Enhancement of Rat Intestinal Calcium Absorption by Vanadate (43315)

WILLIAM A. ROWE,[1] TATJANA K. TOMICIC, AND JEAN-JACQUES HAJJAR[2]
Division of Gastroenterology, Department of Medicine, Veterans Administration Medical Center, Newington, Connecticut 06111 and University of Connecticut Health Center, Farmington, Connecticut 06032

Abstract. **Vanadate alters intestinal transport and may have a role in regulating cell function. To determine whether it influences calcium absorption, we tested the effects of acute and chronic vanadate administration on calcium absorption using single-pass perfusion of jejunal and ileal segments of the *in vivo* rat intestine. Acute vanadate administration increased the lumen-to-mucosa and net fluxes of calcium in both the jejunum and ileum. The increase was largely due to an enhancement of the saturable fluxes of calcium and was observed at 10^{-4} *M* concentration of vanadate, but not at higher or lower concentrations of the oxyanion, except at the highest concentration used, 10^{-2} *M*, where calcium absorption was inhibited. Chronic vanadate administration caused, on the other hand, no changes in calcium absorption. We have demonstrated previously that rat intestinal ($Na^+ + K^+$)-ATPase is inhibited by vanadate, an effect that could raise cell sodium and increase the efflux of sodium across the brush border membrane. The results suggest that the vanadate enhancement of calcium absorption may be related to an increased entry of calcium into the mucosa, possibly as a result of an augmented exchange through the Na^+/Ca^+ antiport system. Alternatively, vanadate may influence access to a calcium channel in the mucosal membrane of the intestinal epithelium, leading to the observed increase in absorption.** [P.S.E.B.M. 1991, Vol 198]

Vanadate, the pentavalent oxyanion of vanadium, has been noted to have varying effects on different transport systems. Present in most mammalian tissues (1, 2), vanadate has been shown to inhibit ($Na^+ + K^+$)-ATPase (1, 3), and a variety of other ATPases (4–6) and phosphatases (7, 8). It has, however, no effect on ($H^+ + K^+$)-ATPase (9) and it is stimulatory to some enzymes, such as adenylate cyclase (10–13) and acetylcholinesterase (14). Its effect on Ca-ATPase has generally been inhibitory and demonstrated in many isolated cell systems, including erythrocytes (4, 5, 12), cardiac (12) and skeletal muscle (15) cells, and squid axons (16). Because of the differing actions of vanadate on these transport systems, we tested its effect on intestinal calcium absorption in the *in vivo* rat intestine. We found that acute exposure of the intestine to vanadate enhances calcium absorption, but chronic vanadate ingestion in concentrations that simulate those that occur with pollution (17) causes no significant changes in calcium absorption.

Materials and Methods

Animal Feeding and Pretreatment. Sprague-Dawley rats were used and housed under controlled conditions with a regulated temperature (24°C), humidity, and light:dark schedule (12:12 hr). They were fed Ralston Purina rat chow (Ralston Purina Co., St. Louis, MO) and water *ad libitum* and kept in our animal facility for at least 7 days before they were used in the experiments described below. The diet contained 1.01% calcium, 0.74% phosphorus, 0.21% magnesium, and 3.3 IU of vitamin D per gram of diet. In one set of experiments, in which the acute effect of vanadate was tested, rats with an average weight of 300 g were used. In another set, rats with a mean initial weight of 175 g were chronically fed vanadate, and, by the time the absorption studies were performed, they had grown to an average weight of 290 g. Chronic vanadate administration was done by a similar method, as we have reported earlier (18). The rats were randomized into a control group ($n = 8$) and two vanadate-fed groups (each group, $n = 8$). The control group was given the

[1] Present address: Division of Gastroenterology, The Johns Hopkins University School of Medicine, Baltimore, MD 21205.

[2] To whom correspondence and requests for reprints should be addressed at Veterans Administration Medical Center, Building 5, 555 Willard Ave, Newington, CT 06111.

Received March 21, 1991. [P.S.E.B.M. 1991, Vol 198]
Accepted June 6, 1991.

0037-9727/91/1982-0754$3.00/0

Table I. Effect of Intraluminal Perfusion of Varying Vanadate Concentrations on Lumen-to-Mucosa and Net Calcium Fluxes in Jejunal Segments of the *In Vivo* Rat Intestine[a]

Vanadate concentration (*M*)	Net absorption (μmol/hr/g wet wt)	Lumen-to-mucosa flux (μmol/hr/g wet wt)
0	4.86 ± 0.21	6.56 ± 0.25
10^{-6}	4.97 ± 0.18	6.71 ± 0.26
10^{-5}	5.46 ± 0.23	7.37 ± 0.32
10^{-4}	7.23 ± 0.23[b]	9.73 ± 0.36[b]
10^{-3}	5.32 ± 0.22	7.18 ± 0.31
10^{-2}	3.11 ± 0.17[c]	4.20 ± 0.18[b]

[a] Values are means ± SE of six rats in each of the six groups randomly allocated to perfusion with varying concentrations of vanadate. Calcium concentration in the perfusate was 2.5 m*M*.
[b] $P < 0.01$, compared with control group not perfused with vanadate (Student's *t* test).
[c] $P < 0.05$, compared with the control group not perfused with vanadate (Student's *t* test).

chow diet and water *ad libitum* and the vanadate-fed groups were pair-fed the same diet, but given vanadate (orthovanadate; Pfaltz and Bauer Inc., Stamford, CT) administered through drinking water at a concentration of 75 ppm. This concentration was chosen because it produces changes in intestinal transport without compromising the nutrition of the rats during vanadate feeding (19). Both the control group and vanadate-fed groups were maintained on their respective regimens for a period of 7–8 weeks, at the end of which they were tested in the experiments described below.

***In Vivo* Absorption Studies.** Intestinal absorption of calcium was determined using a modification of the single-pass *in vivo* perfusion technique described by Younoszai *et al.* (20). Rats were anesthetized with intraperitoneal injections of urethane (1 g/kg body wt). The abdominal cavity was opened by a midline longitudinal incision and the entire small intestine was perfused as two separate jejunal and ileal segments. The jejunal segment was cannulated, with an inlet cannula located 1-cm distal to the ligament of Treitz and a distal cannula at the end of the proximal third of the intestine (30–40 cm). The ileal segment was similarly cannulated at both of its ends, and it comprised the remaining distal two thirds of the intestine, extending to 1-cm orad to the ileocecal junction. The two segments were flushed with isotonic saline and air and then re-placed in the abdomen. The rats were perfused while placed on a heating pad that maintained their rectal temperature at 37°C. The segments were perfused *in situ* at a rate of 0.3 ml/min with a solution containing 120 m*M* NaCl, 5 m*M* KCl, an appropriate concentration of $^{40}CaCl_2$ (0.85, 1.7, 2.5, or 3.4 m*M*), $^{45}CaCl_2$ (1×10^5 dpm/ml), [^{3}H]polyethylene glycol ([^{3}H]PEG) mol wt 4000), and mannitol in sufficient amounts to supplement the solutes and to achieve an osmolality of 310 mOsm/kg. In addition, sodium orthovanadate (frequently 0.1 m*M*) was included in the perfusates of the experiments that tested the acute effects of this oxyanion on calcium absorption. In previous studies (3, 18, 19, 21), this concentration caused changes in the transport of sodium, amino acids, and sugars, as well as changes in $(Na^+ + K^+)$-ATPase and adenylate cyclase. The perfusate collected during the first 60 min was discarded, and samples collected during three consecutive 20-min periods were tested for their $^{40}Ca^{2+}$ concentrations and their contents of ^{45}Ca and [^{3}H]PEG.

Table II. Effect of Acute Vanadate Administration on Lumen-to-Mucosa and Net Calcium Fluxes in Jejunal and Ileal Segments of the *In Vivo* Rat Intestine[a]

	Luminal calcium (m*M*)			
	0.85	1.70	2.50	3.40
Lumen-to-mucosa flux (μmol/hr/g wet wt)				
Jejunum				
Control	5.10 ± 0.21	6.02 ± 0.22	6.60 ± 0.27	6.79 ± 0.28
Vanadate	7.18 ± 0.30	8.27 ± 0.30	8.62 ± 0.36	9.83 ± 0.41
Ileum				
Control	2.22 ± 0.09	2.38 ± 0.09	2.44 ± 0.10	2.49 ± 0.10
Vanadate	3.16 ± 0.13	3.46 ± 0.12	3.55 ± 0.15	3.65 ± 0.15
Net absorption (μmol/hr/g wet wt)				
Jejunum				
Control	3.78 ± 0.16	4.51 ± 0.16	4.89 ± 0.20	5.19 ± 0.22
Vanadate	5.45 ± 0.23	6.85 ± 0.25	7.12 ± 0.29	7.34 ± 0.30
Ileum				
Control	0.82 ± 0.03	0.95 ± 0.16	0.99 ± 0.18	1.03 ± 0.04
Vanadate	1.08 ± 0.04	1.24 ± 0.05	1.30 ± 0.05	1.34 ± 0.06

[a] Values are means ± SE, $n = 6$–8. All flux rates in the presence of vanadate (0.1 m*M*) are significantly higher ($P < 0.05$) than the corresponding rates in control tissues (Student's *t* test).

Total calcium was measured by a fluorometric method using the Calcette Automatic Calcium Titrator (Precision Systems, McGaw Park, IL). ^{45}Ca and ^{3}H were determined in a two-channel liquid scintillation counter (Packard, Tri-Carb 4000). At the end of the experiments, the rats were sacrificed by an added dose of the anesthetic and the intestinal segments were resected and their length, wet weights, and dry weights were determined as described previously (22). Calcium absorption, which represents the disappearance of calcium from the luminal perfusates, and the lumen-to-mucosa (LM) flux, which represents the influx of calcium into the mucosa, were calculated by the following formulas:

$$\text{Net absorption} = \frac{V[^{40}Ca_i - (^{40}Ca_f)\,(PEGR)]}{W \times t} \quad [1]$$

$$\text{LM flux} = \frac{V\,[^{45}Ca_i - (^{45}Ca_f)\,(PEGR)]}{[(SA_i + SA_f)/2]\,W \times t} \quad [2]$$

in which V refers to the volume of the perfusates per unit time; i and f, the initial and final luminal values; ^{40}Ca, the chemical calcium content of the luminal fluid; ^{45}Ca, the radioactive calcium content of the fluid; PEGR, the ratio of initial to final polyethylene glycol concentrations; SA, the specific activity of the luminal fluid; W, the weight of the intestinal segment perfused; and t, the time period of perfusion. Absorption was calculated per unit wet weight, dry weight, and length of the perfused segments, and it varied uniformly among the three parameters under the different experimental conditions. The data are presented as means ± SE of absorption per unit wet weight, as the absorption rates per unit length or dry weight of the perfused segments are not comparatively different from the absorption data calculated per wet weight of the perfused segments. Comparisons between the different control and experimental groups were made using the Student's t test for unpaired independent samples.

Sodium Efflux Measurements from Mucosa to Lumen. In a previous publication (23), ouabain, an inhibitor of the Na^+ pump, was found to increase the efflux of sodium across the mucosal barrier of the intestinal cell. Since vanadate has also been found to have a similar inhibitory action on the Na,K pump in the intestine, we tested its effect on the efflux of sodium from the intestinal mucosa. Such an effect was considered relevant to calcium uptake by the intestine, since an increase in the sodium efflux may enhance calcium influx in exchange for sodium. $^{22}NaCl$ efflux was compared between normal tissues and tissues exposed to vanadate. The efflux was measured by a method similar to that described previously (3, 23). A segment of the jejunum was everted and incubated for 40 min in a phosphate-buffered Krebs solution containing $^{22}NaCl$ (NEN, Research Products-Dupont, Boston, MA). The incubation solution was oxygenated with 100% oxygen and kept at 35°C. At the end of the loading period, the tissue was blotted with Whatman No. 1 filter paper, cut into a flat sheet, and mounted in a Lucite chamber with an exposed rectangular area of mucosal surface equal to 2.5 cm^2. The mucosal surface was exposed to nonlabeled buffer solution (2 ml, 35°C) for a period of 1 min, during which the solution was bubbled with 100% oxygen. The washout solution was removed by suction into a counting vial and fresh solution was replaced in the chamber. Washout was followed for 10 1-min periods, at the end of which the exposed tissue was cut out and extracted in 0.1 N HNO_3. Washout samples and tissue extracts were counted and the rate of change of radioactivity in the tissue was used in calculating the rate coefficient of the efflux process (3, 22). Vanadate was added to the incubation and washout solutions and its effect was compared with control tissues not exposed to vanadate by using the paired Student's t test.

Results

Initial studies shown in Table I revealed that a 10^{-4} M vanadate concentration produced a significant in-

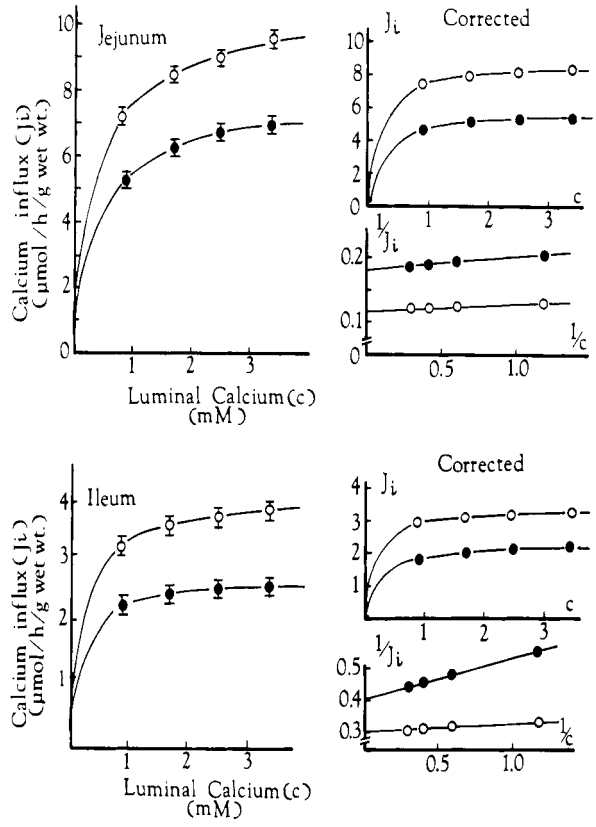

Figure 1. The relationship between luminal calcium concentration and lumen-to-mucosa calcium flux (means ± SE) in jejunal and ileal segments of normal control rats and rats perfused with 0.1 mM vanadate. The saturation curves were drawn using the kinetic constants shown in Table III and as determined by the double reciprocal plots in the right panels. Over the entire concentration range, calcium influx was significantly ($P < 0.05$) higher in the vanadate-treated segments versus the controls. K_a, which reflects nonsaturable calcium transport, was calculated in μmol/hr/g wet wt/mmol, as described in Ref. 25, and was 0.69 and 0.28 for jejunal and ileal influx, respectively, in the controls, and 0.63 and 0.24 in the vanadate-treated tissues.

Table III. The Effect of Acute Vanadate Administration on the Kinetic Constants of the Flux of Calcium from Lumen to Mucosa

	Lineweaver-Burk		Eadie-Hofstee	
	Control	Vanadate	Control	Vanadate
Crude flux data				
Jejunum				
K_t[a]	0.42	0.39	0.44	0.47
V_{max}[b]	7.63	10.37	7.67	10.81
Ileum				
K_t	0.14	0.17	0.14	0.18
V_{max}	2.58	3.82	2.58	3.83
Data corrected for nonmediated fluxes				
Jejunum				
K_t	0.173	0.092	0.192	0.137
V_{max}	5.75	8.40	5.81	8.55
Ileum				
K_t	0.299	0.099	0.303	0.103
V_{max}	2.45	3.30	2.46	3.31

[a] Data expressed in millimolars.
[b] Data expressed in μmol/hr/g wet wt.

crease in both the net and LM fluxes of calcium. The increase was not evident at vanadate concentrations that were lower or higher than 10^{-4} *M*, but a 10^{-2} *M* concentration produced a significant decrease in the calcium fluxes. The effect of intraluminal perfusion of 10^{-4} *M* vanadate on calcium absorption was studied in the presence of four calcium concentrations and compared with a group of controls that were similarly perfused, but without vanadate. Eight rat groups were randomized and their body weights and the lengths and wet weights of their jejunal and ileal segments were comparable. The results of these studies are shown in Table II, in which vanadate, in comparison to the controls, uniformly increased the LM and net calcium fluxes. A significant increase was observed at all of the calcium concentrations tested and was evident in both jejunal and ileal segments. To further evaluate this stimulatory effect of the 10^{-4} *M* vanadate concentration, we computed the changes in the kinetic properties of calcium absorption in the presence and absence of vanadate. Figure 1 shows a plot of the LM flux (influx) as a function of the luminal concentration of calcium. As shown, there is, in both the jejunum and ileum, a tendency for the influx rate to saturate with rising calcium concentrations. Estimates of the nonmediated components of the curves shown in the left panels of Figure 1 were computed as described by Schedl *et al.* (24, 25), and the corrected values that are presented in the right panels of Figure 1 demonstrate the double reciprocal plots shown. The plots suggest that stimulation of the calcium influx may not be mediated through a competitive mechanism that shares the same characteristics as those that exist under normal conditions in the jejunum or ileum. A similar plot (data not shown) was also made for the net calcium absorption data and similar changes in kinetic characteristics were also noted. The kinetic constants of the corrected calcium influx measurements were calculated using both Lineweaver-Burk and Eadie-Hofstee plots (Table III). The effect of vanadate, in both cases, is due to both a decrease in K_t and an increase in V_{max}.

The effect of chronic vanadate administration was studied in three groups of randomized rats ($n = 8$ per group). Two of the groups were given vanadate in drinking water at a concentration of 75 ppm and the third group was given tap water, which is estimated to contain 100 parts of vanadate per billion (26). In previous publications (18, 19), we found that the ingestion of vanadate in drinking water in a concentration of 75 ppm was optimal for testing the effect of chronic vanadate administration. This concentration caused no changes in food intake or in the rate of growth of the rats, both of which remained similar in the three groups studied. As compared with controls, chronic exposure of the intestine to vanadate (Table IV) causes no changes in net calcium absorption or in calcium influx. The vanadate-fed rats, however, still respond to acute vanadate exposure and, as shown in the third column of Table IV, acute vanadate increases their calcium absorptive fluxes.

Table V shows the effect of vanadate on ^{22}Na efflux. The efflux of sodium increased significantly in the vanadate-treated tissues as compared with the untreated controls. This finding is similar to what we have observed previously in the rabbit intestine that was treated with ouabain (23). In the latter case, ouabain inhibited the Na^+ pump and caused a rise in cell sodium that increased the extrusion of sodium from the cell to the intestinal lumen.

Discussion

Vanadate, a known inhibitor of Ca-ATPase (4–6), was demonstrated, contrary to expectations, to have a

Table IV. Effect of Chronic Vanadate Administration on Ca^{2+} Absorption[a]

	Ca^{2+}	Control	Vanadate-fed	
			Chronic	Acute on chronic
Lumen-to-mucosa flux				
Jejunum	0.85	4.98 ± 0.28	5.26 ± 0.27	5.58 ± 0.31
	3.4	6.41 ± 0.36	6.72 ± 0.37	8.85 ± 0.49[b,c]
Ileum	0.85	2.15 ± 0.12	2.24 ± 0.13	2.49 ± 0.14
	3.4	3.19 ± 0.18	3.29 ± 0.18	3.66 ± 0.20
Net flux				
Jejunum	0.85	3.69 ± 0.21	3.89 ± 0.19	4.41 ± 0.25[b]
	3.4	4.74 ± 0.26	4.97 ± 0.28	6.99 ± 0.38[b,c]
Ileum	0.85	0.76 ± 0.04	0.74 ± 0.04	0.99 ± 0.06[b,c]
	3.4	1.01 ± 0.06	1.12 ± 0.06[b]	1.46 ± 0.08[b,c]

[a] Results are means ± SE of lumen-to-mucosa and net fluxes in μmol/hr/mg wet wt ($n = 8$). Ca^{2+} is the initial luminal calcium concentration in millimolars.
[b] Statistically significant ($P < 0.05$), as compared with control.
[c] Significant ($P < 0.05$), as compared with values under the chronic condition (Student's t test).

Table V. Effect of Acute Vanadate Administration on Sodium Efflux Across the Mucosal Border of Rat Jejunum

Sodium efflux		Ratio of vanadate to control
Control (μmol/hr/mg dry wt)	Vanadate (μmol/hr/mg dry wt)	
3.49	4.19	1.20
4.11	5.38	1.30
3.11	3.42	1.10
3.19	3.83	1.20
3.44	3.96	1.15
4.49	4.94	1.10
3.99	4.91	1.23
4.18	5.31	1.27
Mean ± SE		1.19 ± 0.07

stimulatory action on intestinal calcium absorption. This effect was observed mainly when vanadate was administered acutely, and it occurred in both jejunal and ileal segments of the rat intestine. The stimulation of calcium absorption by vanadate was characterized by an increase in V_{max}, as well as a decrease in K_t. The changes in the kinetic parameters and the double reciprocal plots shown in Figure 1 suggest that the vanadate enhancement of calcium absorption occurs through a nonspecific transport process that may or may not be related to the normal mechanism of absorption of calcium.

It is not possible strictly from the present *in vivo* studies to characterize the mechanism of action of vanadate on calcium absorption. However, hypothesis can be suggested based on the current findings and uptake studies in isolated cells and membrane vesicles (4, 5, 27, 28). There is evidence to suggest that vanadate increases the absorption of calcium by increasing the saturable component of calcium absorption. As shown in Figure 1, when correction for the nonmediated component of the calcium influx was made, a saturable absorptive curve was obtained and the tissues exposed to vanadate continued to absorb significantly more calcium than the control tissues. This suggests that the vanadate effect occurs through enhancement of a calcium-saturable transport mechanism. The effect may involve at least two possible sites of action on the calcium transport mechanisms, an effect on the entry across the mucosal border of the cell and another on the exit across the basolateral membrane. We have observed previously (21) that vanadate in a concentration similar to that used in the present communication causes inhibition of $(Na^+ + K^+)$-ATPase of the rat intestine. Such an effect raises the cell Na^+ concentration and increases, as observed in the present study, the extrusion of Na^+ across the mucosal membrane. If we assume that a Na^+/Ca^{2+} exchanger exists in the mucosal brush border membrane, then the extrusion of Na^+ may enhance calcium entry through this antiport mechanism. Such an assumption, however, is only speculative at present and it needs to be confirmed by studies using brush border membrane vesicles. The efflux of Na^+ across the basolateral membrane may also increase, but the exchange of Na^+ for Ca^{2+} across this barrier was found to have a limited role in calcium exchange across this barrier (27, 28). Extrusion is believed to occur primarily through activation of the Ca^{2+}-ATPase, which remains functional despite the presence of the lower concentrations of vanadate that were tested. This is contrary to what is reported in *in vitro* studies of many cell systems in which vanadate was found to inhibit Ca^{2+} pump (4, 5, 12, 15, 16). However, it is possible that the vanadate concentration that was perfused in the *in vivo* conditions of the present experiments, though inhibitory to $(Na^+ + K^+)$-ATPase, was not high enough to inhibit the Ca^{2+} pump. This is conceivable, since Ca^{2+}-ATPase is 3 times less sensitive to vanadate than $(Na^+ + K^+)$-ATPase (5).

Unlike the acute effect of vanadate, chronic vanadate administration causes no changes in calcium ab-

sorption. This may be due to at least three possible reasons. First, the amount of vanadate that was chronically administered may be too small to produce an effect. It was, however, difficult to administer a larger dose of vanadate to rats without interfering with their nutrition. In previous studies (18, 19), we found that this dose is optimal because it causes changes in intestinal transport without interfering with the growth and nutrition of the rats during the period of its administration. Second, the chronic effect of vanadate may cause secondary adaptive changes of the intestine, leading to tolerance and insensitivity to the vanadate effect. This possibility is, however, unlikely, since acute on chronic vanadate administration causes a similar stimulation of calcium absorption by the intestines of control rats and of rats that are given vanadate on a chronic basis. The third possibility is that the specific calcium absorptive rate measured per wet weight of the intestinal tissue may represent an underestimate because of swelling of the intestinal epithelium. Calculation of absorption per dry weight of mucosa reveals an average 8% increase with chronic vanadate, as compared with a 22% increase with acute administration of the oxyanion. These findings are not comparatively different from those obtained when absorption is calculated per wet weight of intestine. The chronic effect of vanadate is, therefore, relatively minimal and not comparable to the acute effect. These findings, again, suggest that the effect of vanadate on calcium absorption is related to differences in the accessibility of the oxyanion to the active Na^+ and Ca^{2+} transport sites.

This study was supported by the Medical Research Service of the Veterans Administration.

1. Cantley LC, Josephson L, Waener R, Yanagisawa M, Lechene C, Guidotti G. Vanadate is a potent (Na,K)-ATPase inhibitor found in ATP derived from muscle. J Biol Chem **252**:7421–7432, 1977.
2. Schroeder HA, Balassa JJ, Tipton IH. Abnormal trace metals in man—Vanadium. J Chronic Dis **16**:1047–1071, 1963.
3. Hajjar JJ, Rowe WA, Tomicic TK. The nonspecific nature of the vanadate inhibition of rat ileal (Na,K)-ATPase. Life Sci **43**:1741–1746, 1988.
4. Barrabin H, Garrahan PJ, Rega AF. Vanadate inhibition of the Ca^{2+}-ATPase from human red cell membranes. Biochim Biophys Acta **600**:796–804, 1980.
5. Bond GH, Hudgins PM. Inhibition of red cell Ca^{2+}-ATPase by vanadate. Biochim Biophys Acta **600**:781–790, 1980.
6. Wajsman R, Walters JR, Weiser MM. Identification and isolation of the phosphorylated intermediate of the calcium pump in rat intestinal basolateral membranes. Biochem J **256**:593–598, 1988.
7. Van Etten RL, Waymack PP, Rehkop DM. Transition metabolism inhibition of enzyme-catalyzed phosphate ester displacement reactions. J Am Chem Soc **96**:6782–6785, 1974.
8. Lopez V, Stevens T, Lindquist RN. Vanadium ion inhibition of alkaline phosphatase-catalysed phosphate ester hydrolysis. Arch Biochem Biophys **175**:31–38, 1976.
9. Sachs G. Vanadate as a transport probe. J Lab Clin Med **96**:379–381, 1980.
10. Krawietz W, Werman K, Erdmann E. Stimulatory effect of vanadate on the adenylate cyclase of cardiac tissue. Biochem Pharmacol **28**:2517–2520, 1979.
11. Schwabe U, Puchstein C, Hannemann H, Sochtig E. Activation of adenylate cyclase by vanadate. Nature **277**:143–145, 1979.
12. Erdman E. Cardiac effects of vanadate. Basic Res Cardiol **75**:411–412, 1980.
13. Schmitz W, Hackbarth I, Scholtz H, Wetzel E. Effects of vanadate on the cAMP system of the heart. Basic Res Cardiol **75**:438–443, 1980.
14. Catalan RE, Martinez AM, Aragones MD, Godoy JE. Activation of acetylcholinesterase by vanadate. Neuropharmacology **24**:1119–1122, 1985.
15. Nelson MT, Blaustein MP. Effect of ATP and vanadate on calcium efflux from barnacle muscle fibers. Nature **289**:314–316, 1981.
16. Dipolo R, Rojas HR, Beauge L. Vanadate inhibits uncoupled Ca efflux but not Na-Ca exchange in squid axons. Nature **281**:228–229, 1979.
17. Goldberg ED, Griffin JJ, Hodge V, Koide M, Windom H. Pollution history of the Savanah River Estuary. Environ Sci Technol **13**:588–594, 1979.
18. Hajjar JJ, Dobish MP, Tomicic TK. Effect of chronic vanadate ingestion on amino acid and water absorption in rat intestine. Archives Toxicology **63**:29–33, 1989.
19. Hajjar JJ, Dobish MP, Tomicic TK. Effect of acute and chronic vanadate administration on sugar transport in rat jejunum. Proc Soc Exp Biol Med **190**:35–41, 1989.
20. Younoszai MK, Nathan R, Yokoyama L. Intestinal calcium absorption is ehnahced by D-glucose in diabetic and control rats. Gastroenterology **88**:933–938, 1985.
21. Hajjar JJ, Zakko S, Tomicic TK. Effects of vanadate on water and electrolyte transport in rat jejunum. Biochim Biophys Acta **863**:325–328, 1986.
22. Green RS, MacDermit RG, Scheig RL, Hajjar JJ. Effect of ethanol on amino acid absorption across the in vivo rat intestine. Am J Physiol **241**:G176–G181, 1981.
23. Curran PF, Hajjar JJ, Glunn IM. The sodium-alanine interaction in rabbit ileum: Effect of alanine on sodium fluxes. J Gen Physiol **55**:297–308, 1970.
24. Iskandarani M, Miller DL, Schedl HP. Kinetics of calcium transport in the hamster duodenum and ileum. Gastroenterology **81**:903–909, 1981.
25. Schedl HP, Wilson HD, Miller T, Ogesen B. Calcium uptake by intestinal brush border membrane vesicles. Comparison with in vivo calcium transport. J Clin Invest **76**:1871–1878, 1985.
26. Mitchell RL. Emission spectrochemical analysis determination of trace elements in plants and other biological materials. In: Yoe JH, Koch HJ, Eds. Trace Analysis. New York: John Wiley, p398, 1957.
27. Favus MJ. Factors that influence absorption and secretion of calcium in the small intestine and colon. Am J Physiol **248**:G147–G157, 1985.
28. Nellans HN, Popovitch JR. Role of sodium in intestinal calcium transport. In: Bronner F, Peterlik M, Eds. Epithelial Calcium and Phosphate Transport. New York: Alan R. Liss, Inc., p301, 1984.