Research Article

Impact of Gastric pH and Dietary Fiber on Calcium Availability of Various Calcium Salts

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Abstract

The objective of this study was to compare calcium release and ionization from commercial calcium supplements and various salts of calcium with different solubilities. The impact of pH of test fluid and dietary fiber co-administered with calcium supplement on calcium availability was also studied. Compressed tablets of six different calcium salts were prepared by direct compression. Calcium release from tablets was tested in simulated gastric fluid without pepsin (SGF, pH 1.2) or fed state simulated gastric fluid (FSSGF, pH 4.8), using USP dissolution apparatus. The ionized calcium concentration in test fluid was determined by calcium ion selective electrode. The results demonstrated that most of the formulations released calcium within 1 hour. Tablets made of higher solubility calcium salts demonstrated a faster calcium release. The calcium release from calcium hydrogen phosphate and calcium carbonate tablets in gastric fluid with higher pH showed a slower calcium release than in SGF. The dietary fiber co-administered with calcium supplement influenced the calcium availability, especially in medium with higher pH. The results suggested that most of the calcium salts could be used as calcium supplement. However, the use of calcium carbonate required an acidic environment in order to be dissolved in the gastrointestinal tract.

Key Words: Calcium; Tablet; Gastric pH; Pectin; Dietary fiber

Introduction

Calcium is an essential mineral found in great abundance in the human body. More than 99% of total body calcium is found in the bones and teeth where it functions to support their structure (Shils, 1999). The remaining 1% is in the blood, muscle and the fluid between cells. Calcium plays important roles in nerve conduction, muscle contraction, and blood clotting. A constant level of calcium is maintained in body fluid and tissues so that these vital body processes function efficiently. When calcium intake is low or calcium is poorly absorbed, bone breakdown occurs because the body must use the calcium stored in bones to maintain normal biological functions such as nerve and muscle function. Bone loss also occurs as a part of the aging process. A key example is the loss of bone mass observed in postmenopausal women because of decreased amounts of the hormone estrogen (Riggs and Melton, 1995). Therefore, it is important to

consume enough calcium to maintain adequate blood and bone calcium levels.

One way to help reduce the risk is to consume adequate amounts of calcium in daily diet. The Dietary Reference Intake (DRI) for calcium for adults is 1000-1300 mg depending on age and gender. A balanced diet with calcium-rich foods, such as milk, dairy products and certain vegetables (broccoli, Chinese cabbage, legumes), would be the best solution. However, one serving of dairy product provides only ~300 mg of calcium so those who do not consume enough calcium from food sources may need calcium supplements to meet their daily requirement. Currently, many calcium supplements are available in the market. Calcium is well absorbed in small intestines but not all calcium taken will be absorbed. The amount of calcium absorbed is dependent on a number of factors such as the acidic condition in our intestines, vitamin D level, estrogen level and the type of calcium supplement. Moreover, calcium availability in calcium supplements is affected by the nature of the calcium complex (Delisle et al., 1995). It is now generally recognized that calcium absorption does not depend solely on the amount of the calcium present in the products but also on other factors such as solubility and ionization.

Therefore, the objective of this study was to compare calcium release and ionization from commercial calcium supplements and various salts of calcium (both inorganic and organic salts) with different solubilities (Table 1). The influence of gastric pH and dietary fiber co-administered with calcium on calcium availability was also studied.

Table 1 Physical properties of different calcium salts investigated in this study (Budavari, 1996).

Calcium salts	Molecular weight	Solubility in water at 20 °C (g/100 mL)	Hygroscopicity	Flavor
Calcium gluconate	430.4	4	Stable	Bland
Calcium lactate (pentahydrate)	308.0	9	Little unstable	Neutral/bland
Calcium lactate gluconate	324.3	20	n/a	Neutral
Calcium acetate (monohydrate)	176.1	34.7	Stable	Vinegary
Calcium hydrogen phosphate (dihydrate)	172.1	0.02	Stable	Sandy/bland
Calcium carbonate	100.1	0.0015	Stable	Soapy/lemon

Materials and Methods

Materials

Two calcium sources commercially available for use in food and dietary supplements were used, i.e., inorganic and organic salts. The inorganic salts of calcium were calcium hydrogen phosphate dihydrate or dibasic calcium phosphate (Carlo Erba Reagenti, Italy), and calcium carbonate (Riedel-de Haën, Germany). The organic salts of calcium were calcium gluconate (Gluconal® CA), calcium lactate pentahydrate (Puracal® DC), calcium lactate gluconate (Puracal® XPro) (PURAC Bioquimina, Spain), calcium acetate monohydrate (Polskie Odczynniki S.A., Poland). Commercial calcium carbonate tablets (Chalkcap®-1000, Pond's Chemical Thailand R.O.P., Thailand) were used for comparison purpose. Low methoxy pectin with degree of esterification of 38% (type CU701) and

that with degree of esterification of 29% and degree of amidation of 20% (type CU020) were kindly provided by Herbstreith & Fox KG (Germany) and referred as LM pectin and LMA pectin, respectively. Commercial dietary fiber drink containing psyllium husk, apple pectin, fructo-oligosaccharide and other ingredients (Truslen® Nite Klean Fiber, Pronova Laboratories, Thailand) was used as a model dietary fiber in this study. All other chemicals were standard pharmaceutical grade or analytical grade and were used as supplied without further purification.

Preparation of calcium tablets from various calcium salts

Six calcium tablet formulations, i.e., calcium gluconate, calcium lactate, calcium lactate gluconate, calcium acetate, calcium hydrogen phosphate, and calcium carbonate, were studied. The compressed tablets (tablet weight of 1000 mg) of different calcium salts were manufactured by direct compression using a single punch hydraulic press (Model 15011, Specac, USA) with 12.7-mm diameter flat-faced tooling, at a pressure of 1 ton with a dwell time of 10 s and kept in a desiccator until used.

Determination of tablet hardness

The hardness of each tablet was tested using a tablet testing instrument (model PTB311, Pharmatest, Germany). The formulated as well as the commercial tablets were placed between the two jaws. The breaking point was determined by gradually increasing the force on the tester. Tablet hardness is the force applied (in kg) to break the tablet radially into two halves.

Measurement of calcium by calcium ion selective electrode system

For calcium measurement, the calcium ion selective electrode system (model HI4522, pH/ISE/EC bench meters, Hanna Instruments, Romania) was used. Calcium ion selective electrode has a polyvinyl chloride (PVC) membrane which is impregnated with an organic molecule which selectively binds and transports calcium ions, and contains an internal solution with a fixed concen-tration of calcium chloride – added to the potassium chloride/silver chloride solution of the internal reference system. The instrument was calibrated with standard solution supplied by the manufacturer (calcium standard 0.10 M, lot G1258, Hanna Instruments, Hungary), and confirmed the calibrations by analyzing aqueous solutions containing 0.1 mole of ionized calcium per liter.

***In-vitro* release studies**

The *in-vitro* method for evaluation of calcium availability from different calcium tablets was performed using USP dissolution apparatus II equipped with paddles which was operated at the speed of 50 rpm. Nine hundreds millilitres of either simulated gastric fluid without pepsin (SGF, pH 1.2) or pH 4.8 acetate buffer (fed state simulated gastric fluid, FSSGF), as the dissolution medium, were placed in the glass vessel, assembled the apparatus, and equilibrated the dissolution medium to 37 ^{o}C. To study the effect of co-administration of dietary fiber with calcium supplement, LM pectin tablet (1 g), LMA pectin tablet (1 g) or commercial dietary fiber drink (18 g powder containing 1 g of apple pectin and 3.4 g of psyllium husk) were added in the dissolution vessel. The amount of calcium released and ionized was measured at the suitable time interval and was then determined by calcium ion selective electrode system. Each *in-vitro* release study was performed in triplicate.

Results and Discussion

Calcium exists in nature only in combination with other substances which are called compounds. Different calcium compounds are used in supplements. These compounds contain different amounts of elemental calcium, which is the actual amount of calcium in the supplement. Figure 1 shown the percentage of elemental

calcium of the calcium salts investigated in this study. Inorganic salts, such as carbonates and phosphates, contain more calcium than the organic salts. For instance, a calcium carbonate supplement contained 40% calcium while a calcium lactate supplement only contained 13% calcium as lactate is a larger molecule than carbonate.

In this study, the tablets of different calcium salts were prepared and compared with commercial calcium carbonate tablets. Table 2 shows the hardness of tablets made of different calcium salts and commercial calcium tablets. The prepared tablets were hard enough (3-4 kg) to withstand mechanical stress during packaging and shipment. The commercial tablets had a slightly higher hardness, compared to the formulated calcium tablets.

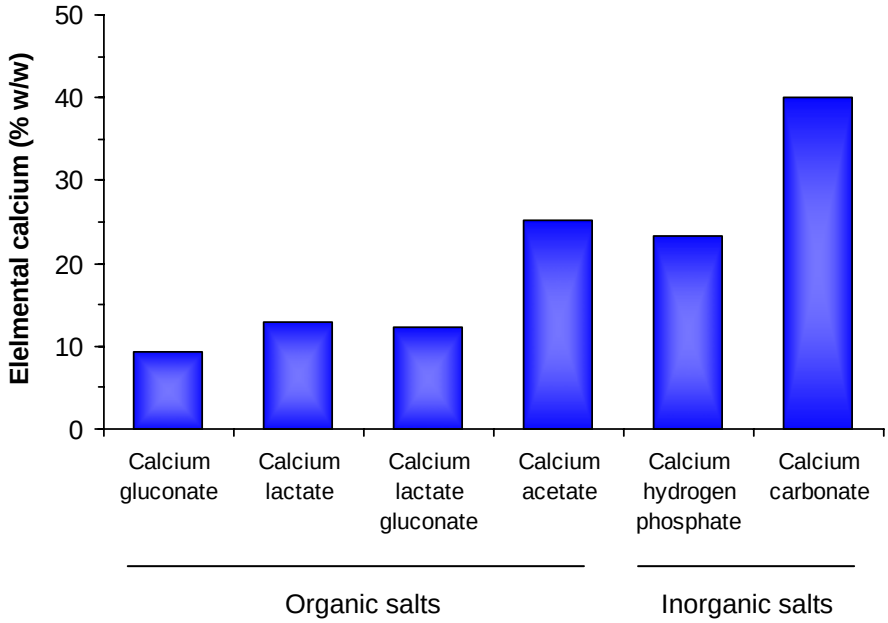
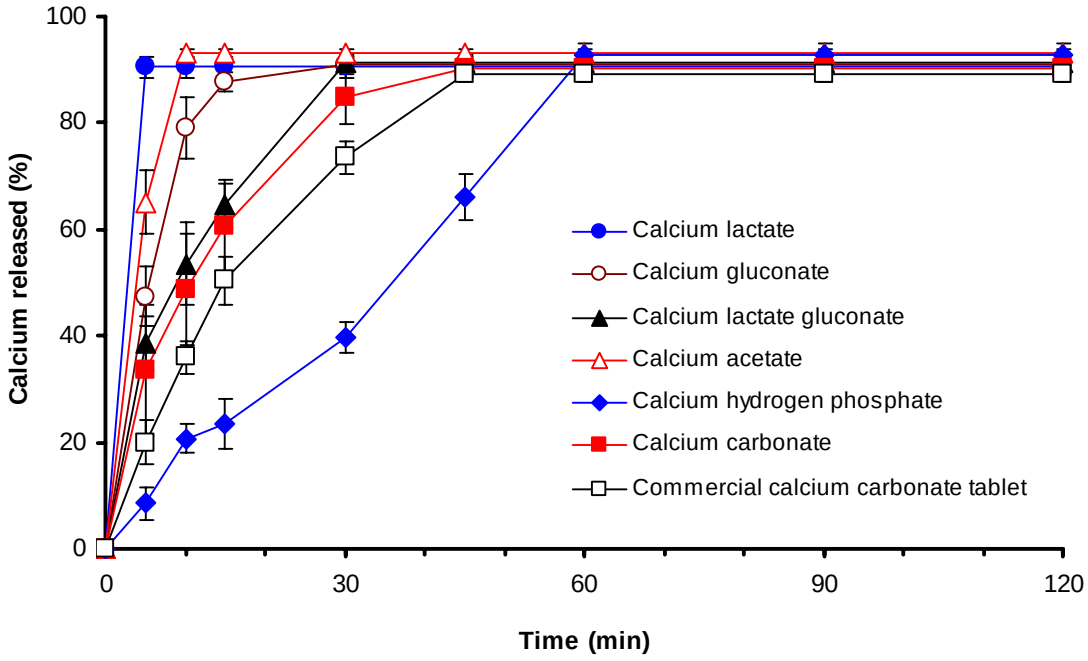


Figure 1 Comparison of the amount of calcium (elemental calcium) found in various calcium salts.

Table 2 Hardness of tablets made of different calcium salts and commercial calcium tablets.

Products	Tablet hardness (kg, n=5)
Calcium gluconate	3.6 ± 0.4
Calcium lactate	3.2 ± 0.3
Calcium lactate gluconate	3.8 ± 0.2
Calcium acetate	3.6 ± 0.1
Calcium hydrogen phosphate	3.6 ± 0.3
Calcium carbonate	3.1 ± 0.1
Commercial calcium carbonate	4.4 ± 0.2

(a)



(b)

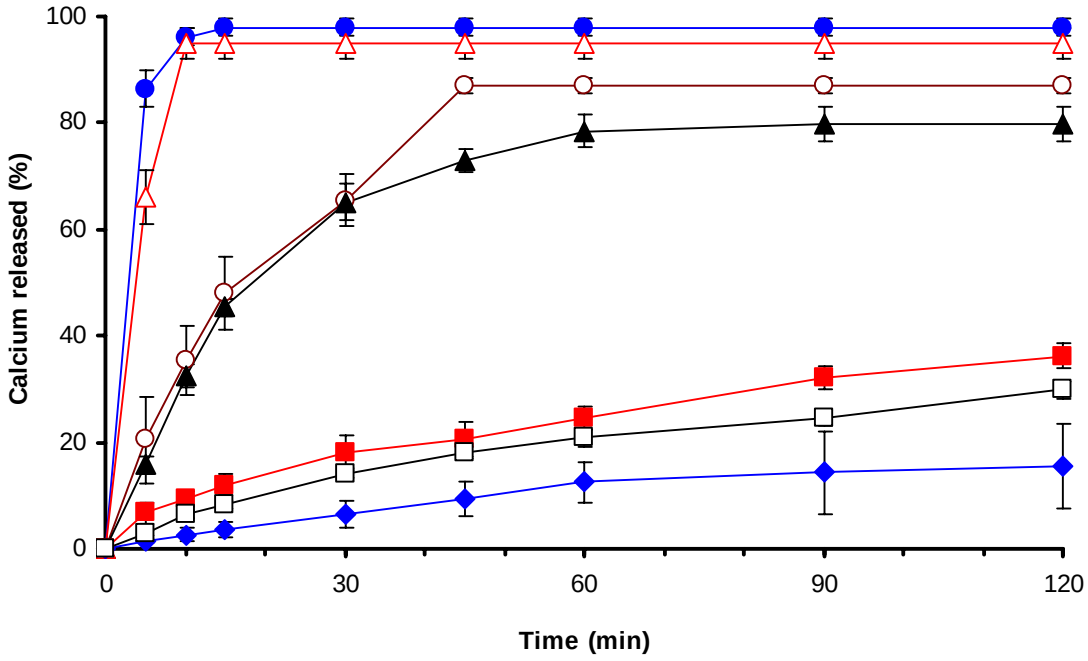


Figure 2 Release profiles of different calcium salts and commercial calcium tablets in (a) simulated gastric fluid (SGF, pH 1.2) and (b) fed state simulated gastric fluid (FSSGF, pH 4.8). The means and standard deviation of triplicate data are plotted.

The effectiveness of calcium consumption depended on its bioavailability, which means how well the human body absorbed and utilized it. On average, only about 10% to 30% of calcium was absorbed from a mixed diet by healthy adults (Gerstner, 2003). Several different factors influenced this level, including which salt provided the calcium. According to U.S. Pharmacopeia (USP) standards, a calcium tablet must dissolve and release calcium within 30-40 minutes. Figure 2 shows the release profiles of different calcium salts and commercial calcium tablets. The rate of calcium release from various formulations was compared. In SGF, most of the formulations released calcium within 1 hour. The commercial calcium carbonate tablets and the tablets containing calcium carbonate showed a slow calcium release in SGF with a complete release within 1 hour, resulting from the ionization of calcium carbonate in acidic medium. The ionization of calcium carbonate developed CO_2 in the stomach and then led adverse intestinal effects such as intestinal bloating or excess gas formation (Levenson and Bockman, 1994). The tablets containing calcium hydrogen phosphate showed the slowest calcium release (Figure 2a). This is agreed with the other report (Camara-Martos and Amaro-Lopez, 2002) in which calcium phosphate demonstrated a lower bioavailability than other calcium salts. Furthermore, calcium phosphate is not considered to be an appropriate ingredient for calcium supplement due to the undesired characteristics of the phosphate anion. Intake of phosphate is reported to exceed adult recommended dietary intakes (RDIs). Therefore, the intake of phosphate should be avoided in order to gain a higher calcium-to-phosphorus ratio, which is considered favorable for sufficient calcium absorption (Gerstner, 2003).

Tablets made of the organic salts of calcium with higher solubility, e.g., calcium lactate, calcium acetate, and calcium gluconate, demonstrated a faster calcium release than the commercial calcium carbonate tablets (Figure 2a). Tablets of calcium lactate released calcium faster than those of calcium gluconate and calcium acetate. It is likely that the elemental weight of calcium had an effect on the release and ionization of these salts. The higher the elemental weight of calcium in a compound, the less soluble the calcium is. Therefore, calcium lactate and gluconate with lower elemental weights have higher solubility, which allows for absorption at the digestive tract. Spencer et al. (1966) reported the greater *in-vivo* absorption of calcium from calcium lactate than from calcium gluconate, as evidenced by the increase in the average calcium plasma level and the decrease in average fecal calcium excretions during the intake of calcium lactate. However, both calcium lactate and calcium gluconate showed higher calcium absorption with narrow variation, compared to calcium carbonate, calcium citrate and calcium phosphate (Levenson and Bockman, 1994).

The calcium release from most formulations in gastric fluid with higher pH (FSSGF) showed a slower calcium release, except for calcium acetate and calcium lactate (Figure 2b). This indicated that bioaccessibility of calcium acetate and calcium lactate was not influenced by gastric conditions. They did not require the presence of gastric acid to dissolve and be absorbed and could be taken on an empty or full stomach. The results also suggested that other calcium salts, especially calcium carbonate and calcium hydrogen phosphate, required an acidic environment in order to be dissolved in the stomach and absorbed into the blood. Hence, it could be said that the type of calcium salts influenced the calcium availability in the different gastric conditions.

The co-administration of dietary fiber with calcium supplement can influence the calcium availability. In this study, we found that the dietary fiber (i.e., LM pectin, LMA pectin and

commercial dietary fiber drink) slowed the calcium release (Figures 3-4). The results were not obvious in SGF (Figure 3) as the fibers were less soluble. The interaction was obviously seen in the case of calcium lactate. For calcium carbonate, the co-administration of LM pectin or LMA pectin slightly increased the calcium release, compared to those without pectin. On the contrary, the co-administration of commercial dietary fiber drink with calcium carbonate decreased the calcium release. The presence of psyllium husk and fructo-oligosaccharide along with apple pectin in dietary fiber drink may increase the viscosity of SGF, thus the calcium release from the tablets was slowed down.

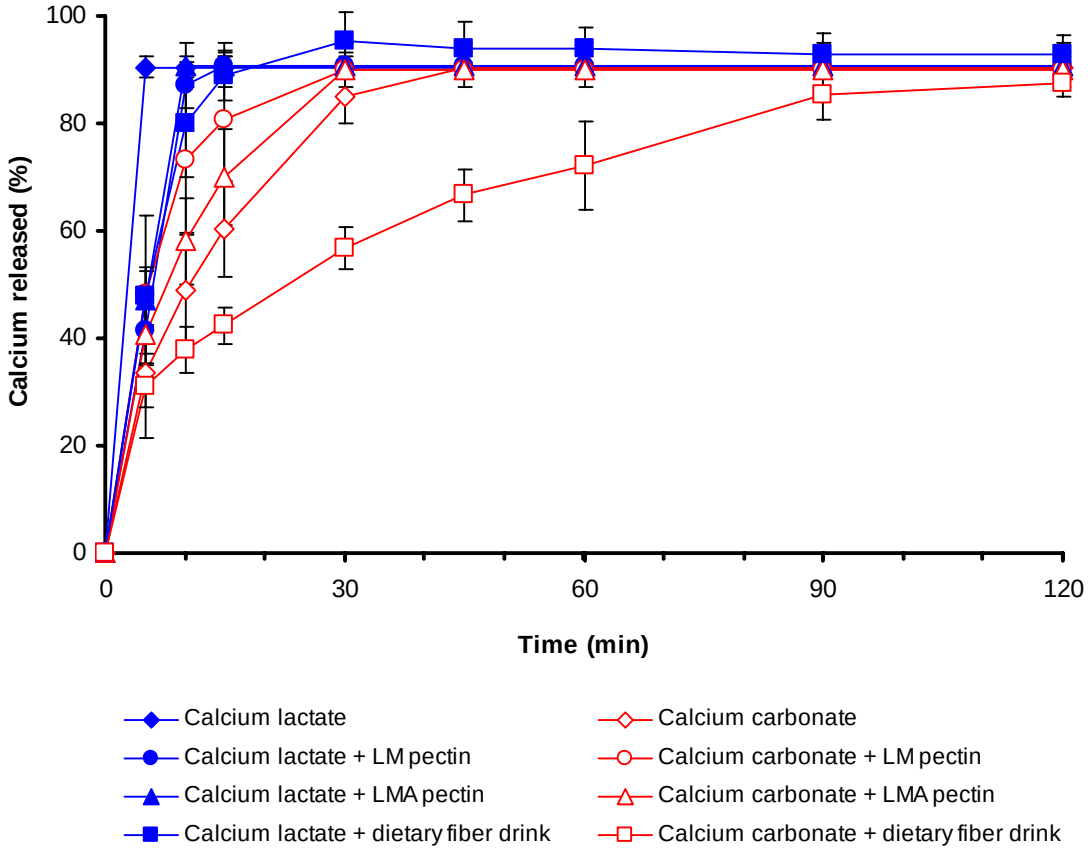


Figure 3 Effect of pectins and dietary fiber on calcium release from calcium supplement tablets in simulated gastric fluid (SGF, pH 1.2). The means and standard deviation of triplicate data are plotted.

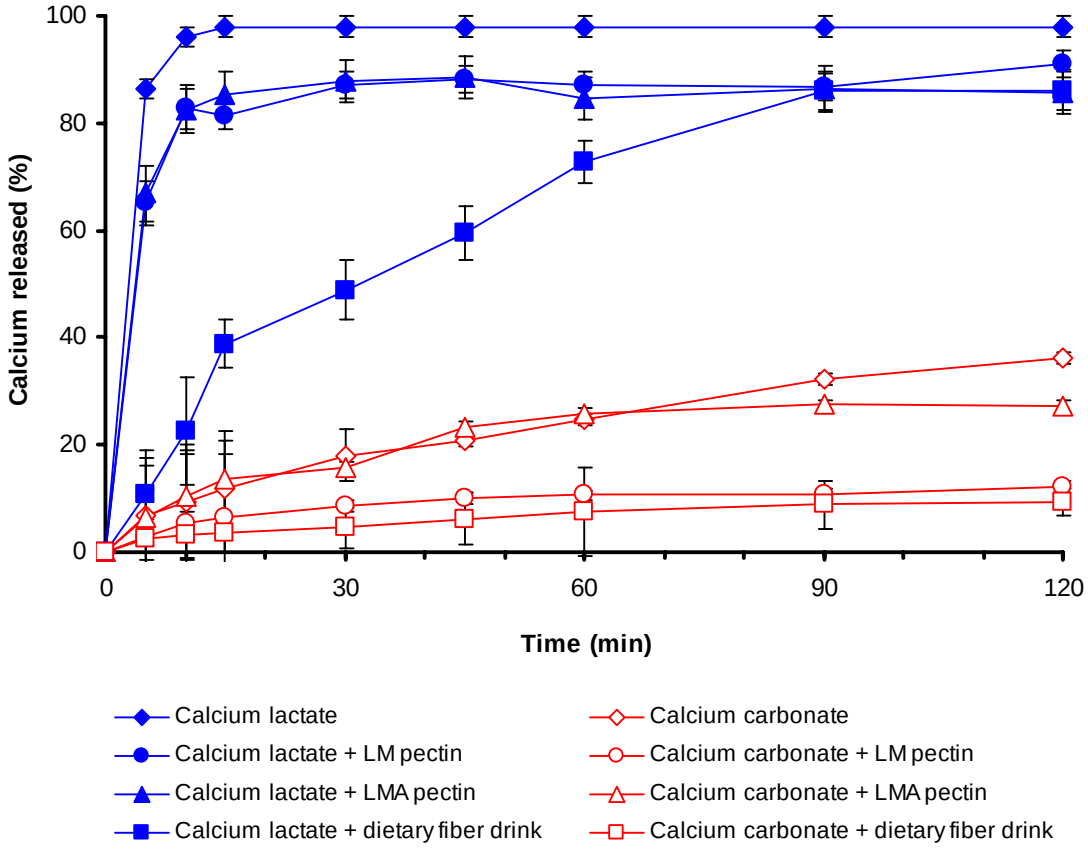


Figure 4 Effect of pectins and dietary fiber on calcium release from calcium supplement tablets in fed state simulated gastric fluid (FSSGF, pH 4.8). The means and standard deviation of triplicate data are plotted.

The profound effect was seen in medium with higher pH (FSSGF) (Figure 4). At pH 4.8, pectin can rapidly form viscous solutions and gels on contact with aqueous media (Sriamornsak et al., 2007), thus the slow calcium ionization was observed. This may be the reason why the calcium release was retarded when co-administration of calcium tablets with LM pectin or LMA pectin. Moreover, pectin with degree of esterification less than 50% can form gels by the action of calcium ions, which crosslink the galacturonic acid chains (Sriamornsak, 2003). Therefore, the free calcium, from tablets of calcium lactate and calcium carbonate, in the FSSGF medium were reduced. The co-administration of calcium tablets with commercial dietary fiber drink was clearly reduced the amount of calcium available in the FSSGF medium. The released calcium may be trapped by fibers (i.e., pectin, psyllium husk and fructo-oligosaccharide) then the free calcium was diminished. The results absolutely agreed with the *in-vivo* results performed in pre- and perimenopausal women in which a high-fiber (especially wheat bran fiber) diet could reduce calcium absorption (Wolf et al., 2000). In contrast, fructo-oligosaccharide showed to stimulate apparent calcium absorption in both the animal and human gut (Morohashi et al., 1998; Zafar et al., 2004).

Conclusion

Most of the formulations released calcium within 1 hour. Tablets made of higher solubility calcium salts demonstrated a faster calcium release. The tablets containing calcium hydrogen phosphate showed the slowest calcium release, followed by calcium carbonate, in SGF. The calcium release from calcium hydrogen phosphate and calcium carbonate tablets in FSSGF showed a slower calcium release than in SGF. The dietary fiber co-administered with a calcium supplement influenced the calcium availability, especially in FSSGF. The results suggested that most of the calcium salts could be used as calcium supplement. However, the use of calcium carbonate required an acidic environment in order to be dissolved in the gastrointestinal tract. The co-administration of calcium supplement together with dietary fiber should be cautiously used.

Acknowledgments

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