with a dramatic increase in the prevalence of non-insulin-dependent diabetes mellitus (NIDDM) (1). The River Pima (Akimel O'odham) tribe of arid, central Arizona currently has the highest documented prevalence of diabetes of any ethnic group in the world, with up to 35% of their population developing the disease and associated complications by adulthood (2). Their diets today are considerably different from those before 1930, which were dominated by wild and cultivated desert legumes (3), with cacti, fish, and small seeds as supplements (4). Mesquite, corn, and legumes together provided ~40–50% of food intake by weight. Modernization and the donation of government-surplus commodity foods led to the daily consumption of Western mainstays such as wheat flour, lard, sugar, coffee, and processed cereal products (5). Thorburn et al (6) showed that the traditional Australian Aboriginal and Pacific island foods produce significantly lower glycemic and insulin responses than do Western starchy staples. It was hypothesized that the slow digestibility of native foods protected genetically-susceptible populations such as Australian Aborigines from developing diabetes.

The purpose of this study was to gather further evidence to test the hypothesis by determining the nature of the major carbohydrate sources in the traditional diet of the River Pima Indi-

ans. Six traditional Pima Indian meals were assessed by analyzing in vivo plasma glucose and insulin responses and the in vitro digestibility of the starch.

### Subjects and methods

Six traditional River Pima staple foods were investigated: lima beans (*Phaseolus lunatus* L), white tepary beans (*Phaseolus acutifolius* Gray), yellow tepary beans (*Phaseolus acutifolius* Gray), mesquite pods (*Prosopis velutina* Wootton), corn (*Zea mays* L), and acorns (*Quercus emoryi* Torrey).

The foods were either grown by Pima Indians on the Gila River Indian Reservation by using traditional methods of cultivation or they (acorns and mesquite) were wild harvested nearby. The four cultivated staples were traditional landraces (folk varieties), having remained unchanged for  $\geq 500$ –2000 y. The flour corn was left on the plant until partially dry according to traditional practice. Four of the cultivated foods (lima beans, corn, and white and yellow teparies) were stored at room temperature for 12 mo after harvest. The wild acorns and mesquite were freshly harvested in August 1986 when all foods were air freighted to Sydney. On arrival the foods were stored at room temperature for 3 mo until analyzed. Meals were prepared according to methods that simulated those traditionally used by the Pima Indians.

#### Mesquite: mesquite cakes

Mesquite pods were cleared of dust and debris by washing and drying. They were then heated in a 130 °C oven for 4 h to destroy bruchid beetle larvae. Pods were ground with mortar and pestle to make a fine flour and sifted twice to remove seeds and seed coats. The flour was mixed with a small amount of water and formed into a cake, which was left to dry at room temperature for 2 d. Before eating, the cakes were mixed with water and heated to ~65 °C to enhance palatability (7, 8).

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*Corn hominy*

Corn kernels were removed from the dried cobs by hand. Approximately 30 cc of powdered lime [Ca(OH)<sub>2</sub>] were dissolved in 1.8 L of water and the kernels added. Corn was cooked at 100 °C until the hulls loosened from the kernels (~2 h). The liquid was then drained and the corn washed in running water until all the lime taste was removed, ~12 h (9).

*Teparies and lima beans*

Beans (500 g) were cleaned and soaked overnight. Two cloves of garlic, one chopped onion, and just enough water to cover was added and the mixture boiled until the beans softened (several hours). Some of the beans were then mashed and stirred into the liquid to form a thick broth. (9)

*Acorns: acorn and venison stew*

Acorn shells were cracked open by pestle blows and placed in a bucket of water to separate the seed from the husk. This leaching also served to remove some of the bitter taste of the seed. The seeds were laid out to dry, then ground with a mortar and pestle to make a coarse meal. Venison (200 g) was gently simmered for 5 min in just enough water to cover the meat. Acorn meal (150 g) was then added and the mixture heated until any bitter taste was removed (9).

After cooking, the meals were stored at -80 °C before analysis of nutrient composition, *in vitro* starch digestibility, and *in vivo* metabolic responses. Available carbohydrate was measured directly based on the method of Southgate (10) and Wills et al (11). Moisture (freeze-drying), fat (12), protein (13), and dietary fiber (14) were also estimated.

*In vitro starch digestibility*

A modification of the method of Jenkins et al (15) was used. Food portions (1 g homogenized) were individually mixed with 3 mL porcine pancreatin (1% wt:vol, grade VI, Sigma, St Louis, MO) and 2 mL of pooled human saliva. Distilled water was added to bring the final food mixture volume up to 15 mL. Plastic tubes containing the mixture were incubated at 37 °C on a shaker for 4 h, heated at 80 °C for 5 min to inactivate  $\alpha$ -amylase in the enzyme solutions, and finally centrifuged at 10 000  $\times$  g for 5 min. Maltose and oligosaccharides in the supernatant were analyzed together as glucose after hydrolysis of a 2-mL sample with 2 mL of 10 mol hydrochloric acid/L for 1 h at 75 °C. This was followed by neutralization with sodium hydroxide. Glucose was analyzed by the glucose hexokinase method on a centrifugal analyzer (Cobas Fara, Roche Diagnostics, Basle, Switzerland). White bread was used as a control. Further *in vitro* experiments were undertaken with saliva and pancreatin inactivated by boiling to account for free sugars already present in the foods and digestive juices. The sugars enzymatically released from the foods gave an index of starch digestibility.

*In vivo studies on the metabolic responses to Pima meals*

Eight healthy Australian Caucasian volunteers (five males, three females) aged 21–24 y with normal glucose tolerance and a body mass index of 22.3  $\pm$  0.8 kg/m<sup>2</sup> ( $\bar{x}$   $\pm$  SEM) were studied. Equal carbohydrate portions (25 g) of each meal (except mesquite) or 25 g glucose were fed on separate mornings after a 12-

h overnight fast. Mesquite cakes (15-g carbohydrate portion) were fed to only four subjects because of quantity restriction and poor palatability; the area under the response curve was adjusted proportionally. Meals were consumed within 15 min with the addition of white tea to increase palatability. Tea consisted of 60 mL of milk and ~300 mL of tea to make up meal volume to 600 mL. Meals were eaten 1 wk apart in random order.

Finger-prick capillary blood samples were taken at fasting (0 min), then at 15, 30, 45, 60, 90, and 120 min after the meal was commenced. Hands were placed in a 45 °C water bath for 3 min before puncturing with an automatic finger-prick pen (Penlet, Lifescan Inc, Palo Alto, CA). Blood samples (500–800  $\mu$ L) were obtained by gentle pressure on the finger tip and collected in EDTA-coated Eppendorf tubes. Plasma was removed after centrifugation (12 500  $\times$  g for 1 min) and stored at -80 °C before analysis.

Plasma glucose was analyzed in duplicate by the hexokinase method. Plasma insulin was analyzed by radioimmunoassay with the BIO-RIA <sup>125</sup>I-Insulin Chromacode Radioimmunoassay kit (Bio-Mega Diagnostic Inc, Hamon, Montreal, Canada). The glycemic index (GI) and insulin index of each meal was calculated as described previously (16) by using 25 g oral glucose as the standard.

Statistical differences were assessed by analysis of variance with Fisher's test and Tukey's HSD method used for multiple comparisons (17). The protocol was approved by the Medical Ethical Review Committee of the University of Sydney.

**Results**

The composition and weight of the meals is shown in **Table 1**.

*In vitro starch digestibility*

The percentage of starch digested *in vitro* ranged from 8  $\pm$  0.2% for acorns to 36  $\pm$  1.2% for traditional Pima corn (**Table 2**). The acorn and mesquite meals were digested more slowly ( $p < 0.01$ , Fisher's test) than the other meals. The starch in the bread control was 55% digested after 4 h, which was significantly higher than that for the other meals ( $p < 0.01$ , Fisher's test).

*The glycemic and insulin indices of the meals*

All the meals gave low GIs ranging from 16  $\pm$  2 for acorns to 40  $\pm$  5 for corn (**Table 2**). The value for corn (40  $\pm$  5) was significantly higher ( $p < 0.05$ , Tukey's HSD method) than that for the other meals. All meals gave significantly smaller GIs than did the oral glucose load ( $p < 0.01$ , Tukey HSD method). The insulin responses to the meals varied proportionately with the glycemic responses and correlated with the GIs ( $r = 0.9$ ,  $p < 0.01$ ). (**Table 2**).

There was a significant positive correlation between the *in vitro* digestibility of the meals and their glycemic index ( $r = 0.9$ ,  $p < 0.01$ , **Fig 1**). Fat and fiber content of the meals showed significant negative correlations with glycemic index ( $r = 0.8$ ,  $p < 0.01$  and  $r = 0.9$ ,  $p < 0.001$ , respectively, **Fig 2**).

TABLE 1  
Nutrient composition and weight of meals

Meals based on	Moisture	Protein	Fat	Available carbohydrate	Fiber	Weight of meal
	<i>g/100 g</i>	<i>g/100 g</i>	<i>g/100 g</i>	<i>g/100 g</i>	<i>g/100 g</i>	<i>g</i>
Corn	65.1	4.3	2.3	20.0	4.2	125
Lima beans	67.8	7.5	2.6	12.8	6.1	195
White teparies	67.4	7.4	0.8	12.7	6.3	197
Yellow teparies	75.2	5.4	0.6	10.5	4.8	239
Mesquite	66.4	3.1	5.0	6.2	9.4	241*
Acorns	47.5	6.5	17.9	5.5	18.4	454

\* 15-g carbohydrate portion.

## Discussion

The low glycemic and insulin responses to Pima Indian foods, which we found in this study to be similar to those seen with traditional Australian Aboriginal and Pacific Island foods, contrast with the rapid and high responses to Western staples such as potatoes, bread, and other processed cereal products (18). Although legumes were previously shown to be a source of slowly digested carbohydrate (19), this is the first study to confirm that the particular legumes, which dominated the Pima Indian diet (lima beans, tepary beans, and mesquite pods), are also slow-release. These results provide further support for the hypothesis that the nature of the starch in traditional foods was an important factor in protecting indigenous populations from developing diabetes. We believe it is valid to extrapolate these findings in Caucasian subjects to Pima Indians on the basis of our previous work in Australian Aborigines, who are also at high risk of developing diabetes. We showed that responses to traditional Aboriginal foods with low GIs were similar in Caucasians and healthy Aborigines (20).

The GI of the traditional flour corn ( $40 \pm 5$ ) was significantly lower than the published value for commercial sweetcorn,  $59 \pm 11$  (18), suggesting that modern sweetcorn, which has replaced native corn in the Pima diet, contains a different type of starch. The amylose-amylopectin ratio in starch is an important determinant of rate of digestion (19) and may account for differences between the corns. The difference may also be due

to developmental stage at consumption. The Pima flour corn is harvested when fully mature whereas sweet corn is consumed when immature.

The glycemic indices of foods based on 25-g carbohydrate portions are comparable with values based on 50 g carbohydrate. Jenkins et al (21) reported a linear dose response to full (50 g carbohydrate) and half (25 g carbohydrate) bread portions. The half portion gave a plasma glucose area that was almost half of the 50-g carbohydrate value. Jenkins et al (18) tested a number of foods in 25-g portions including beetroot, carrots, parsnips, and swede, which had GI values of 64, 92, 97, and 72, respectively, with 25 g glucose as the standard.

The in vitro system used to determine relative starch digestibility showed that all six meals were digested significantly more slowly than were the Western starchy staple, white bread. Acorns, which were particularly high in fiber, had the lowest digestibility. The difference between the two tepary varieties suggests that differences in the rate of starch digestion are responsible for their different glycemic responses. The rates of starch digestion in the Pima Indian staples are similar to those values we reported (6) for traditional staples of the Aboriginal and Pacific Island communities.

The GIs of the meals varied inversely with dietary fiber con-

TABLE 2  
Glycemic index (GI) and insulin index (II) and in vitro starch digestibility of the meals

Meal based on	GI	II	Starch digested in vitro
			%
Corn (hominy)	$40 \pm 5$	$53 \pm 4$	$36 \pm 1.2$
Lima beans (broth)	$36 \pm 3$	$51 \pm 3$	$38 \pm 0.5$
White teparies (broth)	$31 \pm 3$	$38 \pm 5$	$33 \pm 0.4$
Yellow teparies (broth)	$29 \pm 3$	$38 \pm 5$	$26 \pm 0.2$
Mesquite (cakes)	$25 \pm 3$	$18 \pm 3$	$9 \pm 0.2$
Acorns (stewed with venison)	$16 \pm 1$	$19 \pm 2$	$8 \pm 0.2$

\*  $\bar{x} \pm \text{SEM}$ .

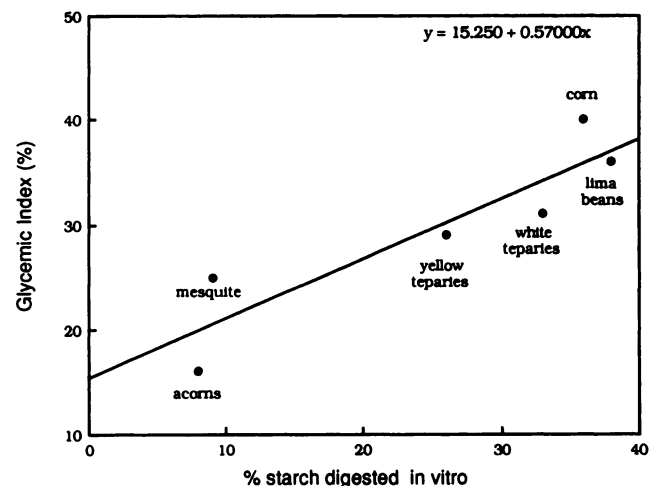


FIG 1. Relationship between glycemic index and starch digestibility in vitro ( $r = 0.9$ ,  $p < 0.01$ ).

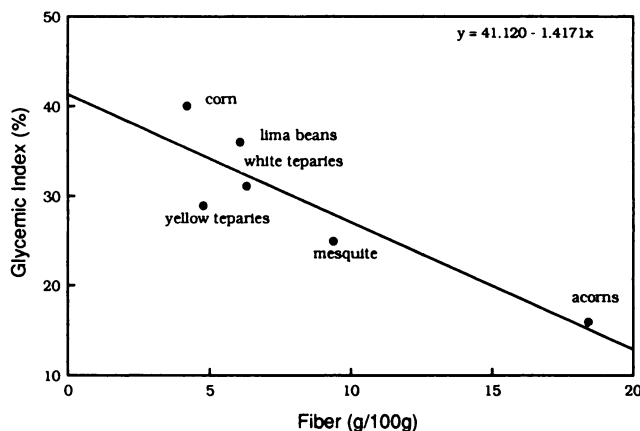



FIG 2. Relationship between glycemic index and fiber content of meals ( $r = 0.9$ ,  $p < 0.001$ ).

tent despite the fact that this method underestimates soluble fiber. Certain types of fiber, particularly the soluble viscous fibers, slow absorption in the small intestine, delay gastric emptying, and decrease upper gastrointestinal motility (22). Insoluble fiber such as that found in corn appears to have little effect on glycemic response.

Traditional methods used in the preparation of the meals may also have contributed to their low GI. Plasma glucose and insulin responses depend on particle size and cooking time as well as the type of cooking process (23). Previous studies showed lower responses to home-cooked food compared with their processed equivalents (24). Acorns were ground coarsely in contrast to mesquite, which required grinding into a fine flour. The smaller particle size may have contributed to the higher responses to mesquite. A greater degree of starch gelatinization may take place, which allows easier access of  $\alpha$ -amylase to the starch, increasing the rate of digestion and the subsequent glycemic response (16).

The slow-release carbohydrate hypothesis (6) may be particularly pertinent to ethnic populations with centuries of residence in desert habitats, such as the Pima Indians of the Sonoran Desert and Australian Aborigines. Their gastrointestinal and endocrine systems may have been selected in an evolutionary sense to metabolize the many slow-release complex carbohydrate foodstuffs available in arid zones. North American desert floras are relatively rich in edible legumes as well as in cacti and small seeds with hygroscopic mucilages, many of which contributed to the Piman diet. At one time the Pima and Papago Indians may have consumed more legumes per capita per day than any other ethnic population in the world (3). Mesquite (*P. velutina*), the Pima's most important wild food, contains a viscous galactomannan in its seeds and pods, which has been shown to lower glycemic responses (25). The viscous mucilages in *Opuntia* cactus pads, flower buds, and fruit may be responsible for the hypoglycemic effects ascribed to these Pima foods (26). In addition, the Pima historically utilized the seeds of a number of plants known to have mucilaginous seed coats, including peppergrass (*Lepidium* spp), Indian wheats (*Plantago* spp), chias (*Salvia* spp), and tansy-mustards (*Descurainia* spp). We hypothesize that the same hygroscopic mucilages, which allow desert plants to survive droughts by slowing mois-

ture vapor loss, may be viscous enough to slow digestion and absorption when incorporated into human diets.

To test these hypotheses, the low GI foods, which formerly dominated the diets of indigenous desert peoples, must be shown to improve  $\beta$ -cell function and/or insulin sensitivity over the long term in individuals predisposed to diabetes, as well as to slow digestion and absorption. Short-term consumption of legumes and other low GI foods by Caucasian diabetics has resulted in improved glucose tolerance and glycemic control (27–29). Similar studies comparing traditional desert foods and modern processed foods need to be carried out in healthy and prediabetic Pima Indian subjects. 

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