

Cadmium absorption and retention by rats fed durum wheat (*Triticum turgidum* L. var. *durum*) grain*

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A whole-body radioassay procedure was used to assess the retention and apparent absorption by rats of Cd in kernels of durum wheat (*Triticum turgidum* L. var. *durum*) harvested from plants grown hydroponically in ¹⁰⁹Cd-labelled nutrient solution. Wholegrain wheat, containing 5 µmol Cd (570 µg)/kg dry weight labelled intrinsically with ¹⁰⁹Cd, was incorporated into test meals fed to rats that had been maintained on diets containing marginally adequate, adequate or surplus levels of Zn (0.12 mmol (8 mg), 0.43 mmol (28 mg) or 1.55 mmol (101 mg) Zn/kg respectively), and either 0 or 50 g durum wheat/kg. Regardless of diet, all rats consumed about 99 % of the test meal offered. In rats fed diets without wheat, initial Cd absorption averaged 7.7, 4.6 and 2.4 % of the dose when the diet contained 0.12 mmol (8 mg), 0.43 mmol (28 mg) or 1.55 mmol (101 mg) Zn/kg diet respectively. In rats fed wheat-containing diets, initial Cd absorption averaged 3.8 and 2.6 % of the dose when dietary Zn concentration was 0.12 mmol (8 mg) and 0.43 mmol (28 mg)/kg diet respectively. The amount of Cd retained in the body at 15 d postprandial was <2 % of the dose in all rats, and decreased as Zn in the diet increased. Even at 15 d postprandial, 32 to 44 % of the Cd retained in the body was still in the gastrointestinal tract. The results show that: (1) the bioavailability to rats of Cd in wholegrain durum wheat was depressed when wholegrain wheat was part of the regular diet; (2) increased intake of dietary Zn lowered Cd absorption and retention; (3) retention of Cd in the body at 15 d postprandial from diets containing adequate Zn was <1.3 %.

Cadmium retention: Cadmium absorption: Durum wheat: Rats

The amount of Cd in food and agricultural crops varies, and is influenced by several agronomic factors, particularly plant species and soil conditions (Wolnick *et al.* 1983; Mench, 1998; Welch & Norvell, 1999). In this respect, Cd concentrations in durum wheat (*Triticum turgidum* L. var. *durum*) grain harvested in some areas of the northern Great Plains in the USA and adjoining regions of Canada may exceed 0.89 µmol (100 µg)/kg dry weight (Wolnick *et al.* 1983). The Cd content of food is important because excessive intake of Cd may cause several adverse health problems in human subjects (Goyer, 1995; Jarup *et al.* 1998; Satarug *et al.* 2000). To minimize dietary Cd intake, restrictions on the Cd content of unprocessed food products may be imposed. Thus, an upper limit of 0.89 µmol (100 µg) Cd/kg unprocessed durum wheat grain intended for export from the USA may be established

(CODEX Alimentarius Commission, 1999). The proposed limit to the amount of Cd allowable in plant foods is based on the assumption that about 5 % Cd in plant foods is bioavailable (Andersen *et al.* 1992; World Health Organization, 1992). The study presented here was conducted to assess the absorption and retention by rats of Cd in wholegrain durum wheat. Bioavailability represents that portion of the element in the diet that is potentially absorbable from the lumen of the gastrointestinal tract (Welch & House, 1984). The term 'potentially absorbable' is used because the actual amount absorbed may be affected by numerous factors (House, 1999).

Many factors affect Cd bioavailability, including the nutritional status of the test subject, dietary composition and the form of Cd (Fox, 1988). Estimates of absorption or retention of dietary Cd by animals (Flanagan *et al.* 1978;

Abbreviations: MT, metallothionein; WOW8, diet without wheat, containing 0.12 mmol (8 mg) Zn/kg diet; WOW28, diet without wheat, containing 0.43 mmol (28 mg) Zn/kg diet; WOW101, diet without wheat, containing 1.55 mmol (101 mg) Zn/kg diet; WW8, diet with wheat, containing 0.12 mmol (8 mg) Zn/kg diet; WW28, diet with wheat, containing 0.42 mmol (28 mg) Zn/kg diet.

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Jacobs *et al.* 1978; Kostial *et al.* 1978, 1980; Welch *et al.* 1978; Kello *et al.* 1979; Welch & House, 1980; Kiyozumi *et al.* 1982; Jackl *et al.* 1985; Schäfer *et al.* 1986; Fox, 1988; McKenna *et al.* 1992; Reeves *et al.* 1994; Reeves & Vanderpool, 1998; Reeves & Chaney, 2001) and by human subjects (Flanagan *et al.* 1978; McLellan *et al.* 1978; Strehlow & Barltrop, 1988; Berglund *et al.* 1994; Crews *et al.* 2000; Reeves *et al.* 2001; Vanderpool & Reeves, 2001) have been reported, but studies to assess the bioavailability of Cd in wheat are very few, and apparently limited to varieties of common wheat (*Triticum aestivum* L.).

The bioavailability to rats of Cd in wheat bran, wheat endosperm and whole wheat (presumably common wheat, although authors do not specify) has been determined (Wing *et al.* 1992, 1995; Wing, 1993). The accumulation of Cd in tissues of mice provided with either grain of winter wheat (Wagner *et al.* 1984) or wheat bran (Lind *et al.* 1998) has also been reported. In studies with human subjects, Crews *et al.* (2000) studied dietary Cd absorption from porridge made with flour from hydroponically-grown winter wheat (*Triticum aestivum* L.) labelled intrinsically with the stable isotope ^{106}Cd . Results for Cd absorption from diets containing common wheat may not be representative of the absorption from durum wheats. Common wheat and durum wheat differ in many respects, including their ability to absorb and translocate Cd to grain (Hart *et al.* 1998). Many types of durum wheat accumulate two to three times as much Cd in the grain as do common wheats under similar conditions (Chaney *et al.* 1996). To our knowledge, the research presented here is the first to obtain information on the bioavailability to rats of Cd in grain of durum wheat.

Materials and methods

Wheat

Kernels of intrinsically labelled durum wheat (*Triticum turgidum* L. var. *durum*) were obtained from a commercial variety (cultivar Renville), after growth to maturity in hydroponic nutrient solution as described by Hart *et al.* (1998). Radiocadmium as $^{109}\text{CdSO}_4$ (47 MBq, 0.1243 μmol Cd/MBq) was added to the nutrient solution at the beginning of flowering (anthesis), and allowed to accumulate in the developing grain for 2 months. At maturity, the grain was harvested from the plants, and a portion of the grain was autoclaved at 250°C for 15 min,

homogenized, and lyophilized to dryness. Samples of the dried homogenates were assayed for ^{109}Cd activity (model 5530; Packard Instrument Co., Meriden, CT, USA) and wet digested for chemical analysis in $\text{HNO}_3\text{--HClO}_4$ (9:1, v/v). A sample of field-grown durum grain was selected and homogenized for incorporation into experimental diets, which contained whole wheat. Grain from a variety reported to accumulate relatively low amounts of Cd (cultivar Biodur) was chosen to lessen the coincidental addition of Cd into the diets. All grain homogenates were analysed for mineral elements using inductively coupled plasma atomic emission spectrometry with appropriate standards and a reference material (Wheat flour no. 1567a; National Institute of Standards and Technology, Gaithersburg, MD, USA) as described by House *et al.* (1997). Phytate in extracts (Lehrfeld, 1994) of subsamples of dried wheat was determined using a liquid ion chromatography method (DIONEX, 1990).

Animals and diets

Male Sprague–Dawley rats (n 30) were purchased (Charles River, Wilmington, MA, USA) and were housed individually as described previously (House *et al.* 1997). All rats had free access to deionized water and, except during presentation of the test meal, to basic or experimentally modified diets based on the AIN-93G formulation (Reeves *et al.* 1993).

Initially, all rats were fed the basic AIN-93G diet for 4 d. By analysis, concentrations of Cu, Fe, Mn and Zn in the basic diet were about 0.09 mmol (5.7 mg), 0.63 mmol (35.2 mg), 0.18 mmol (9.9 mg) and 0.46 mmol (30.1 mg)/kg diet respectively; Cd concentration was <0.004 μmol (0.450 μg)/kg diet. Rats were then allotted by body weight to six groups of five rats each. Subsequently, each rat within a group was assigned randomly to one of five treatment groups. Body weight of rats (n 6) in each treatment group averaged 75.0 (SEM 0.9) g, and each group was assigned randomly to one of the dietary treatments (WOW8, diet without wheat, containing 0.12 mmol (8 mg) Zn/kg diet; WOW28, diet without wheat, containing 0.43 mmol (28 mg) Zn/kg diet; WOW101, diet without wheat, containing 1.55 mmol (101 mg) Zn/kg diet; WW8, diet with wheat, containing 0.12 mmol (8 mg) Zn/kg diet; WW28, diet with wheat, containing 0.43 mmol (28 mg) Zn/kg diet). Diet composition was confirmed by analysis (Table 1).

Table 1. Amounts of wheat, zinc and cadmium in experimental diets*
(Mean values for duplicate analyses)

Diet treatment	Diet name	Wheat (g/kg)†	Zn		Cd	
			$\mu\text{g/g}$	mmol/kg	$\mu\text{g/kg}$	$\mu\text{mol/kg}$
Without wheat	WOW8	0	8	0.12	<2	<0.02
Without wheat	WOW28	0	29	0.44	<2	<0.02
Without wheat	WOW101	0	101	1.55	<2	<0.02
Plus wheat	WW8	50	8	0.12	6	0.05
Plus wheat	WW28	50	28	0.43	6	0.05

* Except for Zn, all diets contained adequate amounts of nutrients (National Research Council, 1995).

† Wheat added at expense of maize starch to AIN-93G diet (Reeves *et al.* 1993).

Experimental diets contained either 0 or 50 g wholegrain wheat/kg, harvested from field-grown plants. Whole grain was added to the diets to accustom the rats to a wheat diet before dosing. The grain was autoclaved, homogenized, dried and added to the basal diet at the expense of maize starch. In addition, dietary Zn content was adjusted with ZnCO₃ to form three treatments representing marginal, adequate or surplus levels of dietary Zn. By analysis, the Zn levels treatments were 0.12 mmol (8 mg), 0.43 mmol (28 mg) and 1.54 mmol (101 mg)/kg diet respectively.

The experimental diets were fed to the rats for 7 d. After the 7 d adjustment period, rats were denied food for 12 h and then were fed test meals containing wholegrain wheat labelled intrinsically with ¹⁰⁹Cd. Each meal contained 1.0 g dried ¹⁰⁹Cd-labelled wheat homogenate, 1.0 g relevant experimental diet and 0.5 g sucrose. The amount of grain in each test meal was based on our estimate of the amount of ¹⁰⁹Cd required for accurate radioassay. Each test meal contained about 31 kBq ¹⁰⁹Cd and 5.0 nmol Cd (562 ng)/g. Meals were offered to the rats for 3 h and then were replaced with the appropriate diets. Any uneaten portion of the meal was collected and assayed.

As described previously (Welch & House, 1980), rats were assayed for radioactivity in a custom-built whole-body γ -spectrometer immediately after they were fed the test meals, and then at about 24 h intervals for 15 d. Briefly, the assay system consisted of a multi-channel analyser (Canberra Series 35 Plus, Meriden, CT, USA) connected to a special TI-activated NaI crystal (230 × 180 mm) that had a centre well (95 × 100 mm) into which the rat was placed. The crystal was optically coupled to four 76 mm photomultiplier tubes with external magnetic shield and low background tube base assemblies.

Retention of ¹⁰⁹Cd in the whole body was determined daily. Retention data were plotted as a function of postprandial time. Cd absorption at day 0 postprandial was calculated by extrapolation from data collected for days 7 to 15, as described previously (Welch *et al.* 1978; Welch & House, 1980). The calculated % ¹⁰⁹Cd absorbed represented % Cd in the wheat that was bioavailable to the rats. After rats were assayed for ¹⁰⁹Cd on day 15, they were anaesthetized, killed and the gastrointestinal tracts (tissues and contents) and livers were removed and assayed for radioactivity.

Statistical analysis

Data were evaluated statistically using ANOVA procedures; Tukey's procedure was used for mean comparisons (Steel & Torrie, 1960). Animal care and use procedures followed National Institutes of Health and US Department of Agriculture guidelines and were approved by an Institutional Animal Care and Use Committee.

Results and discussion

Wheat

Table 2 shows concentrations of trace elements and of phytate in wholegrain durum wheat. The radiolabelled grain was obtained from hydroponically-grown plants and the field-grown wheat was harvested in ND, USA. Concentrations of micronutrient elements in the field-grown wheat (Table 2) were similar to amounts reported for durum wheat grown in the USA (Zook *et al.* 1970; Erdman & Moul, 1982) and in Italy (Micco *et al.* 1987). Concentrations of Zn in the field-grown and the radiolabelled grain were more than adequate to supply the needs (0.18 mmol (12 mg)/kg diet) of growing male rats (National Research Council, 1995). The concentration of Cd in the field-grown grain incorporated into the experimental diets with wheat was about 1 μ mol (112 μ g) Cd/kg dry weight, which is well within the range commonly found in durum wheat (Zook *et al.* 1970; Erdman & Moul, 1982; Chaney *et al.* 1996). Because a modest amount of this field-grown wheat was added to some of the diets (50 g wheat/kg diet), rats fed these diets continuously ingested a small amount of Cd (about 0.9 nmol (100 ng)/d or 8.9 nmol (1000 ng)/kg body weight). This amount approximates the provisional tolerable weekly intake of Cd suggested as a limit for human subjects (World Health Organization, 1989). The concentration of Cd in the hydroponically-grown ¹⁰⁹Cd-labelled grain was about 5 μ mol (562 μ g) Cd/kg dry weight, which is greater than the range typically found in field-grown durum grain, but not markedly so (Chaney *et al.* 1996).

Cadmium absorption and retention

Regardless of the diet fed prior to providing the test meals, all rats consumed about 99% of the test meal offered

Table 2. Concentrations of trace elements and of phytate in wholegrain durum wheat (*Triticum durum* L. var. *durum*), and certified and observed concentrations of trace elements in a standard reference material

(Mean values for triplicate analyses)

Item	μ mol/kg dry weight						Phytate*	
	Cd	Cu	Mo	Fe	Mn	Zn	μ mol/g	%
Field-grown wheat	0.98	85	8.3	1.11	0.55	0.60	18.8	1.24
¹⁰⁹ Cd-labelled wheat	5.0	56	20.0	1.08	0.24	0.45	5.8	0.38
Wheat flour†								
Certified	0.23	33.1	5.0	0.25	0.17	0.18		
Measured	0.22	29.9	5.0	0.24	0.17	0.18		

* Myo-inositol hexaphosphate plus myo-inositol pentaphosphate.

† Standard reference material no 1567a; National Institute of Standards and Technology, Gaithersburg, MD, USA.

(Table 3). Moreover, because each meal contained the same amount of intrinsically-labelled wheat, all rats consumed the same amount of radio-labelled Cd and about the same amount of total Cd (5–6 nmol (562–674 ng)) from the test meals.

The initial absorption of Cd from the lumen of the rat gut was calculated as the best assessment of the bioavailability of Cd in test meals. Table 3 shows the calculated amount of Cd absorbed by rats fed test meals that contained wholegrain wheat labelled intrinsically with ^{109}Cd . Absorption of Cd was affected by dietary Zn content and by prior dietary intake of wheat. In rats maintained on diets without added wheat, Cd absorption decreased with increased Zn intake and averaged 7.7, 4.6 and 2.3% of the dose when fed diets WOW8, WOW28 and WOW101 respectively. Rats fed the wheat-containing diets absorbed 3.8 and 2.6% of the Cd dose when fed WW8 and WW28 respectively. Generally, postprandial retention of ^{109}Cd declined exponentially with time. Fig. 1 shows the influence of dietary Zn intake on reducing whole-body retention of ^{109}Cd . In addition, as shown in Figs. 2 and 3, rats previously fed wheat-containing diets absorbed and retained less ^{109}Cd from test meals than did rats fed diets without added wheat.

The values presented here for absorption of Cd from the meal (2.4–7.7% dose) are similar to those we reported previously (2–8%) for rats fed lettuce leaves (Welch & House, 1980). Results for apparent absorption of Cd by rats fed sunflower seeds have been more variable, ranging from about 12 to 15% (Reeves & Vanderpool, 1998) to <1% (Reeves & Chaney, 2001). Some of the variation in reported values for Cd bioavailability is the consequence of differences in methods of calculating Cd absorption. If absorption is defined as Cd that has accumulated in the body after transfer from mucosal cells, then calculated values of absorption are much reduced. This occurs because a substantial portion of Cd absorbed initially into the mucosa is not transferred into other tissues, but instead moves through the gut as a result of the sloughing of

mucosal cells. For example, in our present experiment, the calculated % 'absorption' for Cd is reduced by factors of approximately two to four when calculated by the method of Reeves & Chaney (2001), which is based on the transfer of ^{109}Cd to body tissues from mucosal cells. Using this approach, the calculated mean 'absorption' for our present experiment would be 1.9 (SD 0.3), 1.4 (SD 0.3) and 1.2 (SD 0.2)% in rats fed diets WOW8, WOW28 and WOW101 respectively. In rats fed diets with wheat, Cd 'absorption' would have been 1.8 (SD 0.3) and 1.4 (SD 0.4)% for diets WW8 and WW28 respectively. These lower values for 'absorption' are comparable with our results for whole-body retention of ^{109}Cd at 15 d postprandial (Table 3). Regardless of the method used to calculate Cd 'absorption', it is clear that only a relatively small portion of the dose was bioavailable under the conditions described here.

Absorption of dietary Cd is influenced by nutritional status and by dietary intake of several mineral elements (e.g. Zn, Fe and Ca), protein, fibre and phytate. Generally, Cd absorption is enhanced when dietary concentrations of Ca, Fe and Zn are low (Flanagan *et al.* 1978; Jacobs *et al.* 1978; Kello *et al.* 1979; Kostial *et al.* 1980; Hoadley & Cousins, 1985; Schümann *et al.* 1990; McKenna *et al.* 1992; Wing *et al.* 1992; Reeves & Chaney, 2001). In addition, increased intake of phytate and some dietary fibres appears to depress Cd uptake (Kiyozumi *et al.* 1982; Jackl *et al.* 1985; Lind *et al.* 1998), but increased intake of protein appears to enhance Cd absorption and retention (Schäfer *et al.* 1986; Kimura *et al.* 1998). In our present study, dietary amounts of essential nutrients, except for Zn, were provided at or near recommended levels (National Research Council, 1995). Dietary Zn concentration was marginally deficient in two of the diets (WOW8 and WW8, Table 1) but the other diets contained more than adequate amounts of Zn.

Absorption of Cd was higher in rats fed the marginally Zn-deficient diets than in rats fed nutritionally replete diets (Table 3). This inverse relationship between Cd absorption

Table 3. Absorption and retention of cadmium by rats provided single test meals that contained wholegrain durum wheat (*Triticum turgidum* L. var. *durum*) labelled intrinsically with $^{109}\text{Cd}^*$

Diet	Meal†		^{109}Cd absorbed (% dose)‡	^{109}Cd retained at 15 d postprandial (% dose)§		
	Zn content	Consumed		Whole body	Intestine	Liver
	$\mu\text{mol/g}$	% eaten				
WOW8	0.12	99.2	7.7 ^a	1.81 ^a	0.78	0.55
WOW28	0.44	98.9	3.8 ^b	1.33 ^{ab}	0.58	0.40
WOW100	1.55	98.9	2.4 ^b	1.22 ^b	0.41	0.44
WW8	0.12	99.0	4.6 ^b	1.66 ^{ab}	0.62	0.55
WW28	0.43	99.3	2.6 ^b	1.26 ^b	0.41	0.46
SEM		0.1	0.6	0.13	0.12	0.08
HSD¶		0.6	2.6	0.54	0.50	0.33

WW, with wheat; WOW, without wheat; HSD, honestly significant difference.

^{a,b}Mean values with unlike superscript letters were significantly different: $P < 0.05$.

* For details of diets and procedures, see Tables 1 and 2 and p. 500.

† Meals contained 1.0 g ^{109}Cd -labelled wheat, 1.0 g appropriate diet and 0.5 g sucrose.

‡ Calculated from whole-body retention (Welch & House, 1980).

§ ^{109}Cd retained in the whole-body (total), intestinal tract (content and tissues) and liver.

|| Pooled SEM calculated from ANOVA.

¶ Tukey's HSD at $P = 0.05$ (See Steel & Torrie (1960)).

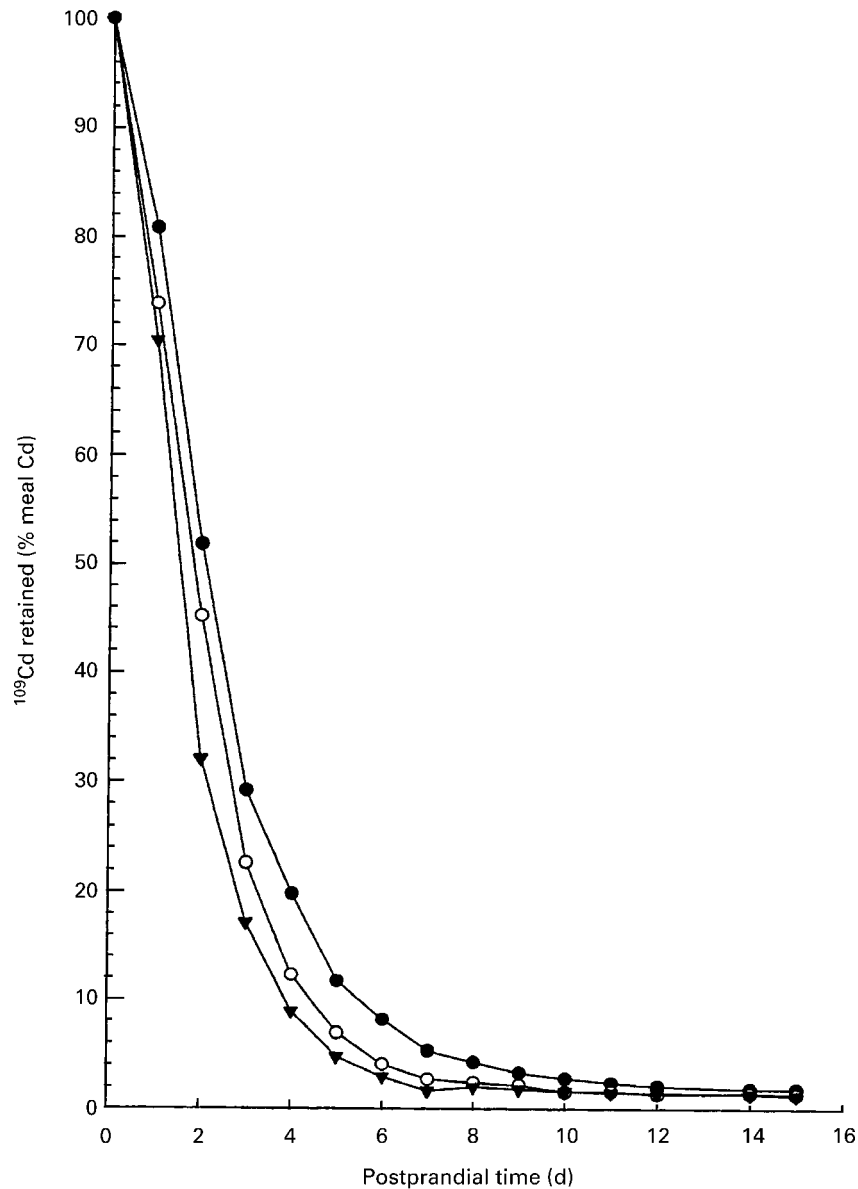


Fig. 1. Whole-body postprandial retention of ^{109}Cd by rats fed diets without wheat grain that contained either 0.12 mmol (8 mg) (●, WOW8), 0.43 mmol (28 mg) (○, WOW28) or 1.55 mmol (100 mg) (▼, WOW100) Zn/kg diet. For details of diets and procedures, see Tables 1 and 2 and p. 500. Values are means for six rats per group.

and dietary Zn content generally agrees with results of previous studies (Jacobs *et al.* 1978; Welch *et al.* 1978; Hoadley & Cousins, 1985; Schümann *et al.* 1990; McKenna *et al.* 1992). Competitive interactions between Cd and Zn may decrease Cd uptake. Moreover, metallothionein (MT), induced within the intestinal mucosa by Zn, may bind Cd because the stability constant of the Cd–MT complex is greater than that of the Zn–MT complex (Schümann *et al.* 1990). Studies with mice lacking MT (Kimura *et al.* 1998) and *in vitro* studies with Caco-2 cells (Blais *et al.* 1999) indicate that intestinal MT has a role in the regulation of Cd absorption. However, saturation of the Cd-binding capacity of intestinal MT in mice did not result in increased absorption of Cd (Lind & Wicklund, 1997).

In our present study, rats that regularly consumed wheat in their diet prior to being fed the test meal absorbed less ^{109}Cd than rats fed the diets without wheat (Table 3). Decreased absorption of Cd by the wheat-fed rats may be attributed to phytate or fibre in the wheat because of formation of insoluble Cd–phytate or Cd–fibre complexes. Others (Wing, 1993; Lind *et al.* 1998) attributed decreased Cd absorption in animals fed wheat bran to the presence of phytate or fibre in the bran. In addition, Jackl *et al.* (1985) reported that phytate markedly depressed Cd absorption in rats. Some dietary fibres have also been reported to depress Cd absorption (Kiyozumi *et al.* 1982). However, in addition to consuming phytate and fibre, rats fed wheat continuously ingested small amounts of Cd. This, too, may have contributed to decreased absorption of Cd

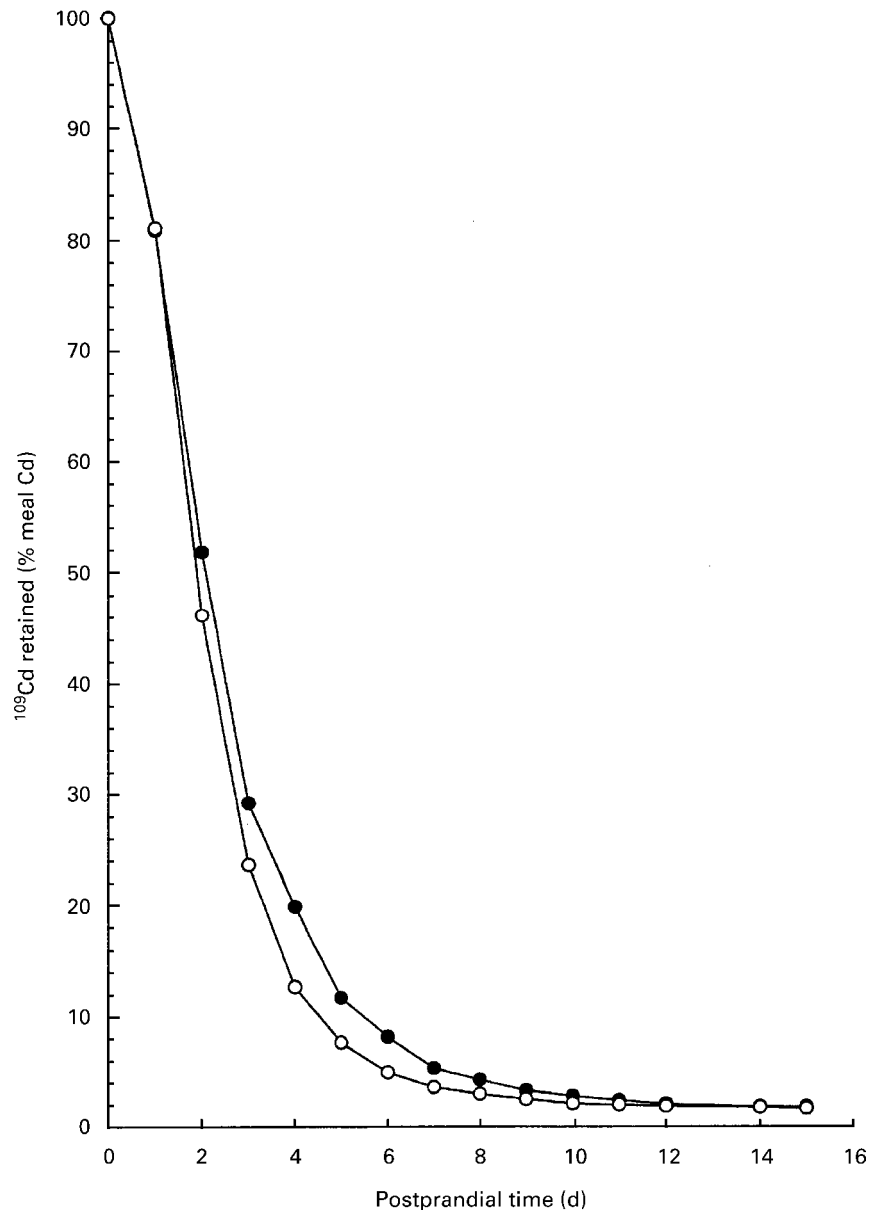


Fig. 2. Whole-body postprandial retention of ^{109}Cd by rats fed diets that contained 0–12 mmol (8 mg) Zn/kg diet and either 0 (●, WOW8) or 50 (○, WW8) g wholegrain durum wheat (*Triticum turgidum* L. var. *durum*)/kg. For details of diets and procedures, see Tables 1 and 2 and p. 500. Values are means for six rats per group.

from the test meal because of adaptation of the intestinal mucosa to the Cd. In this respect, Lind *et al.* (1997) observed that fractional absorption of Cd was higher in mice that received an occasional high dose of Cd than in animals that regularly ingested low amounts of Cd, even though the weekly intake of Cd was the same in both groups. Lind *et al.* (1997) postulated that Cd from an occasional high Cd dose may be more readily absorbed because the Cd may induce non-specific damage to the intestinal mucosa, saturate the capacity of the intestinal mucosa to bind Cd, or alter permeability of tight-junctions between mucosal cells.

A study with mice indicated that Cd in the grain of a variety of common wheat was absorbed less efficiently than was

CdCl_2 (Wagner *et al.* 1984). In addition, Cd in lettuce leaves was more bioavailable to rats than was Cd in spinach (McKenna *et al.* 1992). These studies indicate that the bioavailability to animals of Cd in plant foods may be affected by the form of Cd in the food. Although the forms of Cd present in durum wheat grain are not known, results for common wheats suggest that much of the Cd is likely to be present as complexes of organic compounds. For example, in the common wheat studied by Wagner *et al.* (1984), Cd in the grain occurred mainly as an 11 kDa, aqueous-soluble complex. In a study of human subjects fed ^{106}Cd -labelled wheat, the ^{106}Cd collected in ileostomy fluid was associated with a chemical species having a molecular mass of about 14 kDa (Crews *et al.* 2000).

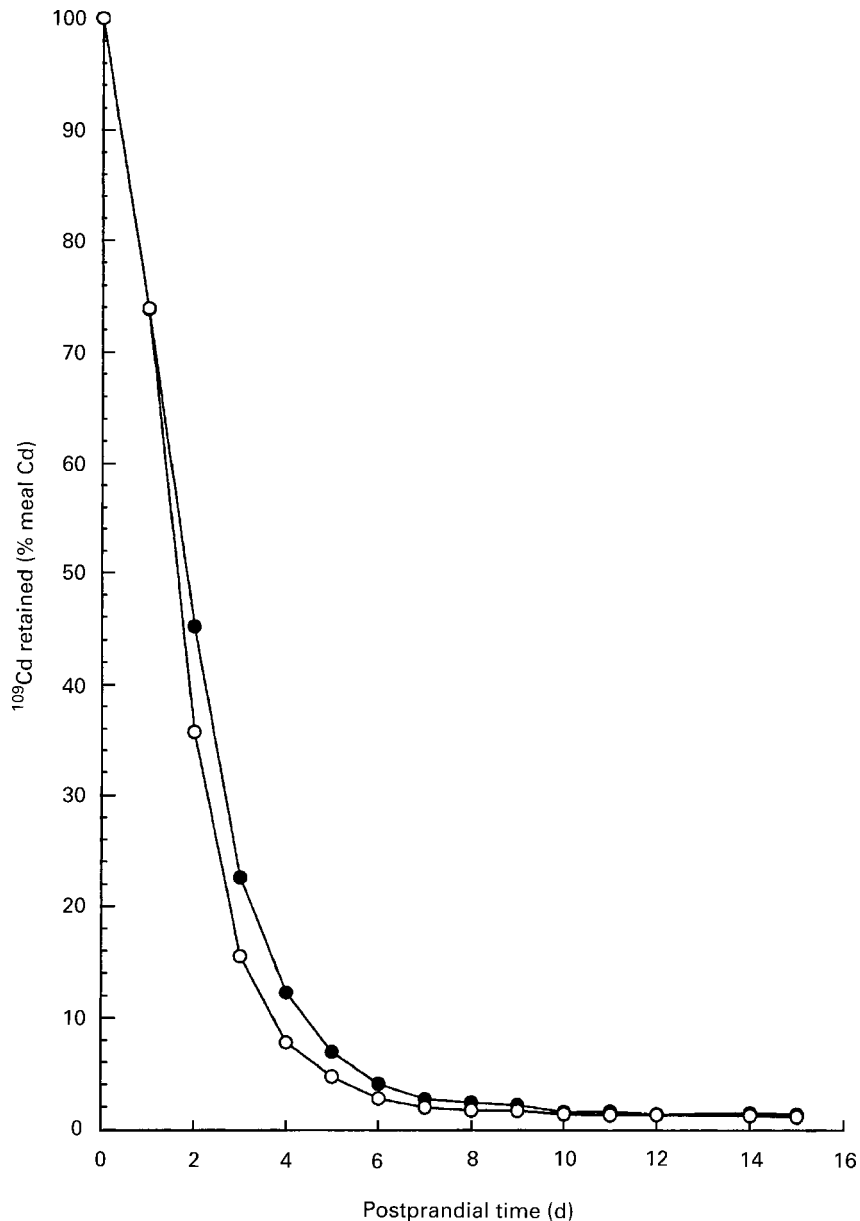


Fig. 3. Whole-body postprandial retention of ^{109}Cd by rats fed diets that contained 0.43 mmol (28 mg) Zn/kg diet and either 0 (●, WOW28) or 50 (○, WW28) g wholegrain durum wheat (*Triticum turgidum* L. var. *durum*)/kg. For details of diets and procedures, see Tables 1 and 2 and p. 500. Values are means for six rats per group.

Table 3 shows the % ^{109}Cd ingested that was retained in the whole body at 15 d postprandial. The amount of ^{109}Cd retained generally declined with increased Zn intake, as would be expected, because increased Zn intake had lowered initial Cd absorption. As the whole-body burden of ^{109}Cd declined, there was a concomitant reduction in the retention of ^{109}Cd in the alimentary tract (tissues and contents) and in the liver. Similarly, Cd retention in various tissues of quail (*Coturnix coturnix*) declined as plant-Zn content increased (McKenna *et al.* 1992). When the diets used in our present study contained the same amount of Zn, retention of ^{109}Cd in the intestine tended to be lower in rats fed the wheat-containing diets (WW8 and WW28) compared with those fed the diets without wheat (WOW8

and WOW28); however, the amount of ^{109}Cd in the liver was similar in rats fed diets with or without wheat. Increased retention of ^{109}Cd in the intestines of the rats fed the low-Zn diets may be an indication that Cd is bound to sulfhydryl sites on cellular membranes, sites that normally would be occupied by Zn. The retention of ^{109}Cd in the liver was less than that previously reported (Jackl *et al.* 1985) when $^{109}\text{CdCl}_2$ was administered to rats fed a low-phytate diet, but our present results on ^{109}Cd retention in the liver are similar to those observed in rats that were fed either high- or low-phytate diets when the ^{109}Cd was administered as ^{109}Cd -labelled phytate (Jackl *et al.* 1985). Moreover, assuming a 7 d half-life for liver Cd (Reeves & Vanderpool, 1998), then the amount

of ^{109}Cd we observed in the liver on day 15 (about 0.5 % of the dose, Table 3) might have declined to an amount similar to that observed (0.3 %) by Reeves & Vanderpool (1998) 20 d after rats were fed ^{109}Cd -labelled sunflower seeds. In our present study, the gastrointestinal tract and liver were the only tissues assayed for ^{109}Cd content; Cd levels were not determined in other animal tissues.

Conclusions

Young, growing rats were used as the model to investigate the bioavailability of Cd in durum wheat. Although the rat may not be an ideal quantitative model for assessing mineral bioavailability for man, the dietary factors that adversely affect the bioavailability of minerals to rats generally affect the bioavailability of minerals to man. Thus, rats can be used to obtain qualitative estimates of mineral bioavailability in foods, and this alternative to studies with human subjects is particularly useful when working with potentially toxic elements or radioactive tracers. Our present results indicate that Zn nutritional status and dietary composition affected the bioavailability to rats of Cd in wholegrain durum wheat. Although our present results do not delineate the specific mechanisms by which Zn or prior intake of wheat suppressed alimentary absorption of Cd by the rats, it is very likely that the bioavailability to people of Cd in durum wheat would be affected in a similar way.

With regard to the bioavailability of Cd in human foods, it is important to recognize that most durum wheat is milled, processed into pasta and then consumed together with other foods as a part of a meal. Many components of wholegrain durum wheat are removed in the milling process. These include most of the phytate and much of the Zn and Cd, because these constituents accumulate in the aleurone layer and germ, which are removed with the bran during milling. Micco *et al.* (1987) reported that about 36, 20 and 44 % of the Cd in wholegrain durum wheat was recovered in semolina, middlings and bran respectively. Thus, much of the Cd in the whole grain is lost during processing, and the bioavailability of the Cd remaining in the food may be different than in the original whole wheat, because of losses during processing of Cd-binding compounds such as phytate.

The bioavailability of mineral elements in plant foods depends on a complex set of interacting factors, including meal composition, form of the element in the food, presence of putative antinutrients (e.g. phytate) that may suppress mineral absorption, and physiological and nutritional status of the individual. In addition, man eats a variety of foods and the importance of a specific food as a source for minerals may be different when the food is consumed as part of a complex diet than when it is consumed separately. Nevertheless, our present results support the concept that animals and human subjects with marginal mineral nutritional status are likely to absorb more Cd than individuals who are nutritionally replete (Reeves & Chaney, 2001). Our present results also suggest that the bioavailability to nutritionally replete people of Cd in durum wheat may be <5 %, because rats typically absorb minerals more efficiently than do human subjects. Controlled

studies with human subjects consuming nutritionally complete diets that contain durum wheat products are needed to fully assess the significance of durum wheat as a source of Cd. Other studies are needed to assess both the form of Cd in durum wheat and the speciation of Cd in the intestinal tract.

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