

border hydrolysis is the rate-limiting step in the absorption of a tetrapeptide, tetraglycine, indicating that the higher oligopeptides are not absorbed but have to be further hydrolyzed prior to uptake. The membrane peptidases aminopeptidase N, aminopeptidase A, and dipeptidylpeptidase IV are probably those responsible for this final digestion of luminal oligopeptides and thus the complete uptake of a dietary protein. Aminopeptidase N constitutes 5-15% of the total membrane protein and should thus be the dominant one (Sjöström et al., 1978). Studies by Friedrich et al. (1980a,b) indicate that the enzyme in fact plays a central role in the brush border digestion of peptides in vitro. This enzyme was strongly inhibited by the LMW fraction.

About 40% of the LMW fraction is absorbed from the small intestine in the rat (unpublished results). A part of the fraction may thus be present in the epithelial cells at the time of intracellular peptide hydrolysis and may affect the final digestion of (di)peptides, since an effect on an intracellular dipeptidase (Gly-Leu dipeptidase) was observed as well.

The results from the studies on chromatographically separated fractions of the Maillard reaction mixture revealed that no single compound could account for the inhibition of aminopeptidase N or carboxypeptidase A. In addition, the profiles of inhibition of the enzymes by the various fractions were different (see Figure 2). This indicates that the compounds may affect the two enzymes to a different extent. The radioactivity in each fraction may roughly be considered proportional to the total amount of Maillard reaction compounds. The profiles of inhibition did not coincide with radioactivity; hence, the inhibitory effect was not a feature common to the majority of compounds in the mixture. The nature of some of the specific compounds that in fact have inhibitory effects will be reported.

The present study supports the assumption that low molecular weight glucose-lysine reaction compounds influence the uptake of protein in rats (Öste and Sjödin, 1984) due to an inhibition of intestinal proteolytic enzymes. It seems most likely that this effect is mainly the result of specific inhibitions of enzymes of the small intestinal mucosa, rather than an interference with the pancreatic

enzymes. In particular, the inhibition of aminopeptidase N might be crucial.

Registry No. Carboxypeptidase A, 11075-17-5; aminopeptidase N, 9054-63-1; pepsin, 9001-75-6; chymotrypsin, 9004-07-3; aminopeptidase A, 9074-83-3; dipeptidylpeptidase IV, 9032-67-1; glycylleucine dipeptidase, 9025-31-4; alanylproline dipeptidase, 60831-96-1; trypsin, 9002-07-7; carboxylpeptidase B, 9025-24-5.

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Determination of Energy from Moisture Content in Foods Containing Small Amounts of Fat and Dietary Fiber

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It is shown that for a range of 46 foods of all types that contain small amounts ($\leq 3\%$) of fat or dietary fiber, or approximately equal amounts of fat and dietary fiber, the available energy E (kJ/100 g) is inversely correlated with the percent moisture (M) by the equation $E = -17.38M + 1699$, $r = -0.998$, over the range $M = 0-96\%$. The underlying reason for the empirical correlation is given. Comparison of the calculated energies with nearly 200 energies obtained by standard methods for cereals and root crops of moisture content 38-90% shows that the agreement is within about 5-10%. The method is very simple and rapid and should be useful for routine determinations of energies of cereals, bread, root crops, fruit, and vegetables, particularly in simple laboratories in developing countries and elsewhere.

Although there are many different nutrients that are required in the human diet, the two most important are

adequate intakes of energy and protein (FAO/WHO, 1973). The energy intake of a particular diet or of a single food source is normally calculated from the amount of carbohydrate (starch, sugar), protein, fat, and (if present) alcohol that it contains, by the use of suitable factors originally due to Atwater and accepted today with only slight

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modification (Southgate and Durnin, 1970; Davidson et al., 1979). Another method of determination of energy is to measure the heat produced by complete combustion of the food in a bomb calorimeter (gross energy), but corrections are required due to incomplete digestion of components of the diet, incomplete oxidation of protein by the body, and only slight absorption of dietary fiber (Southgate and Durnin, 1970; Van Soest et al., 1982).

Both of these methods require a considerable amount of work and involve equipment and expertise that may not be available in some laboratories in developing countries. The possibility of the development of a simple method arises from a general principle that foods of high energy have low moisture content and vice versa. Indeed, inverse correlations between moisture and energy content and also between moisture and protein were observed for Gambian foods of moisture contents of 40–90% (Hudson et al., 1980).

The basis of an inverse correlation between moisture and energy content can be derived in the following way. The Atwater factors for starch (17.2 kJ/g) and protein (17 kJ/g) are essentially the same, that for sugar (15.7 kJ/g) is somewhat smaller, and that for fat (37 kJ/g) is more than twice that of the others (Davidson et al., 1979). By contrast, the only other constituents of food that contribute appreciably to its weight do not contribute to its energy (water, ash, dietary fiber). Thus, for a food that consists only of starch, protein, sugar, and water, the energy contribution from the first three will depend to a first approximation on their total weight percent (100 - % water); hence, the energy and the water content should be inversely correlated. The presence of large amounts of fat would be expected to increase the energy content of the food and destroy the inverse linear correlation, but equal amounts of fat and [dietary fiber + ash] should approximately balance out in energy terms.

In this paper published data for foods containing low fat, dietary fiber, and ash are used to obtain an energy-moisture correlation over the moisture range (0–96%). Energy values obtained by this method are then compared with those calculated using the Atwater factors from chemical analyses for starch, sugar, fat, and protein or from calorimetry.

RESULTS AND DISCUSSION

The data in Table I from 46 different foods fit a straight line as shown in Figure 1 and give the relation

$$E = -17.38M + 1699 \quad (1)$$

where E is the available energy (kJ/100 g), M is the moisture in percent, correlation coefficient $r = -0.998$ and standard deviation $E = 35.3$. There is virtually no difference in the equation if the 11 protein-rich entries (from jelly to milk in Table I) are removed from the analysis. The 46 foods used to produce Figure 1 and eq 1 were each chosen to contain $\leq 3\%$ fat or dietary fiber or, as in the case of Special K and muesli, to have a difference between fat and dietary fiber of $\leq 3\%$. As would be expected, foods that contain appreciable amounts of fat give much greater energies and fall above the line in Figure 1, whereas those that contain large amounts of dietary fiber (or ash) have lower energies and fall below the line in Figure 1. A representative sample of these foods are tabulated in Table I, but none of these fat-rich or dietary fiber-rich foods are included in the inverse correlation. It is noted that in eq 1 and Figure 1 $E = 0$, when $M = 97.8\%$, indicating that there is on the average 2.2% of material in the food (dietary fiber, ash, etc.) that does not contribute to the energy as calculated.

Table I. Energy and Moisture Contents of Selected Foods

food	moisture, %	energy, kJ/100 g	ref ^a
sugar (sucrose)	0	1680	1
spaghetti, raw	10.5	1612	1
rice	11.8	1530	2
starch	12.0	1480	2
wheat	12.8	1521	2
maize (whole kernel, dried)	13.6	1459	2
rye flour	15.0	1428	1
glucose, liquid	20.4	1355	1
white bread	39.0	991	1
chapatis without fat	45.8	860	1
jelly packet cubes	29.9	1104	1
cod, dried salt boiled	64.9	586	1
haddock, smoked, steamed	71.6	429	1
veal, fillet raw	74.9	459	1
turkey, light meat	75.2	435	1
haddock, steamed	75.1	417	1
whiting, steamed	76.9	389	1
oysters, raw	85.7	217	1
yogurt, low fat	85.7	216	1
eggs, white, raw	88.3	153	1
milk, fresh, skimmed	90.9	142	1
cassava	65.5	564	2
cooking banana	68.2	468	2
barley, pearl, boiled	69.6	510	1
spaghetti, boiled	71.7	499	1
sweet potato	72.3	451	2
breadfruit	72.9	401	2
taro	75.4	393	2
mangoes, canned	74.8	330	1
yam (<i>Dioscorea alata</i>)	76.4	364	2
potatoes, old, raw	75.8	372	1
potato	78.3	343	2
potatoes, boiled	80.5	343	1
pineapples, fresh	84.3	194	1
apples, eating	84.3	196	1
peaches, fresh	86.2	156	1
porridge	89.1	188	1
asparagus, boiled	92.4	75	1
onions, raw	92.8	99	1
radishes, raw	93.3	62	1
melons, cantelope, raw	93.6	102	1
pumpkin, raw	94.7	65	1
lettuce, raw	95.9	51	1
cucumber, raw	96.4	43	1
Special K ^b	2.7	1650	1
muesli ^b	5.8	1556	1

fat-rich foods ^c	moisture, %	energy, kJ/100 g	fat, %	ref ^a
soya flour, full fat	7.0	1871	23.5	1
fruit pie	22.9	1554	15.5	1
yorkshire pudding	56.4	902	10.1	1
avocado pears	68.7	922	22.2	1

dietary fiber-rich foods ^c	moisture, %	energy, kJ/100 g	dietary fiber, %	ref ^a
puffed wheat	2.5	1386	15.4	1
All Bran	2.3	1156	26.7	1
spinach, boiled	85.1	128	6.3	1

^aReference 1: Paul and Southgate, 1979. Reference 2: Thaman and Thomas, 1982. ^bThese entries fit the graph (Figure 1) because the extra energy from fat is roughly balanced by the near zero energy from dietary fiber. Fat and dietary fiber contents are respectively Special K 2.5% and 5.5% and muesli 7.5% and 7.4%. ^cThese results are graphed (but not used in the inverse correlation) to show the effects of larger amounts of fat on increasing energy and larger amounts of dietary fiber on decreasing energy.

The quality of the empirical correlation between energy and moisture given in Figure 1 and eq 1 has been checked out by comparison with three sets of data from the literature and with our extended data set on sweet potato. The results are shown in Table II as the mean difference between the energy determined by standard methods (E_s)

Table II. Comparison of Energies (kJ/100 g Fresh) Determined by Standard Methods (E_s)^a with Energies Calculated from Moisture Content (E)

type of food	no. of samples	range of moisture content, %	$E_s - E$		ref
			mean, over all samples ^b	std dev	
Gambian foods, millet, sorghum, rice, maize, etc.	36 ^c	38-90	-14.0 (-3.2%)	44	Hudson et al. (1980)
taro	36	61-73	-28.2 (-5.5%)	25	Wills et al. (1983)
sweet potato	38	58-78	-6.3 (-1.2%)	12	Heywood and Nakikus (1982)
sweet potato	84	63-89	-9.3 (-2.1%)	26	ACIAR/ANU (1985)

^aThe standard methods involved calculation of energy from the protein, fat, starch, and sugar in the samples except for the results of Hudson et al. (1980) where the gross energy obtained by calorimetry was used, multiplied by 0.87 to give average available energy. ^bThe mean value obtained over all samples is given in terms of kilojoules/100 grams and also in brackets as a percentage of the total mean energy (E_s). ^cTwo results from Table II of Hudson et al. (1980) were omitted that contained raw groundnuts, because of likely high fat content.

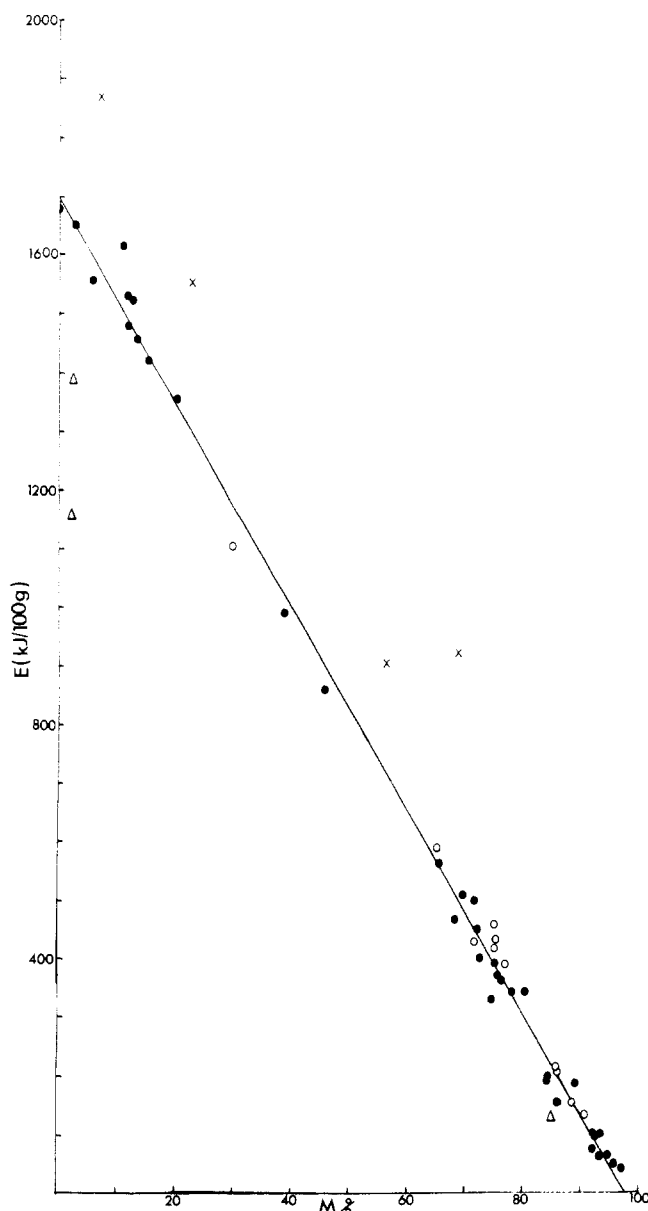


Figure 1. Graph of energy (E in kilojoules/100 grams of fresh weight) against percent moisture (M) for 46 foods: O, protein rich; ●, carbohydrate rich. Some foods rich in fat (x) and some rich in dietary fiber (Δ) are plotted but are not included in the inverse correlation.

and the energy calculated from the moisture content (E) by eq 1. Results are given of $\overline{E_s - E}$ (kJ/100 g) and also as a percent of the total energy (in brackets). This quantity is negative, which indicates that, on average, E calculated by eq 1 overestimates the energy by about

1-5%. However, the magnitude of the standard deviation of $E_s - E$ in Table II shows that for particular samples there is considerable variability in $E_s - E$ from positive to negative values. The results in Table II apply to a range of cereals and root crops with moisture contents in the range of 38-90%. The energies calculated by eq 1 are seen to be in reasonable agreement with those obtained by standard methods.

The likely range of application of this empirical method of calculation of energy from moisture content can be delimited by considering cases in which it will not be applicable. Thus, the method is inapplicable to foods containing >3% fat (unless the dietary fiber is also equally large), which includes meat products, nuts, chocolates, etc (see Table I). Foods containing >3% dietary fiber (unless balanced by an equally large amount of fat) give unacceptably low values, but these tend to be limited largely to breakfast foods (Table I). The method would therefore be useful for those foods that fall outside the above range including cereals, root crops, fruit and vegetables, and low-fat fish and dairy products. It should be useful as a rapid method for routine analysis of foods of generally known composition and low-fat content such as cereals, bread, root crops, vegetables, and fruit. In such cases a careful moisture determination (made by drying to constant weight at 100 °C in an oven) should allow the calculation of the energy content with an accuracy of 5-10% (see Table II). Because of its simplicity, the method should be particularly useful in those laboratories in developing countries and elsewhere that do not have the equipment and/or the expertise to make analyses for protein, fat, starch, and sugar or to use a bomb calorimeter.

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Registry No. H₂O, 7732-18-5; sucrose, 57-50-1; starch, 9005-25-8.

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Legumes and a Cereal with High Methionine/Cysteine Contents

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The amino acid and chemical composition of seeds from three lesser known legume species and an African cereal with high Met + Cys contents is presented. Mesquite (*Prosopis spp.*): 41% protein, 2.5% Met + Cys. Djenkol bean (*Pithecellobium lobatum*): immature seeds, 32% protein, 2.8% Met + Cys; mature seeds, 16% protein, 3.9% Met + Cys. Tamarind seeds (*Tamarindus indica*): 18% protein, 3.5% Met + Cys. Acha (*Digitaria exilis*): 8% protein, 7.3% Met + Cys. Threonine is the first limiting amino acid for mesquite and tamarind while leucine is for Djenkol bean. The overall chemical scores are as follows: mesquite, 55; Djenkol bean, (immature) 31, (mature) 38; tamarind, 80. Tamarind seed protein has a very favorable amino acid balance and deserves further study. These legumes can not only complement cereals but supplement legumes with lower Met + Cys contents as well. The exceptionally high Met + Cys content of the cereal Acha makes it an excellent complement to legumes.

INTRODUCTION

Legumes, as protein-rich crops, are becoming increasingly important sources of plant proteins for human food and animal feed. In 1972-1974, legumes contributed 7% to the total protein supply worldwide (Hoshiai, 1980). With the ability to fix nitrogen from the atmosphere and a much higher efficiency to produce protein per unit land area than animals, legumes hold a promise of meeting protein needs of an increasing world population. However, legume proteins are of lower quality than animal protein due to the limiting amounts of Met + Cys, poor digestibility, and the presence of antinutritional factors.

The low levels of Met + Cys are usually corrected by supplementation with methionine, as in animal feed, or complementation with cereals, as practiced by various human populations. However, there are good reasons for improving the amino acid balance of legumes without resorting to these practices. Addition of methionine to foodstuffs may result in off-flavors caused by bacterial degradation and release of volatile sulfides (Damico, 1975; Bookwalter et al., 1975). Certain populations consume edible roots such as cassava and other starchy foods such as plantain as their chief carbohydrate sources instead of cereal (Bressani, 1973). Due to the very low protein content and a deficient amino acid profile of these starchy foods, which are not complementary with legumes, such population groups can benefit from legumes with improved amino acid balance.

Our objective in this study was to identify lesser known legumes and other plant seeds with high Met + Cys contents. Traditional plant breeding techniques have so far failed to improve the amino acid balance of legumes and other plant foods. Thus, there is a continuing need to

identify legumes with relatively high Met + Cys to provide the genetic base for traditional plant breeding methods or the genes for the rapidly developing tools of genetic engineering. On the molecular level, the genes coding for the (Met + Cys)-rich proteins can be studied with the tools of molecular biology with the aim of enhancing the biosynthesis of such proteins. In our search, we include a cereal of reported high Met + Cys content. We present, in this paper, the amino acid and chemical composition of some lesser known legumes and an African cereal with high Met + Cys.

MATERIALS AND METHODS

Legumes and Cereal Samples. *Prosopis velutina*, *Prosopis alba*, and *Prosopis pubescens* seeds were collected by one of us from mature trees in the Sonoran Desert of Southern California while *Prosopis chilensis*, *Prosopis tamarugo*, and *Prosopis strombulifera* seeds came from mature trees in the Atacama Desert near Pica, Chile. Sample code numbers indicate the field number of the tree and year of collection. Seeds of tamarind (*Tamarindus indica*) were from the Philippines, seeds of Djenkol bean (*Pithecellobium lobatum*) from Indonesia, and seeds of the African cereal Acha (*Digitaria exilis*) from Senegal. The seed coats of Djenkol bean and tamarind were removed before grinding the cotyledon to 80-mesh size. Seeds of *Prosopis spp.* and Acha were ground whole to 80 mesh in an Udy Mill.

Proximate Analysis. All proximate analyses were carried out according to AOAC methods (AOAC, 1980).

Amino Acid Analysis. Ether-extracted milled samples were heated in 6 N HCl under vacuum for 24 h at 110 °C. After filtering, the HCl was removed by evaporation and the amino acid composition of the acid hydrolysates determined with a Durrum amino acid analyzer, Model D-500, by the modified Spackman et al. (1958) ion-exchange method. Cysteine and methionine were determined separately as cysteic acid and methionine sulfone after per-

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