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Potential of Mussel Shell as a Biosorbent for Stormwater Treatment

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1 Executive Summary

The Auckland Regional Council's (ARC) Stormwater Action Plan (SWAP) has several work streams, one of which is to investigate innovative technologies. As part of this, ARC has been investigating the potential of using crushed shellfish shells to remove contaminants from stormwater. The ARC has undertaken a series of projects investigating the potential of New Zealand green-lipped mussel (*Perna canaliculus*) shells for the removal of dissolved metals from stormwater. This report compiles findings from that series of projects.

The first part of the work was a literature review, which found that marine shells have considerable potential for removing metals from aqueous solution. A laboratory experiment was performed in which crushed *Perna canaliculus* shell was exposed to dissolved zinc (Zn^{2+} (aq)) and copper (Cu^{2+} (aq)). The analytical results show that the mussel shell has a significant capacity for removing these metals from solution, most probably due to an exchange mechanism with calcium (Ca^{2+}) ions from $CaCO_3$. Further laboratory experiments were carried out to investigate how crushed *Perna canaliculus* shell could be implemented in stormwater filter systems. This was done by using flow-through rather than static experiments, the first of which had a long solution-solid contact time of around six hours and found that the absorption capacity was being exceeded at a loading of around 650 µm of Zn per gram of mussel shell. The second flow-through experiment decreased the solution-solid contact time to 30 mins, extended the shell particle size range and increased the amount of shell being used, each column containing 150 g of shell in contrast to 20 g initially used. The results of the second flow-through experiment demonstrated the near-equivalent relationship between zinc loading and calcium release and found that the column absorption efficiency was not compromised by the reduction in solution:solid contact time. Overall, these results also suggest that there is good potential for the development of mussel shell material for the removal of metal contamininants from stormwater.

2 Introduction

The Auckland Regional Council (ARC) has investigated the use of mussel shell as an adsorbent to remove dissolved chemical contaminants, particularly zinc (Zn) and copper (Cu) from stormwater. Existing stormwater treatment devices, such as sand filters and rain gardens, typically use peat mixed with sand to provide adsorption of dissolved metals and filtration of particulate metals. Peat has the following disadvantages; 1) it degrades over time, 2) good quality peat is a relatively expensive and scarce resource, and 3) peat is a non-renewable resource.

Mussel shell, a waste product from shellfish processing, has neither of these disadvantages. If it proved effective for metal removal from stormwater, then the shellfish processing industry would be able to provide a large amount of material at relatively low cost. Like sand, shell material can be crushed and graded for a particular use.

Finding a commercial use for mussel and oyster shells would also be of major benefit to New Zealand's shellfish industry. Currently, most shell material is dumped as waste or given away to farmers. For example, Sealord Shellfish Ltd in Nelson pay $40/tonne for removal of mussel shell waste material (pers. comm Graham Fox, Sealord) and Sanford Ltd in Marlborough leave their mussel shell waste in open pits in forests for one year before giving it away to vineyards and farmers (pers. comm Mike Mandeno, Sanford). A major source of mussel shell in the North Island is from North Island Mussel Processors Ltd (NIMPL) in Tauranga, which is co-owned by Sanford Ltd, Sealord Shellfish Ltd and Greenshell New Zealand Ltd. Mussel shell is also available within the Auckland region, for example WestPac Ltd in Henderson produce 1-3 $m^3 d^{-1}$ of crushed mussel shell. Other companies such as Sea Products Ltd in Drury also process mussels, and Kia Ora Seafoods Ltd in Manukau produce 4 $m^3 d^{-1}$ of oyster shell waste throughout the winter.

This report is presented in four main sections. Firstly, a literature review on the ability of shell material to adsorb heavy metals and the potential application of mussel shell material to enhance stormwater treatment devices. Secondly, preliminary batch absorption experiments using solutions containing Zn and Cu at similar concentrations as commonly encountered in stormwater (Craggs et al. 2006). Thirdly, flow-through experiments with solutions of Zn and Cu passed through beds of crushed *P. canaliculus* shell (500- 2000 µm with 20 g shell per column) over a 24 hour period (Pickmere and Mathieson 2007). Fourthly, further experimentation using a flow-through column methodology for Zn removal but with modifications such as, an increased flow rate, an extended range of shell particle size, and an increase in the scale of the trial, with each column containing approximately 150 g of shell (Macaskill 2008).

3 Literature review

3.1 Heavy Metal Pollutants of Stormwater

Contamination of waters all over the world by heavy metals continues to present a serious threat to the environment and human health. Many heavy metals are toxic even at low concentrations and often accumulate throughout the food chain (An et al. 2001). The main heavy metals that are found in dissolved form in stormwater are Zn, Cu and cadmium (Cd) (Minton, 2005). In New Zealand, the Zn is thought to primarily come from galvanized roofing material and vehicle tyres, while the Cu originates from vehicle brake linings.

3.2 Biosorbents

Sand filters for stormwater treatment can efficiently remove large particulates (Färm 2002) but do not adequately remove dissolved heavy metals. The development of innovative technologies to remove dissolved metals remains a challenge, as current technologies have many limitations (Wan et al. 2004). One relatively simple way to enhance the heavy metal removal performance of existing stormwater treatment filters would be to incorporate materials with a high affinity for dissolved heavy metals. Peat and compost are currently used for this purpose, but compost is frequently contaminated with metals to the extent that it becomes a source of metals rather than an adsorbent. Peat degrades over time and good quality peat is a relatively expensive and scarce resource.

Many natural and industrial waste materials have potential for use as biosorptive material including arthropod (crab and prawn) exoskeletons, crustacean and mollusc shells and plant-derived materials (wood, bark leaves, stalks, nut shells, grasses and seaweed) containing lignocellulose (Macchi et al. 1986; Chen and Chang 1994; Kratochvil and Volesky 1998; Bailey et al. 1999; Han 1999; An et al. 2001; Kim and Park 2001; Wojtenko and Ray 2001; Färm 2002; Johnson et al. 2003; Basso et al. 2004; Lee et al. 2004; Tobiason 2004; Lodeiro et al. 2005; Schiewer 2005; Vijayaraghavan et al. 2005; Genç-Fuhrman et al. 2007; Park et al. 2007; Fu and Wang 2011).

Several reviews of heavy metal uptake by biosorbent materials can be found in the literature (Bailey et al. 1999; Vegliò and Beolchini 1997; Wase and Forster 1997; Volesky 2003). Biosorbent materials often have many beneficial features as sorptive media including metal selectivity, lack of concentration dependence, tolerance for organics, and potential for regeneration (Brierley 1990; Volesky 1990; An et al. 2001). These properties illustrate the potential to incorporate biosorbents into existing stormwater treatment filters (An et al. 2001), provided the capacity shows that such addition is effective. The mechanisms of metal capture from solution vary in different materials. For example, bark is effective because of its generally high tannin content containing polyhydroxy polyphenol groups which actively chelate metal cations by displacing some of the phenolic hydroxyl groups (Randall et al. 1974). In contrast, the absorption properties of zeolite are a result of the complex structure of this microporous mineral which contains many negatively charged sites which hold positively charged ions, such as sodium, calcium, and potassium, which are readily replaced by heavy metal ions (Leppert 1990).

3.3 Shell Material

The literature review initially focuses on chitin, a specific biopolymer component of shell. Both chitin and its derivative chitosan have been found to have very high sorption capacities for heavy metals (Chen and Chang 1994; Bailey et al. 1999; An et al. 2001; Rae and Gibb 2003; Schiewer 2005; Vijayaraghavan et al. 2005). The whole shell material is also covered by the review, because recent studies have indicated that an alternative mechanism to adsorption may be responsible for high removal rates of dissolved metals. While the review encompasses shell material in general, the analysis of the review is directed towards mussel shell in particular.

3.3.1 Shell Composition and Derivatives

The shells of molluscs (e.g., oyster, clam and mussel) and crustaceans (e.g., lobster, crab and shrimp) are composed of a mixture of calcium carbonate, protein and chitin, with relatively small amounts of lipid, phosphate and pigment (Wase and Forster, 1997; Zuo et al. 2001). As an example, crab shell is typically composed of 30% protein, 41% calcium carbonate, 27% chitin and 2% lipid (No and Meyers, 1995). There is considerable variation between crustaceans and molluscs in the relative amounts of these substances (for example, see chitin content data in Table 1).

Chitin is similar structurally to cellulose except it has an acetylglucosamine instead of a hydroxyl unit. It has a molecular weight of over one million Daltons (Bornet and Teissedre, 2005). Specifically, chitin is N-acetylated aminopolysaccharide (an unbranched polymer of (1-4) N-acetyl D-glucosamine) (Chen and Chang 1994; Zuo et al. 2001). Chitin and its derivatives are used for a variety of applications because of their versatile biological and chemical activities. For example, deacetylated chitin oligomers act as hydrating agents in cosmetic and hair care products, and as floculating and clarification agents in the food industry. In agriculture, chitin derivatives have been used as fertilisers and fungicides.

Chitin can be isolated from shell material by decalcification using acid (e.g., HCl) (Coughlin et al. 1990).

The chitin content of different shell materials varies appreciably (Table 1). No specific reference could be found for mussel shell. Falini and Fermani (2004) noted that the nacre of mussel shell was high in chitin, whereas the crossed lamellar structure of the shell matrix was low in chitin. Sim-Smith (pers. comm) reported that the green-lipped mussel shell was about 96% inorganic and, as Giffinet and Jeuniaux (1979) reported that the organic fraction of mollusc shells contained at most 25% chitin, it is suggested that green-lipped mussel shells will comprise at most 1 % chitin. Therefore, if sorption within the chitin structure was the main mechanism for heavy metal removal, one might expect mussel shell to have a relatively low capacity for heavy metal removal, compared with shell from crustaceans such as crab, lobster and shrimp.

Table 1.

Chitin content of various shell materials.

Shell material	% Chitin	Reference
Crab shell	27	No and Meyers (1997)
Crawfish	23.5	No et al. (1989)
Shrimp	22.6	Ferrer et al. (1996)
Molluscs	0.1-1	Goffinet and Jeuniaux (1979)
Green-lipped mussel	Max of 1	Estimated from Sim-Smith (pers.comm) who determined that mussel shells had maximum of 3.5% organic matter

Chitin can be transformed to chitosan, a linear biopolymer of D-glucosamine (polyglucosamine), by de-acetylation using concentrated alkali (e.g., 40% sodium hydroxide) at high temperature (Coughlin et al. 1990; Bornet and Teissedre 2005). Chitosan is purported to have even greater capacity than chitin to adsorb heavy metals. Typically, chitosan is not a single substance but a group of products that have been de-acetylated to various extents (Zuo et al. 2001).

3.3.2 Heavy Metal Uptake by Chitin

Zuo et al. (2001) reviewed the potential uses of chitin and chitosan, including removal of heavy metals, pesticides, PCB and dyes. Wantanaphong et al. (2005) found that chitin could remove more than 70% of the metals from a solution of synthetic groundwater containing 10 mg l^{-1} Cu, Pb and Zn and 1 mg l^{-1} Cd.

Crude (without removal of protein) shrimp chitin adsorbed over 95% and 96% of Cu and Cr respectively from a solution with 20-100 mg l^{-1} concentrations, which was comparable to the removal by crab chitosan (Chui et al. 1996). Nickel was not so easily removed by the shrimp chitin (44-70% removal efficiency) compared to 82-99% removal by the crab chitosan.

Adsorbed heavy metals can easily be eluted from chitin. Recovery efficiencies of Cu, Cr and Ni from crude shrimp chitin using 0.1 M EDTA were similar to than those of crab chitosan (80-100%) (Chui et al. 1996).

3.3.3 Heavy Metal Uptake by Chitosan

Chitosan is a natural polycationic polymer that is a more efficient chelator of heavy metals and adsorber of organic compounds than other polymers such as synthetic resins, activated charcoal and chitin (Deshpande 1986; Gandhi 1997; No and Meyers 2000; Evans et al. 2002; Wan et al. 2004).

The high affinity for heavy metal ions is due to its high content of free amine groups that are exposed in chitosan when chitin is deacetylated (Bornet and Teissedre 2005). Chitosan has been shown to chelate five to six times greater amounts of heavy metals than chitin (Nomanbhay and Palanisamy 2005).

For example, the sorption capacity of chitosan for Cu ions from aqueous solution has been found to be 40-200 mg·per gram of chitosan (Findon et al. 1993; Schmuhl et al. 2001).

Heavy metal removal by chitosan varies for particular metal ions, ionic strength and metal ion to chitosan ratio (Vold et al. (2003), but has been shown to be independent of chitosan particle size (for the range 0.65 - 3.38 mm diameter; 300 to 540 pore diameters) indicating that adsorption takes place largely in the pore space (Evans et al. 2002).

Heavy metal uptake by chitosan is not directly proportional to the number of free amine groups present, but rather the conformation of the polymer, thus, chitosan may be able to be manipulated to improve or make metal uptake more selective (Park 1986; Nomanbhay and Palanisamy 2005). The amine functional group in chitosan can be altered chemically to produce chitosan derivatives exhibiting different physicochemical characteristics, i.e., molecular weight, crystallinity, deacetylation, particle size and hydrophilicity (No and Meyers 2000).

The practical use of chitosan is limited due to the relatively high costs of constructing a treatment system for chitosan alone (Wan et al. 2004). Chitosan is soft and has a tendency to agglomerate or form a gel in aqueous solutions, therefore, it is necessary to provide physical support and increase the accessibility of the functional groups for metal binding (Nomanbhay and Palanisamy 2005). One option is to coat sand with chitosan (Wan et al. 2004), although the economics and practicalities of achieving this have yet to be determined.

Crustacean shell such as crab or shrimp can be partially converted to chitosan by mild deacetylation of the periphery of the shell surface producing a heavy metal sorbent with similar adsorption, elution and metal recovery performance to pure chitosan (Coughlin et al. 1990; Chu 2002; Pradhan et al. 2005).

3.3.4 Heavy Metal Uptake by Raw Shell Material

Several authors have found that comminuted, ground or powdered shell material has very high heavy metal sorption capacities, which are similar to, or can even exceed those of chitin, chitosan and other sorbent materials (Tudor 1999; Kim and Park 2001; Petrisor et al. 2002; Rae and Gibb2003; Tudor et al. 2006; Köhler et al. 2006; Ok et al. 2010; Yan-jiao 2011). Shell material has also been used to remove dye (Figueiredo, et al. 2005) and reclaim waste lubricating oils (Osakwe and Maduako 2004).

Hung and Li-Ming Han (1977) investigated the heavy metal (Hg, Cu, Cd and Zn) adsorption capacity and ion selectivity of powdered shell from several shrimp (*Penaeus japonicus, Parapenaeopsis hardwickii, Metapenaeus monoceros* and *Parapenaeus fissurus*) and crab (*Portunidae tribercultatus*) species. They found that shell material had similar performance to pure chitin and chitosan, adsorbing 90% of the heavy metals within 15 minutes.

Lee et al. (1997) determined that crab shell particles could remove 1300 mg Pb g^{-1} shell material, while An et al. (2001) found that crab shell had higher heavy metal (Pb, Cd, Cu, Cr) uptake compared to cation exchange resin, zeolite, powdered activated carbon and granular activated carbon. They also found that the removal of heavy metals was selective, with Pb and Cr being removed in preference to Cd and Cu. Kim and Park (2001) measured higher Pb removal by crab shell than with either chitin or chitosan and similarly, Rae and Gibb (2003) showed that ground crab shell removed >90% of Cu from solution compared to chitin (~35%).

The above findings indicate that there is another component within shell material that is providing superior metal removal to chitin.

This was confirmed in studies by Tudor et al. (2006) who compared the metal (Pb, Cd, Cu, Cr, Hg, As, Zn) uptake ability of several shell materials including molluscs (Quahog clam (*Mercenaria mercenaria*), Eastern oyster (*Crassostrea virginica*)) and crustaceans (American lobster, *Homarus americanus*) to that of chitosan and other calcium carbonate materials (calcite, aragonite and limestone) and showed that shell is usually superior, in some cases by orders of magnitude, in terms of metal uptake rate and total absorption capacity. An example of the effectiveness of heavy metal uptake by shell material is the reduction of Pb concentration from 30,000 ppm to less than 0.1 ppm in 5 minutes by comminuted clam shell (Tudor et al. 2006). All the shell materials tested displayed very fast kinetics and were able to extract more than their own weight of metal (Pb) within a few minutes (Tudor et al. 2006). Oyster shell extracted 150% of its own weight of Pb from a 30,000 ppm solution in 24 hours and ultimately, both clam and oyster shells are able to extract almost twice their own weight of Pb (Tudor et al. 2006).

In an earlier publication, Tudor (1999) suggested that the effectiveness of shell material is not just due to the chitin and mineral $CaCO_3$ content, but that the sub-micron carbonate structure of the shell material provides an extensive surface area embedded within a complex, multi-component organic matrix. This makes the shell material highly reactive, as the initial rate of metal extraction is a function of surface area.

An alternative mechanism to adsorption onto chitin is that metal uptake occurs through ion exchange and conversion of calcium and magnesium carbonates to mixed-metal carbonates in the inorganic fraction of the shell structure. However, some authors consider that chitin is indirectly associated with metal removal through adsorption of these metal carbonates to shell particles. Whether or not chitin is implicated, the formation of metal carbonates should be accompanied by the release of calcium ions (and other constituent cations) from the shell matrix (Lee et al. 1997; Kim and Park 2001). Kim and Park (2001) confirmed that K^+, Na^+, Mg^{2+} and Ca^{2+} were released from crab shell in association with Pb^{2+} uptake to the crab shell suggesting that the mechanism of uptake was through ion exchange and the formation of mixed-metal carbonates.

Dissolved heavy metal removal by shell material has been shown to be dependent upon several variables including pH, temperature, contact time, particle size, metal concentration, metal type and the physio-chemical characteristics of the material (Tudor 1999; Kim and Park 2001; Rae and Gibb 2003). For example, heat treatment (>750 °C) of bivalve shells removes the organic component and converts the mineral component of the shell, calcium carbonate ($CaCO_3$), to calcium oxide (CaO) (Currie et al. 2007, Ok et al. 2010). This more chemically active form of calcium is more effective at capturing and retaining phosphates and some metal ions under certain conditions, such as elevated pH (Kwon et al. 2004, Lee et al. 2005, Currie et al. 2007).

Previous research has indicated that there are considerable differences in the abilities of the shells of different species of bivalves to adsorb metals from solution. Kwon et al. (2004), Lee et al. (2005), and Namasivayam et al. (2005) report on the effectiveness of oyster shell for removing phosphates from solution, without naming the species of oyster they used for their research. Given the research location, it is most likely they used the Pacific oyster, *Crassostrea gigas*, which is also widely farmed in New Zealand. Currie et al. (2007) reported on the effectiveness of the shell material from this same oyster species and the New Zealand green-lipped mussel, *Perna canaliculus*, for removing phosphates from solution.

3.3.5 Regeneration

If metal is adsorbed onto shell, or shell components, either directly or indirectly, then it may be possible and desirable to elute the metals periodically, and so regenerate the ability of the material to adsorb more metal ions. A number of authors have reported that regeneration is possible for both shell material (Vijayaraghavan et al. 2005) and chitosan (Wan et al. 2004; Wan Ngah et al. 2004) by elution with aqueous ethylenediaminetetraacetic acid (EDTA). If the principal mechanism for metal removal is carbonate formation, then EDTA might not be expected to be so effective in eluting the metals.

3.3.6 Modelling

A number of authors have attempted to model their experimental results using equilibrium adsorption isotherms that describe the relationship between the amounts of adsorbed and dissolved species at a given temperature. As is common practice for experimental scientists working with solid-liquid systems, most authors have used the adsorption equations originally developed by Freundlich or Langmuir to describe gas adsorption by solids. The Freundlich equation is considered purely empirical in nature, whereas the Langmuir equation has the major advantage that it is possible to calculate an adsorption maximum and a relative binding energy for adsorption.

Examples were found within the literature of heavy metal adsorption to shell material that has given good fit to either the Freundlich model (e.g., Vijayaraghavan et al. 2005), Langmuir model (Sag and Aktay 2002), both the Freundlich and Langmuir models (Schmuhl et al. 2001; Evans et al. 2002; Taboada et al. 2003; Wan et al. 2004; Wan Ngah et al. 2004; Rojas et al. 2005), and to a hybrid Langmuir-Freundlich models with either pH-dependent or pH-independent parameters (e.g., Cu adsorption on crab shell; Chu and Hashim 2003). Without debating the merits of any of these alternative approaches it is important to point out that the tenuous theoretical basis for either equation in a liquid-solid system only applies if adsorption is the sole metal removal mechanism. If precipitation of carbonates is an important mechanism as suggested by Tudor et al. (2006) then the equilibrium adsorption isotherm approach has no theoretical basis. Carbonate precipitation may explain why Pradhan et al. (2005) did not find a fit with either the Langmuir or Freundlich models for Ni sorption by deacetylated crab shell.

4 Batch experiments

4.1 Methods

Freshly-shucked mussel shells were obtained from a commercial source in Auckland. The shells were placed on an estuarine mudflat, within the inter-tidal zone, for three weeks until the remaining flesh on the shells had been removed. The shells were then washed with tap water and crushed. A size fraction of 500-2000 μm was obtained by dry-sieving. This fraction was washed thoroughly with distilled water on a 500 μm sieve.

Static sorption experiments were carried out in acid-washed 50 ml plastic centrifuge tubes. Stock solutions of Zn (10,000 mg l^{-1}) and Cu (1000 mg l^{-1}) were made with the corresponding nitrate compound. The accuracies of the stock solutions were assessed by creating by dilution, and analysing [Zn] test (theoretically 50 mg l^{-1}) and [Cu] test (theoretically 10 mg l^{-1}) solutions, which contained no shell material.

Aliquots of 500 mg of 500-2000 μm shell material was tested in triplicate using 40 ml solutions with initial metal concentrations of 50, 100, 500 and 1000 mg l^{-1} (2, 4, 20, 40 mg total Zn) and 10, 25, 50 and 100 mg l^{-1} (0.4, 1, 2, 4 mg total Cu) for Zn^{2+} and Cu^{2+} respectively. A 20 ml sub-sample of each solution was removed after 2 days and tested for pH, [Ca], [Zn] and [Cu]. Identical analyses were carried out on the remaining 20 ml after 9 days. All solutions were filtered (0.45 μm) prior to analysis. A blank (40 ml distilled water) and shell blank (500 mg of shell in 40 ml distilled water) were also analysed. All analyses were performed by Hills Laboratories Ltd; the full data sets for day 2 and day 9 are displayed in Appendix 1, and the detailed results are reported in Craggs et al. (2006).

4.2 Results and discussion

4.2.1 pH changes

The pH data are displayed in Table 2 (mean and standard error) and Figures 1 & 2. Predictably, the pH increased between day 2 and day 9 for all solutions in contact with $CaCO_3$ shell material, but was typically lower in the solutions with higher metal loadings.

Table 2.

pH data for solutions after 2 and 9 days.

Metal Soln.	Conc. (mg l^{-1})	Mean pH @2 days	S.E.	Mean pH @ 9 days	S.E.
Zn	50	6.1	0.4	7.3	0.1
Zn	100	6.8	0.0	7.6	0.0
Zn	500	6.8	0.0	7.0	0.0
Zn	1000	6.6	0.0	6.8	0.0
Shell blank	0	6.2	-	7.3	-
Cu	10	6.6	0.0	7.3	0.1
Cu	25	6.4	0.0	7.0	0.0
Cu	50	6.2	0.0	6.8	0.0
Cu	100	6.1	0.0	6.6	0.0

Figure 1.

Relationship between pH and concentration of Zn solutions in contact with crushed mussel shell.

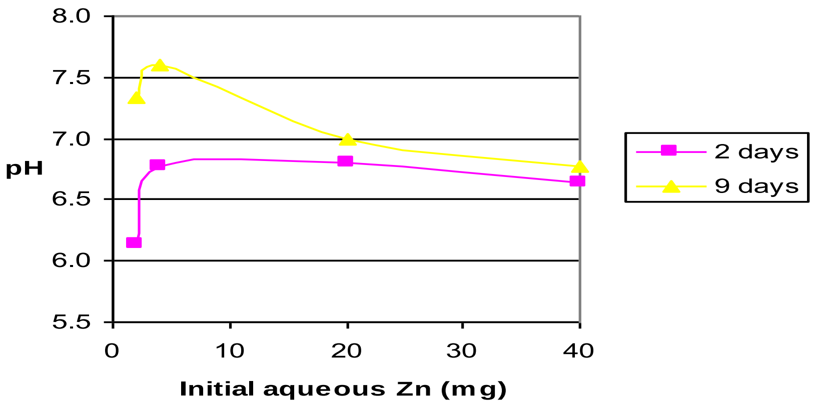
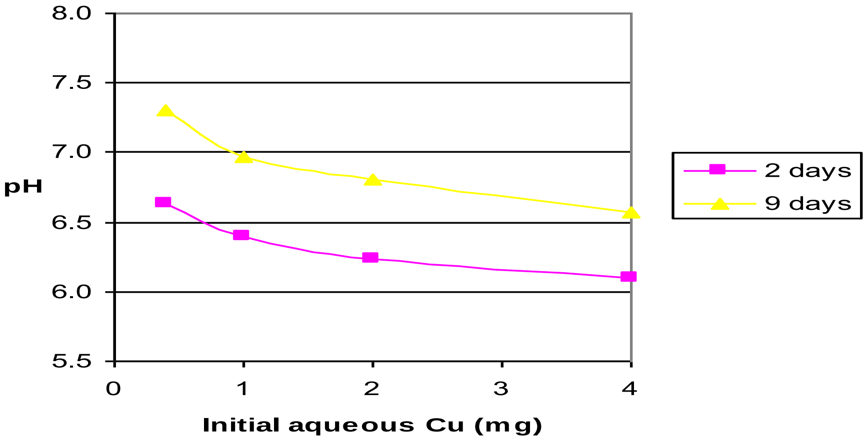


Figure 2.

Relationship between pH and concentration of Cu solutions in contact with crushed mussel shell.



4.2.2 Phase changes of Zn, Cu and Ca

The amount (mg) of dissolved Zn^{2+} (aq) and Cu^{2+} (aq) removed from the various test solutions are displayed in Figures 3 and 4 respectively. Figure 3 shows that 500 mg of 500-2000 µm mussel shell was able to remove considerable quantities of Zn from solution. For example, after 2 days, from an available total of 40 mg of Zn, 8 mg was retained by the mussel shell.

Note that half the remaining aqueous metal was removed at day 2, which (at least partially) explains the slope reduction in each plot after day 2 (Figures 3 and 4).

The plots in Figures 3 and 4 indicate that 500-2000 µm mussel shell material was not as effective at retaining Cu^{2+} (aq), compared with Zn^{2+} (aq). Initially (after 2 days) the shell was particularly ineffective when exposed to the two highest Cu^{2+} (aq) concentration solutions. This is difficult to explain, but the relatively low pH observed in these solutions could be an important factor. However, after 9 days significant Cu was observed to be retained, up to a maximum of 1.5 mg of Cu per gram of shell (Figure 4).

Figure 3.

Aqueous Zn (mg) removed by 500 mg of mussel shell (500–2000 µm) from 40 ml test solutions with initial Zn loadings of 2, 4, 20 and 40 mg.

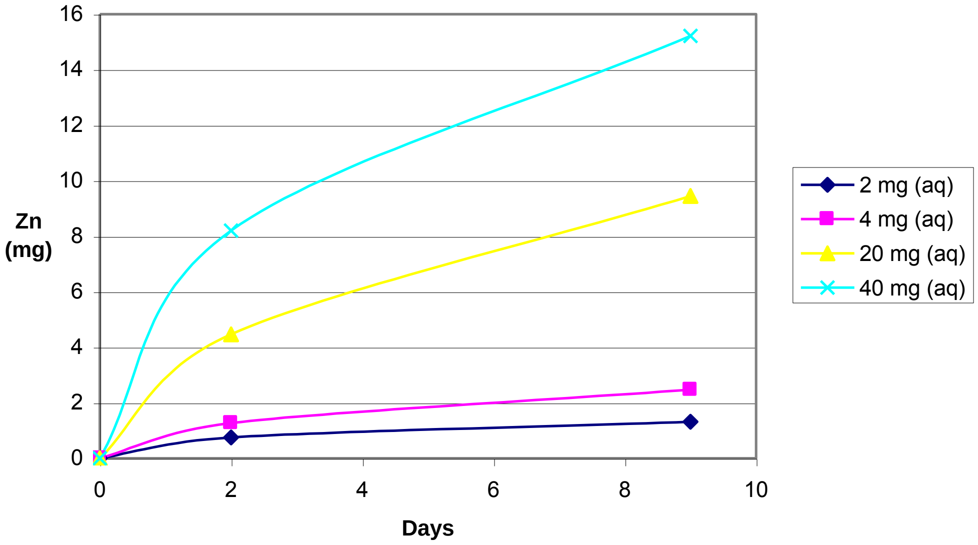
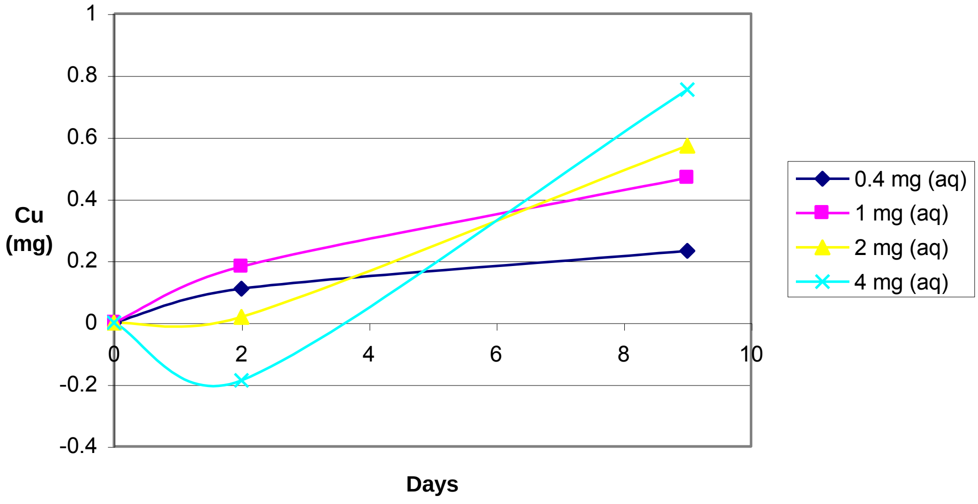


Figure 4.

Aqueous Cu (mg) removed by 500 mg of mussel shell (500–2000 µm) from 40 ml test solutions with initial Cu loadings of 0.4, 1, 2 and 4 mg.



Significant concentrations of Ca were observed in solution after shell material was placed in contact with the Zn^{2+} (aq) or Cu^{2+} (aq) solutions (see Appendix 1 for analytical data). In Figures 5 and 6, for Zn and Cu respectively, the total Ca (µMol) released into solution during the 9-day course of the experiment is compared with the total (µMol) metal (Zn or Cu) removed from the solution. The results indicate the removal of Zn^{2+} (aq) and Cu^{2+} (aq) involved an exchange mechanism between Zn^{2+} (aq) or Cu^{2+} (aq) and the Ca^{2+} from the $CaCO_3$ in the mussel shell matrix. Furthermore, the 500-2000 µm shell material turned green upon contact with the Cu^{2+} solutions, probably due to the precipitation of $CuCO_3$.

Figure 5.

Comparison of Zn removal from solution with release of Ca into solution.

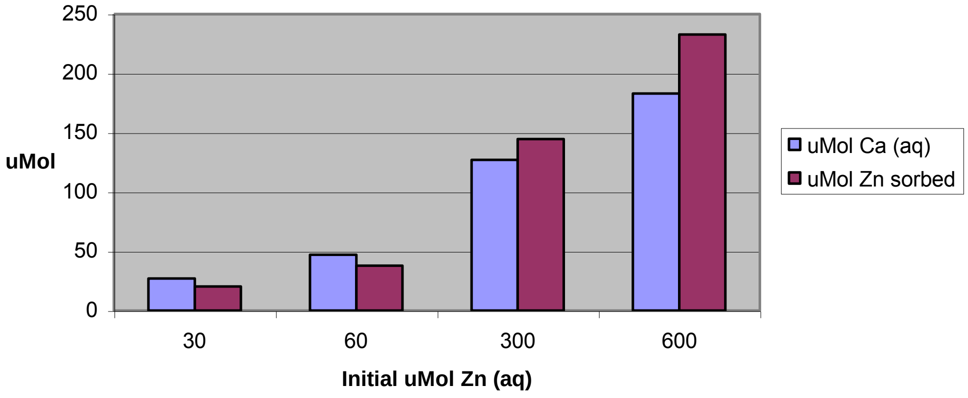
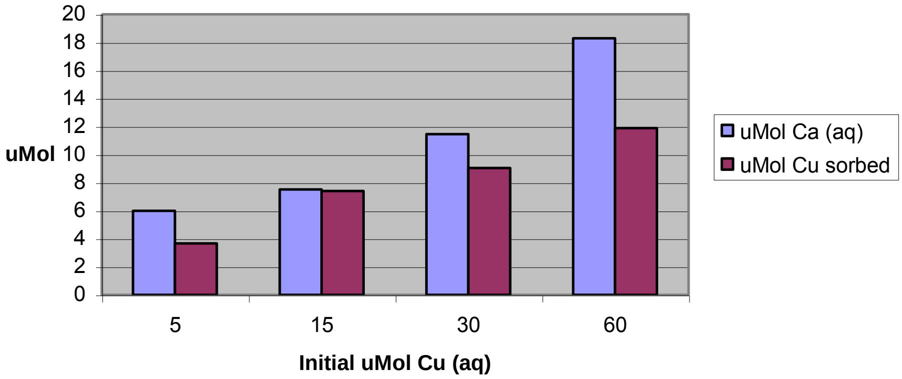


Figure 6.

Comparison of Cu removal from solution with Ca release into solution.



These results give an indication of the potential of 500-2000 μm shell material from green-lipped mussels (*Perna canaliculus*) to remove Zn^{2+} or Cu^{2+} from aqueous solution. The contact of aqueous Zn^{2+} or Cu^{2+} ions with crushed shell probably resulted in the formation of solid-phase $ZnCO_3$ and $CuCO_3$. Although the retention of Zn^{2+} (aq) was more effective, compared with Cu^{2+} (aq), the results indicate the crushed mussel shell has the potential to significantly reduce the aqueous concentration of both metals in stormwater.

Based on these results further work was recommended to investigate the potential for the use of crushed *Perna canaliculus* shell in stormwater filter systems. The shell material should be presented to the metal-contaminated stormwater in an effective way to promote the sorption process. Furthermore, when significant quantities of Zn^{2+} or Cu^{2+} have precipitated, the design should permit a convenient procedure for the recovery of metal-enriched material and simultaneous regeneration of an active filter system.

5 Column experiment one

5.1 Methods

While the preliminary experiments demonstrated the ability of mussel shell to adsorb metals from solution, it is an unlikely scenario that stormwater containing metals would be held in static conditions with shell material for nine days. Therefore, further experiments were undertaken to assess the removal of Zn and Cu from solutions passed through a column of crushed mussel shell (Pickmere and Mathieson 2007).

Four borosilicate glass columns, 20 mm in diameter, were set up with a 50 mm depth of mussel shell crushed and sieved to a particle size range of 500-2000 µm (approximately 20 g shell weight). One of the four columns was used to run a control treatment which consisted of distilled water as the test solution, whilst the remaining three columns were used as replicate columns for test solutions of metal cations.

Four experiments were run with the columns, without the columns being cleaned or the shell material replaced between experiments. All four experiments consisted of adding fresh volumes of a known concentration of metals in solution three times in succession to each of three replicate test columns, and concurrently an equivalent volume of distilled water to the control column. Each addition of the test solution, or distilled water for the control, was allowed to pass very slowly through the mussel shell filter over the subsequent 24 hours.

The four experiments used a range of concentrations of dissolved Zn and Cu at the higher end, and exceeding, what has previously been found for stormwater run-off from New Zealand roads (Moores 2009). The following solutions were used:

(1) 30 ml of 0.5 µg ml^{-1} of Zn and 0.1 µg ml^{-1} of Cu

(2) 50 ml of 5 µg ml^{-1} of Zn and 1 µg ml^{-1} of Cu

(3) 50 ml of 20 µg ml^{-1} Zn and 2 µg ml^{-1} Cu

(4) 50 ml of 100 µg ml^{-1} Zn and 20 µg ml^{-1} Cu

(Note:The test solution volume was increased from 30 ml to 50 ml after experiment 1 to provide larger samples for chemical analyses).

For each experiment, the eluent from each column was filtered (0.45 mm) and retained for chemical analyses of total Ca, Zn and Cu, along with a sample of the original stock solution that was taken prior to the solution passing through the columns. All analyses were performed by Hill Laboratories Ltd in Hamilton.

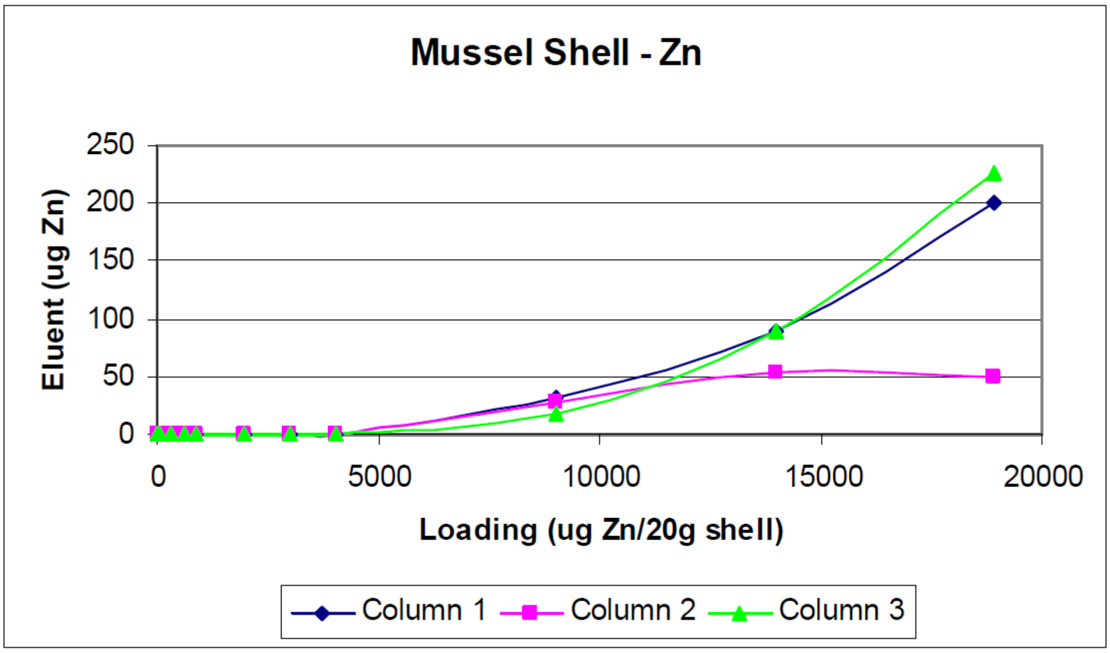
The data sets are displayed in Appendix 2 and reported in more detail in Pickmere and Mathieson (2007). Progressive Zn and Cu loadings (µg) on the shell columns were calculated and are displayed, respectively, in Figures 7 and 8. Figure 9 illustrates the correlation between the weight of Ca leached from the shell and the total weight of Zn and Cu sequestered by the shell.

5.2 Results and discussion

Insignificant amounts of dissolved Zn were found in the eluent for the control using distilled water indicating that any Zn arriving in the eluent in the experiments using metal ion solutions were not derived from the mussel shells.

Figure 7.

Comparison of cumulative loading of Zn on shell and Zn observed in eluent after additions of combined Zn and Cu solutions.



The columns effectively removed almost 100% of the dissolved Zn during the first three experiments. During experiment four, increasing amounts of Zn passed through the shell material for each subsequent 50 ml of solution passed through at 24, 48 and 72 hours. However, the shell material was continuing to retain a significant but decreasing proportion of the dissolved Zn. For example, in third run of the fourth experiment 96.8± 1.3% of the Zn from the solutions were retained on average across the three columns.

At the end of experiment three the 20 g of mussel shell material had successfully absorbed an estimated total of 4,044.5 ± 2.4 µg of Zn. Whilst at the end of experiment four the 20 g of mussel shell material had successfully absorbed an estimated total of 18,675.3 ± 78.2 µg of dissolved Zn. Experiment four, loaded a solution containing 100 µg ml^{-1} Zn and 20 µg ml^{-1} Cu into the experimental columns three times (Figure 7). In comparison, aqueous Zn and Cu in Auckland stormwater are at much lower concentrations, i.e., approximately 0.2 µg ml^{-1} and 0.05 µg ml^{-1} respectively.

Insignificant amounts of dissolved Cu were found in the eluent for the control using distilled water indicating that any Cu arriving in the eluent for experiments using the metal ion solutions were not derived from the mussel shells.

With the addition of solutions containing dissolved Cu, the columns effectively removed almost 100% of the Cu from the solutions during the four experiments (Figure 8). For example, in the third and final run of the fourth experiment, on average across the three columns 99.93 ± 0.06% of the Cu from the solutions was retained by the shell material.

At the end of experiment four the 20 g of mussel shell material had successfully absorbed an estimated total of 3,574.8 ± 0.5 µg of dissolved copper.

Figure 8.

Comparison of cumulative loading of Cu on shell and Cu observed in eluent after additions of combined Zn and Cu solutions.

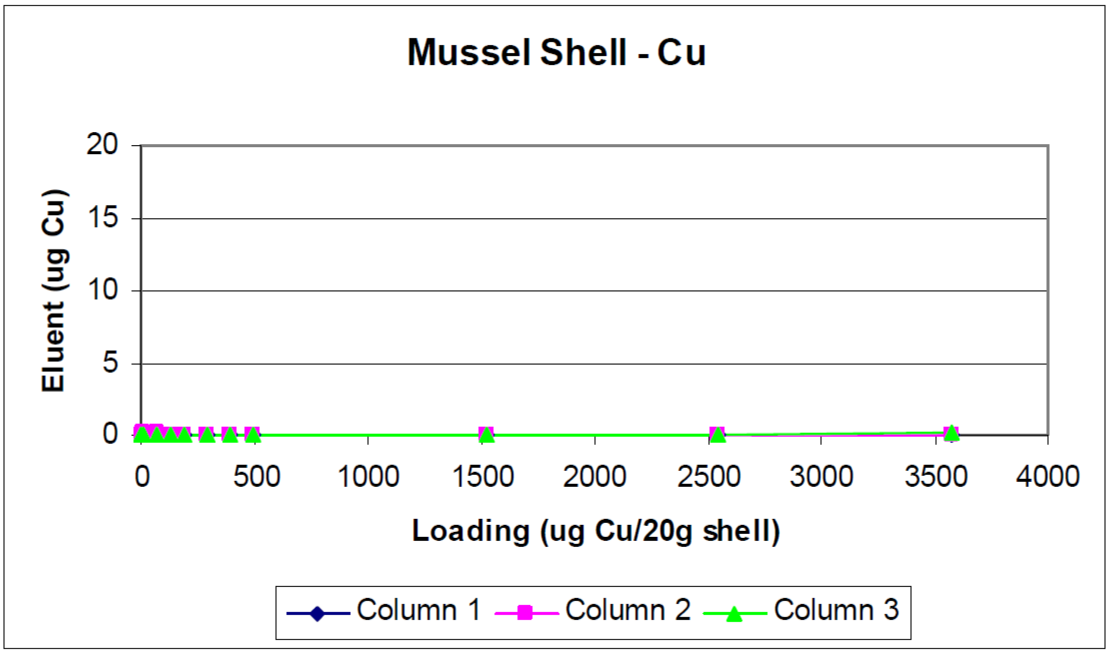
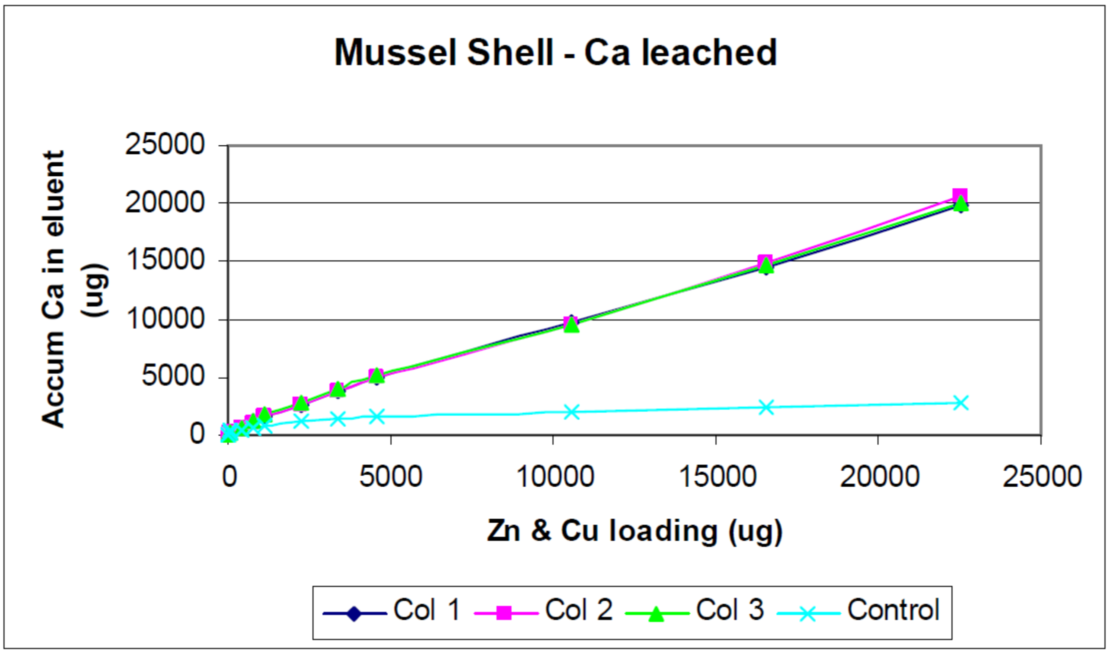


Figure 9.

Comparison of cumulative loading of Zn and Cu on shell and Ca observed in eluent after additions of combined Zn and Cu solutions.



Small amounts of dissolved Ca were found in the eluent for the control using distilled water indicating that for the experiments using metal solutions some of the calcium arriving in the eluent was not solely the result of ion exchange with the mussel shell material. The release of calcium into the eluent continued to increase slightly in the control possibly due to the dried and crushed shell becoming wetted for the first time (Figure 9).

With the addition of solutions containing dissolved Zn and Cu, much greater quantities of Ca were released into the eluent than for the control (Figure 9). The release of Ca into the eluent increased with the increasing load and retention of Zn and Cu suggesting that the removal of these metals is largely due to the formation of insoluble metal carbonates through ion exchange, although it is possible that the organic component in the shell material was also involved in the removal of metal ions from the solution.

Although it was not specifically tested for, there was no clear evidence of displacement between Zn and Cu for binding sites on the shell material (Köhler et al. 2006). However, in experiment four a reduced ability to retain Zn ions while Cu continued to be as efficiently retained suggested there may be competition between the different metals for binding sites.

Cu is generally considered to be adsorbed more easily than other metal ions (Coşkun et al. 2006), however, this has been shown to not always be the case as the capture of Cu by some sorbents is highly dependent on pH (Wu and Zhou 2009). Unfortunately, in this current study no measurements of pH were recorded despite the potential for changes in pH to be caused by chemical reactions between the dissolved metal compounds and the shell material (Genç-Fuhrman et al. 2007, Wu and Zhou 2009).

Overall, the results of these experiments indicate that crushed mussel shell is effective at capturing Zn and Cu in a flow-through situation at concentrations that can be experienced in stormwater. The results also indicate that the mussel shell has considerable overall capacity to capture and retain Zn and Cu, with approximately 1 mg of Zn and 0.2 mg Cu per gram of crushed mussel shell sequestered by the test column, with a continuing capacity to capture metals indicated. In comparison, the earlier nine day static experiment indicated that up to 30 mg of Zn and 1.5 mg of Cu per gram of mussel shell can be removed from solution (Craggs et al. 2006). It is not possible to compare these metal ion capture values with those for published research on shell material or other dissolved metal ion sorbents for a number of reasons, including different metal species and different measurement units that cannot be converted.

In a system where stormwater is filtering through a bed of shell material, the contact time in comparison with the kinetics of the contaminant binding will be an important factor in controlling removal efficiency. This current experiment demonstrated that crushed *P. canaliculus* shell (size fraction 500 – 2000 μm) was effective at removing Zn and Cu from solution over a 24 hour period that would be sufficient for practical application in stormwater filtration. Only when applying solutions containing high metal loadings (far exceeding those typically found in stormwater) did measurable quantities of Zn pass through the shell material.

6 Column experiment two

6.1 Methods

While the prior flow-through experiments demonstrated the ability of mussel shell to retain Zn and Cu from a solution flowing through crushed mussel shell, the low flow rates used in the previous experiments were not entirely representative of what might be encountered in an urban stormwater treatment situation (Pickmere and Mathieson 2007). Therefore, further experiments were undertaken to assess the removal of Zn and Cu from solutions passed through a column of crushed mussel shell at higher flow rates (Macaskill 2008).

Previous studies have indicated that the retention of dissolved metals by bioasorbants is highly dependent on the surface area of the media (Bailey et al. 1999, Köhler et al. 2006, Genç-Fuhrman et al. 2007). Therefore, further experiments were undertaken to assess whether particle size of crushed mussel shell affected the performance of this media for the removal of dissolved Zn and Cu (Macaskill 2008).

6.2 Column preparation

Cleaned and dried green-lipped mussel shell (Perna canaliculus) was crushed in the laboratory with a hammer and initially dry sieved, then wet sieved in order the remove residual fines adhering to the material. The following seven particle size fractions were isolated by sieving; 63-125, 125-250, 250-500, 500-1000, 1000-1400, 1400-1700, and 1700-2000 µm.

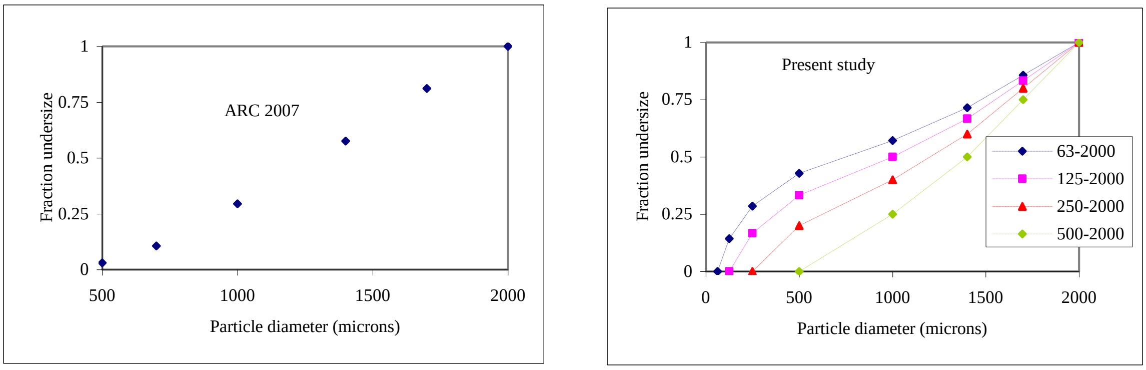
Since it was unlikely that the particle size distribution of any laboratory-scale grinding would closely resemble that of an industrial scale grinding of shells, no attempt was made to produce a realistic particle size distribution. Rather, the desired mix of particle sizes was made from combining a measured mass of particles of the appropriate size fractions.

For the experiments four blends of mussel shell particle size ranges were used; 63-2000, 125 – 2000, 250- 2000, 500 – 2000 µm. For example, the 500 – 2000 µm blend of mussel shell particles consisted of a blend of 25% by mass of each of four largest particle fractions; i.e., 500-1000, 1000-1400, 1400-1700, and 1700-2000 µm fractions, whereas the 63-2000 µm material consisted of 14.3% of each of all seven particle fractions.

This approach allowed for the particle size range to be well characterised, although it is possible it may have over-represented the abundance of the finer fractions that would be potentially be found from an industrial shell crusher. For example, the 63-2000 µm blend contains more than 40% w/w of mussel shell material finer than 500 µm. The 500 – 2000 µm blend had a similar particle size distribution to the material used in the previous study by Pickmere and Mathieson (2007) who sieved crushed mussel shell to a 500-2000 µm range rather than from mixing narrower ranges of particle size fractions (Figure 10).

Figure 10:

Particle size distribution of crushed mussel shell used in the present study and in the earlier study by Pickmere and Mathieson (2007).



A bed of glass beads topped with glass wool was laid in the base of four borosilicate glass columns that were 50 mm in diameter. Then 100 cc of each of the four blends of mussel shell particles were measured and loaded into the separate columns on top of the glass wool. This required around 150 g of crushed mussel shell. This arrangement provided good support for the mussel shell material and during the experiments there was no visual evidence of fine material from the mussel shells passing out of the columns and into the eluent.

6.2.1 Absorption experiments

The four columns containing different crushed mussel of different particle size ranges were used for a series of two subsequent absorption experiments.

The aim of the first experiment was to determine the concentration of dissolved Zn at which the column of crushed mussel shell material was incapable of retaining the sufficient Zn so the eluent did not exceed an arbitrary threshold of 0.05 mg l^{-1} of Zn not captured by the mussel shell. Aliquots of 250 ml of Zn solutions of increasing concentration were passed serially through the crushed mussel shell, i.e., 0, 2, 5, 10, 20, 50, 100, 200, 200 (i.e., 200 ml × 2) mg l^{-1}. After each aliquot had passed through a column the eluent was retained for chemical analyses of total Ca and Zn, along with a sample of the original stock solution prior to being passed through a column. All analyses were performed by Hill Laboratories Ltd in Hamilton with total metals being determined by nitric acid digestion followed by trace level ICP-MS. Retaining and analysing a representative subsample of each eluent provided an integration of column absorption efficiency for that solution and therefore the total column-loading is calculable at every step of the experiment.

The aim of the second experiment was to assess the performance of the four mussel shell columns in removing Zn when continuing to be loaded with dissolved Zn solutions. These loadings were adjusted for the individual columns based on their performance in the first experiment. The column containing crushed mussel shell in the 500-1000 µm range was supplied sequentially with 10 aliquots of 250 ml of each of the following concentrations of dissolved Zn solution 50, 50, 50, 2, 2, 50, 50, 50, 50, and 2 mg l^{-1}. The 1000-1400 µm, 1400-1700 µm, and 1700-2000 µm fractions were each supplied sequentially with 12 aliquots of 250 ml of each of the following concentrations of dissolved Zn solution 100, 100,100, 100, 2, 2, 100, 100, 100, 100, 100, and 2 mg l^{-1}. After each aliquot had passed through a column the eluent was retained for chemical analyses of total Ca and Zn, along with a sample of the original stock solution prior to being passed through a columns. All analyses were performed by Hill Laboratories Ltd in Hamilton.

For all experiments an attempt was made to control the flow rate of the solution through the column to around 200 ml hr^{-1}. This was achieved through a combination of the different permeability of the crushed shell material and the corresponding adjustment of the stopcock outflow on each column. This flow rate represents a velocity of around 10 cm hr^{-1} and a solution:solid contact time of 30 minutes.

Only a single column was run for each size range of crushed mussel shell particles, i.e., there were no replicates. No control columns were used in the experiment.

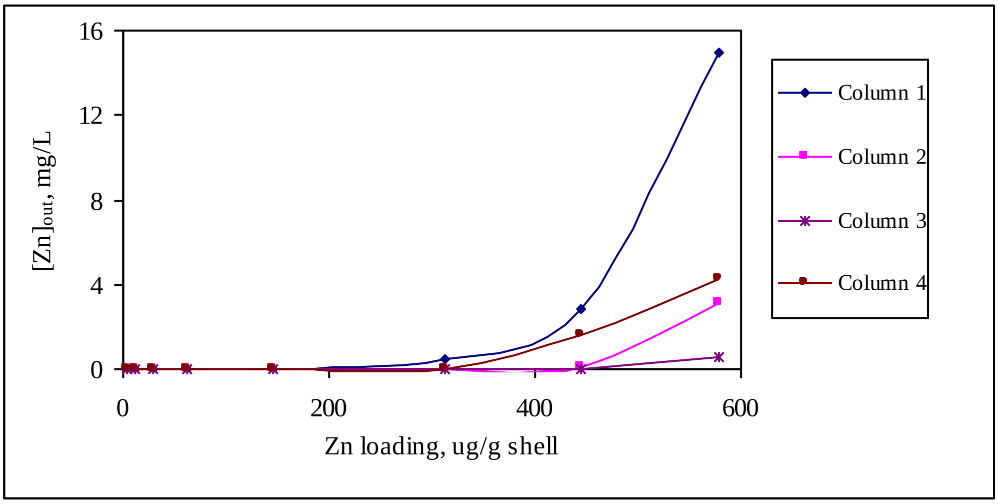
6.3 Results and discussion

The results of the first experiment, fixed volumes of solutions with $[Zn]_{in}$ ranging from 2-200 mg l^{-1}, are shown in Figure 11, with the full data set provided in Appendix 3.

In the first experiment the eluent of all four columns exceeded the pre-set threshold of 0.05 mg l-1 of Zn not captured by the mussel shell (Figure 11). The column containing 500 – 2000 µm mussel shell particles was the first to exceed the threshold when supplied with the 100 mg l^{-1} solution of Zn. Both the 250- 2000 µm and 63-2000 µm columns exceeded the threshold when supplied with the first of the two 200 mg l^{-1} solutions of Zn. The 125 – 2000 µm column only exceeded the threshold when supplied with the second of the two 200 mg l^{-1} solutions of Zn.

Figure 11:

Illustration of loss of absorption efficiency with progressive Zn loading onto columns comprising crushed mussel shell of varying particle size, Column 1 = 500 – 2000 µm, Column 2 = 250- 2000 µm, Column 3 = 125 – 2000 µm, Column 4 = 63-2000 µm.



These breaches in the pre-set threshold appeared to indicate overall differences in the ability of the different particle ranges of mussel shell to retain dissolved Zn. By the end of the first experiment the column of 500 – 2000 µm mussel shell particles had poorest performance in retaining Zn, retaining 96.89% of the Zn loaded into the column. In contrast, the 250- 2000 µm, 125 – 2000 µm, and 63-2000 µm retained 99.44%, 99.90% and 98.99% respectively, of the Zn loaded into each column.

These trends continued with further loading of the columns with Zn solutions in experiment two, although in this experiment the Zn loadings varied among the columns. At the end of experiment two the column of 500 – 2000 µm mussel shell particles had retained 97.93% of a total loading of 14.39 mMol of Zn, compared with 99.43% , 99.90% and 99.48% retention of a total loading of 22.84 mMol of Zn for the columns of 250- 2000 µm, 125 – 2000 µm and 63-2000 µm mussel shell particles respectively. Effectively there was very little, if any, difference in the ability of the three columns containing the finer mussel shell particle for retaining the dissolved Zn. However, the 500 – 2000 µm mussel shell particles had the poorest performance in its ability to retain dissolved Zn.

Smaller particles generally have higher surface areas than the equivalent mass of larger particles made of the same material, although this is not always the case (Köhler et al. 2006). High surface areas are generally associated with greater effectiveness of dissolved heavy metal sorbent media because there is greater contact surface available on which chemical adsorption can occur (Smith 1998; Köhler et al. 2007; Tudor et al. 2006; Genç-Fuhrman et al. 2007). Unfortunately, in this study the surface area of the particles in the mussel shell columns was not measured using standard methodology, such as the nitrogen BET method.

The four mussel shell columns were made up to the same volume (100 cc) but the total mass of shell material was reported to be similar among columns (around 150 g), however, this is unlikely as the packing densities of the different shell particle sizes would have been markedly different. Therefore, it is likely that there were marked differences in the total mass, packing density, and total shell surface area within the different expeimental columns, whilst only the total volume of shell material in the column (100 cc) was kept constant.

This feature of the experimental design makes it a little difficult to interpret the results. For example, differences in the flow rates among the columns were controlled by use of a stopcock on the columns. However, differences in flow rates through the columns would have also have also been affected by the differences in the packing density of varying shell particle size ranges. The solution could be expected to flow more quickly through a column and have less overall contact with shell surfaces, when the shell packing density was lower and there are a greater number of larger spaces through which the liquid can flow. This was likely to be the case for the column with the 500 – 2000 µm mussel shell particles and may have been a major reason why this column was less effective at retaining dissolved metals. Any future research should examine packing density, overall mass of material, surface area in relation to particle size of shell material. Any differences in these variables should be controlled and/or recorded so that rigorous experimental comparisons of metal sorbent performance can be made.

Proportionately more Ca was released from the columns than the amount of Zn retained by the columns (Figure 12). This relationship between the release of Ca and the retenton of Zn appeared to differ among the four different mussel shell particle ranges, with columns containing a greater proportion of large particles tending to release more Ca. This trend became more pronounced as the total quantity of Zn retained by the shell increased, especially for 500 – 2000 µm mussel shell column.

The results indicate that cation exchange between Ca in the mussel shell and the Zn in solution is likely to be the dominant mechanism for the capture of the Zn in the columns. However, this mechanism does not explain the differences in the amount of Ca released among the columns with different ranges of particle size of mussel shell because the most efficient columns for capturing Zn were the columns with proportionately the least amount of Ca released per unit of Zn captured.

This result may be due to differences in the pH of solutions generated among the different columns. Unfortunately, in this current study no measurements of pH were reported despite the potential for changes in pH to be caused by chemical reactions between the dissolved metal compounds and the shell material (Craggs et al. 2006; Genç-Fuhrman et al. 2007; Wu and Zhou 2009). Differences in pH could also influence capture of the Zn because the effectiveness of some metal sorbents is highly dependent on pH (Wu and Zhou 2009).

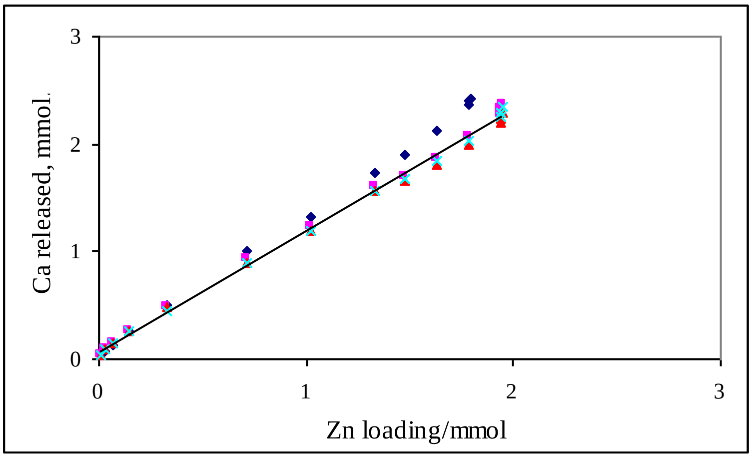
The observed differences in the release of Ca from columns with crushed mussel shell of different particle size ranges may also be due to the presence of other mechanisms involved in retaining metals. For example, it is possible that crushing shell to smaller particle sizes (<500 µm) may make shell chitin more readily available at the surface of particles and in so doing, improve its effectiveness for interacting with, and retaining metals. An improvement in the ability of the chitin within shell for capturing dissolved Zn would also reduce the concentration of Zn available for exchange with the Ca of the shell. This possibility would explain both the higher metal retention performance of the columns with the greater proportion of fine shell particles, as well as their lower release of Ca.

Overall, the results of these experiments demonstrate again that crushed mussel shell is highly effective at retaining dissolved Zn and Cu in a flow-through situation at concentrations that are typical for urban stormwater. The results also indicate that the shell material has considerable capacity to capture and retain Zn from solution, with a total of over 22.7 mMol of Zn sequestered by the crushed mussel shell in three of the experimental columns, and all with an ongoing capacity to capture Zn indicated.

Compared to the previous experiment by Pickmere and Mathieson (2007) the results show that the increased flow rates of solutions through the columns of crushed mussel shell (i.e., 30 minutes versus 6 hours) made no apparent difference to the efficiency of the columns in retaining dissolved metal ions.

Figure 12:

Relationship between Zn loading and column calcium release. Data are presented for all four columns.



7 Conclusions and indicative directions for future work

Overall, the results of this set of experiments clearly demonstrate that the crushed shell of the New Zealand green-lipped mussel, *Perna canaliculus*, is very effective at capturing Zn and Cu from solutions at relatively low concentrations, i.e., down to 0.1 µg ml^{-1} (Macaskill 2008). Such concentrations are within the range that these metal contaminants are typically found in urban stormwater.

In experiments where solutions of dissolved metals were passed through columns of crushed mussel shell the retention of the metals was initially very close to 100%, and continued to remain very high despite subsequent relatively heavy loadings of dissolved metals. For example, a column of 150 g of crushed mussel shell with a particle size range of 125 – 2000 µm removed 99.8% of 0.03 mMol of dissolved Zn, after previously sequestering 22.8 mMol of Zn (Macaskill 2008).

Furthermore, the mussel shell has considerable capacity to retain dissolved metal ions, with up to 30 mg of Zn and 1.5 mg of Cu per gram of shell being removed from a static solution after nine days (Craggs et al. 2006), approximately 1 mg of Zn together with 0.2 mg Cu per gram of crushed mussel shell were sequestered when multiple solutions were each flowed through a test column over 6 hours (Pickmere and Mathieson 2007), and approximately 9.9 mg of Zn per gram of crushed mussel shell was sequestered when multiple solutions of dissolved metals were each flowed through a test column eachover a period of over 30 minutes (Macaskill 2008). In the case of flow-through experiments all of the columns for which data are presented appear to have continuing capacity to capture Zn at the end of the experiments, despite already being subjected to substantial loadings of dissolved metals.

Small columns of crushed shell material remained efficient at removing dissolved metals at flow velocity of around 10 cm hr^{-1} and a solution:solid contact time of 30 minutes (Macaskill 2008). Higher flow rates are likely to be required for the treatment of urban stormwater, so the efficacy of crushed shell material at higher flow velocities needs to be determined through further research.

The particle size range of the crushed mussel shell appears to be important in determining the efficiency of the shell as a sorbent of dissolved metals, with shell material crushed to <500 µm seemingly providing superior performance (Macaskill 2008). The reasons for this improved performance are not clear, but are likely to be related to increased surface area of particles, or greater exposure of organic components within the shell matrix that may also be active in retaining metals (Yan-jiao 2011). However, the release of Ca concomitantly with the capture of dissolved metals by crushed mussel shell indicates that cation exchange resulting in precipitation of insoluble metal carbonates is probably the dominant mechanism by which dissolved metals are retained by the mussel shell (Tudor et al. 2006).

The use of different sized shell particles for the removal of metals from stormater will also need to carefully consider the practicalities of producing the desired particle size ranges. Industrial crushing methods for shell tend to produce a wide range of particle sizes and there would be considerable additional costs of size sorting crushed material to select the desirable particle size range for water treatment.

There is an indication from one set of experiments using solutions with a mix of Cu and Zn that there were differences in the ability of mussel shell to capture the different metal ions. In this experiment the mussel shell showed a reduced ability to retain Zn while Cu continued to be efficiently retained. In other studies Cu has typically been found to be adsorbed more easily than other metals (Coşkun et al. 2006), although this can vary with the type of sorbent media and pH (Wu and Zhou 2009). Unfortunately, in this current experiment no measurements of pH were reported despite the potential for changes in pH to be caused by chemical reactions between the dissolved metal compounds and the shell material (Färm 2002; Genç-Fuhrman et al. 2007; Wu and Zhou 2009). It is important for future research to either measure pH or control for possible effects from varying pH.

Although the on-going retention of captured metals was not specifically examined in the experiments the use of multiple sequential solutions of dissolved metals with subsequent examination of the eluent suggested that there was little or no leaching of previously captured metals. Likewise, where a solution containing the two dissolved metals (Zn and Cu) was used, there was apparently no substitution between one of the metals and the other metal which had been retained previously, thereby resulting in release of this displaced metal into the eluent.

Overall, the combined experimental results strongly indicate that crushed mussel shell has considerable potential for the treatment of urban stormwater containing dissolved Zn, Cu, and potentially other metal cations, such as Pb and Cd. Besides metal ions, crushed bivalve shell material also appears capable of capturing dissolved phosphate especially when the shell has been heat treated (>750 °C) to alter its chemical structure (Kwon et al. 2004; Lee et al. 2005; Namasivayam et al. 2005; Currie et al. 2007).

It is difficult to compare the performance of green-lipped mussel shell with previously published reports of the use of other types of shell and other sorbents for a similar purpose because of differences in the studies including the use of different metal species and different measurement units and methods that are not directly comparable (Tudor et al. 2006; Ok et al. 2010; Yan-jiao 2011).

Mussel shell is a significant waste product from aquaculture in New Zealand and is mostly dumped at land fill sites close to processing plants (Barnaby 2004). Given mussel shell is a waste product it has a low supply cost, although handling, crushing, and freight will add to the cost of utilising this product for stormwater treatment. Crushed shell material also has the potential advantage of being able to replace washed-graded sand in conventional sand filters designed to remove particulate sources of heavy metal in stormwater. By substituting crushed mussel shell for sand in such filters the filter is likely to be able to not only remove particulate, but also dissolved metals.

Further research is recommended to more fully determine the potential of crushed mussel shell for treating dissolved metals in stormwater. The role of particle size and surface area of crushed mussel shell in influencing the efficiency of capturing dissolved metals needs to be determined. Also, the effectiveness of the shell material at higher flow rates needs to be investigated.

In conducting any future research careful attention needs to be paid to some key factors. Most importantly the pH of solutions used and eluents produced need to be carefully monitored because pH is known to greatly affect the efficiency of capture of some metal ions by some sorbents (Wu and Zhou 2009). Chemical reactions between the dissolved metal compounds and the shell material could potential cause changes in the pH of experimental solutions and in turn alter the efficiency of the metal capture (Genç-Fuhrman et al. 2007; Wu and Zhou 2009).

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9 Appendix 1: Batch experiment

Table 1: Day 2 analytical data set.

Sample	Conc (mg l^{-1})	Day 2 pH	Ca (mg l^{-1})	Cu (mg l^{-1})	Zn (mg l^{-1})
Blank	0	5.9	BD	BD	BD
Shell Blank	0	6.2	2.88	BD	BD
[Zn] test soln.	50	6.7	BD		46.2
Zn - a	50	6.4	12.9		31
Zn - b	50	5.4	13.7		30.8
Zn - c	50	6.6	12.4		32.2
Zn - a	100	6.7	22		67.3
Zn - b	100	6.8	23		66.9
Zn - c	100	6.8	21		70.8
Zn - a	500	6.8	51		378
Zn - b	500	6.8	39		392
Zn - c	500	6.8	32		395
Zn - a	1000	6.6	56		809
Zn - b	1000	6.6	53		776
Zn - c	1000	6.7	57		800
[Cu] test soln.	10	6.7	BD	8.88	
Cu - a	10	6.6	2.9	6.65	
Cu - b	10	6.6	2.8	7.49	
Cu - c	10	6.6	2.6	7.62	
Cu - a	25	6.4	3.1	19.9	
Cu - b	25	6.4	3	19.3	
Cu - c	25	6.4	3.4	22.2	
Cu - a	50	6.2	4	50.3	
Cu - b	50	6.3	5	49	
Cu - c	50	6.2	4.2	49.3	
Cu - a	100	6.1	7	106	
Cu - b	100	6.1	6	104	
Cu - c	100	6.1	7	104	

BD – Below detection

Table 2: Day 9 analytical data set.

Sample	Conc. (mg l^{-1})	Day 9 pH	Ca (mg l^{-1})	Cu (mg l^{-1})	Zn (mg l^{-1})
Blank	0	6	BD	BD	BD
Shell Blank	0	7.3	6.85	BD	BD
[Zn] test soln.		5.7	BD		49.5
Zn - a	50	7.1	41.8		2.15
Zn - b	50	7.4	39.9		2.92
Zn - c	50	7.5	41.7		2.89
Zn - a	100	7.6	69		8
Zn - b	100	7.6	72		8.3
Zn - c	100	7.6	75		7.6
Zn - a	500	7	214		135
Zn - b	500	7	220		124
Zn - c	500	7	208		158
Zn - a	1000	6.8	313		462
Zn - b	1000	6.7	303		439
Zn - c	1000	6.8	318		432
[Cu] testsoln	10	6.1	BD	10.2	
Cu - a	10				
Cu - b	10	7.2	8.2	1.35	
Cu - c	10	7.4	9	0.84	
Cu - a	25	6.9	12	4.63	
Cu - b	25	7	12.6	4.84	
Cu - c	25	7	11.1	8.84	
Cu - a	50	6.8	17.9	22.8	
Cu - b	50	6.8	21.5	20.3	
Cu - c	50	6.8	16.3	22.4	
Cu - a	100	6.6	30	58.1	
Cu - b	100	6.6	28	60.3	
Cu - c	100	6.5	32	54.5	

BD – Below detection

10 Appendix 2: Column experiment one

Sample	Description	Time hrs	Addition mls	Ca loading	Ca leach filt ug/ml *	Ca leach partic ug/ml	Ca cum loss ug	Ca cum leach filt ug	Ca cum leach partic ug
M1	Col 1, Expt 1	24	30	0	2.51	0.17	75.3	75.3	5.1
M5		48	30	0	3.14	0.07	169.5	94.2	2.1
M9		72	30	0	3.5	0.22	274.5	105	6.6
M14	Col 1, Expt 2	24	50	0	7.18	0.1	633.5	359	5
M18		48	50	0	9.57	0.01	1112	478.5	0.5
M22		72	50	0	10.3	0	1627	515	0
M27	Col 1, Expt 3	24	50	0	19.6	0.3	2607	980	15
M31		48	50	0	23.3	0	3772	1165	0
M35		72	50	0	23.9	0	4967	1195	0
M40	Col 1, Expt 4	24	50	0	94	0	9667	4700	0
M44		48	50	0	98	3	14567	4900	150
M48		72	50	0	106	0	19867	5300	0
			Sum	0			Sum	19867	184.3
M2	Col 2, Expt 1	24	26	0	2.52	0.18	65.52	65.52	4.68
M6		48	26	0	3.15	0.17	147.42	81.9	4.42
M10		72	26	0	3.61	0.15	241.28	93.86	3.9

Sample	Description	Time hrs	Addition mls	Ca loading	Ca leach filt ug/ml *	Ca leach partic ug/ml	Ca cum loss ug	Ca cum leach filt ug	Ca cum leach partic ug
M15	Col 2, Expt 2	24	50	0	7.1	0.32	596.28	355	16
M19		48	50	0	9.35	0.16	1063.78	467.5	8
M23		72	50	0	9.93	0.27	1560.28	496.5	13.5
M28	Col 2, Expt 3	24	50	0	20.1	0.1	2565.28	1005	5
M32		48	50	0	23	0.5	3715.28	1150	25
M36		72	50	0	23.4	1	4885.28	1170	50
M41	Col 2, Expt 4	24	50	0	94.6	0	9615.28	4730	0
M45		48	50	0	106	0	14915.28	5300	0
M49		72	50	0	113	0	20565.28	5650	0
			Sum	0			Sum	20565.28	130.5
M3	Col 3, Expt 1	24	26	0	2.93	0.13	76.18	76.18	3.38
M7		48	26	0	3.31	0.04	162.24	86.06	1.04
M11		72	26	0	3.57	0.22	255.06	92.82	5.72
M16	Col 3, Expt 2	24	50	0	8.43	0.21	676.56	421.5	10.5
M20		48	50	0	10.5	0	1201.56	525	0
M24		72	50	0	10.6	0.7	1731.56	530	35
M29	Col 3, Expt 3	24	50	0	20.8	0.2	2771.56	1040	10
M33		48	50	0	24.3	0	3986.56	1215	0
M37		72	50	0	24.8	0	5226.56	1240	0
M42	Col 3, Expt 4	24	50	0	86.9	4.1	9571.56	4345	205
M46		48	50	0	104	3	14771.56	5200	150

Sample	Description	Time hrs	Addition mls	Ca loading	Ca leach filt ug/ml *	Ca leach partic ug/ml	Ca cum loss ug	Ca cum leach filt ug	Ca cum leach partic ug
M50		72	50	0	105	2	20021.56	5250	100
			Sum	0			Sum	20021.56	520.64
M4	Blank Col, Expt 1	24	30	0	2.62	0.09	78.6	78.6	2.7
M8		48	30	0	3.26	0.04	176.4	97.8	1.2
M12		72	30	0	4.48	0	310.8	134.4	0
M17	Blank Col, Expt 2	24	50	0	3.39	0.01	480.3	169.5	0.5
M21		48	50	0	3.89	0.16	674.8	194.5	8
M25		72	50	0	4.28	0	888.8	214	0
M30	Blank Col, Expt 3	24	50	0	4.19	0.01	1098.3	209.5	0.5
M34		48	50	0	5.04	0.05	1350.3	252	2.5
M38		72	50	0	5.54	0.05	1627.3	277	2.5
M43	Blank Col, Expt 4	24	50	0	7.05	0	1979.8	352.5	0
M47		48	50	0	7.24	0.02	2341.8	362	1
M51		72	50	0	7.65	0.01	2724.3	382.5	0.5
			Sum	0			Sum	2724.3	19.4
		DL 0.05ug/ml							

Zinc

Sample		Time hrs	Addition mls	Zn loading	Zn leach filt ug/ml *	Zn leach partic ug/ml	Volume mls	Zn cum loading ug		Zn cum leach filt ug	Zn cum leach partic ug
M1	Col 1, Expt 1	24	30	15.03	0.001	0	30	15.03		0.03	0
M5	0.501ug/ml	48	30	15.03	0.002	0	60	30.06		0.06	0
M9		72	30	15.03	0.002	0.006	90	45.09		0.06	0.18
M14	Col 1, Expt 2	24	50	289	0.002	0	140	334.09		0.1	0
M18	5.78ug/ml	48	50	289	0.003	0	190	623.09		0.15	0
M22		72	50	289	0.001	0	240	912.09		0.05	0
M27	Col 1, Expt 3	24	50	1045	0	0.0006	290	1957.09		0	0.03
M31	20.9ug/ml	48	50	1045	0	0	340	3002.09		0	0
M35		72	50	1045	0.0004	0	390	4047.09		0.02	0
M40	Col 1, Expt 4	24	50	4965	0.653	0.016	440	9012.09		32.65	0.8
M44	99.3ug/ml	48	50	4965	1.77	0.07	490	13977.09		88.5	3.5
M48		72	50	4965	4	0.08	540	18942.09		200	4
				18942.09					Sum	321.62	8.51
M2	Col 2, Expt 1	24	26	13.026	0.003	0	26	13.026		0.078	0
M6		48	26	13.026	0.002	0	52	26.052		0.052	0
M10		72	26	13.026	0	0	78	39.078		0	0
M15	Col 2, Expt 2	24	50	289	0.007	0	128	328.078		0.35	0
M19		48	50	289	0.002	0	178	617.078		0.1	0
M23		72	50	289	0.001	0	228	906.078		0.05	0

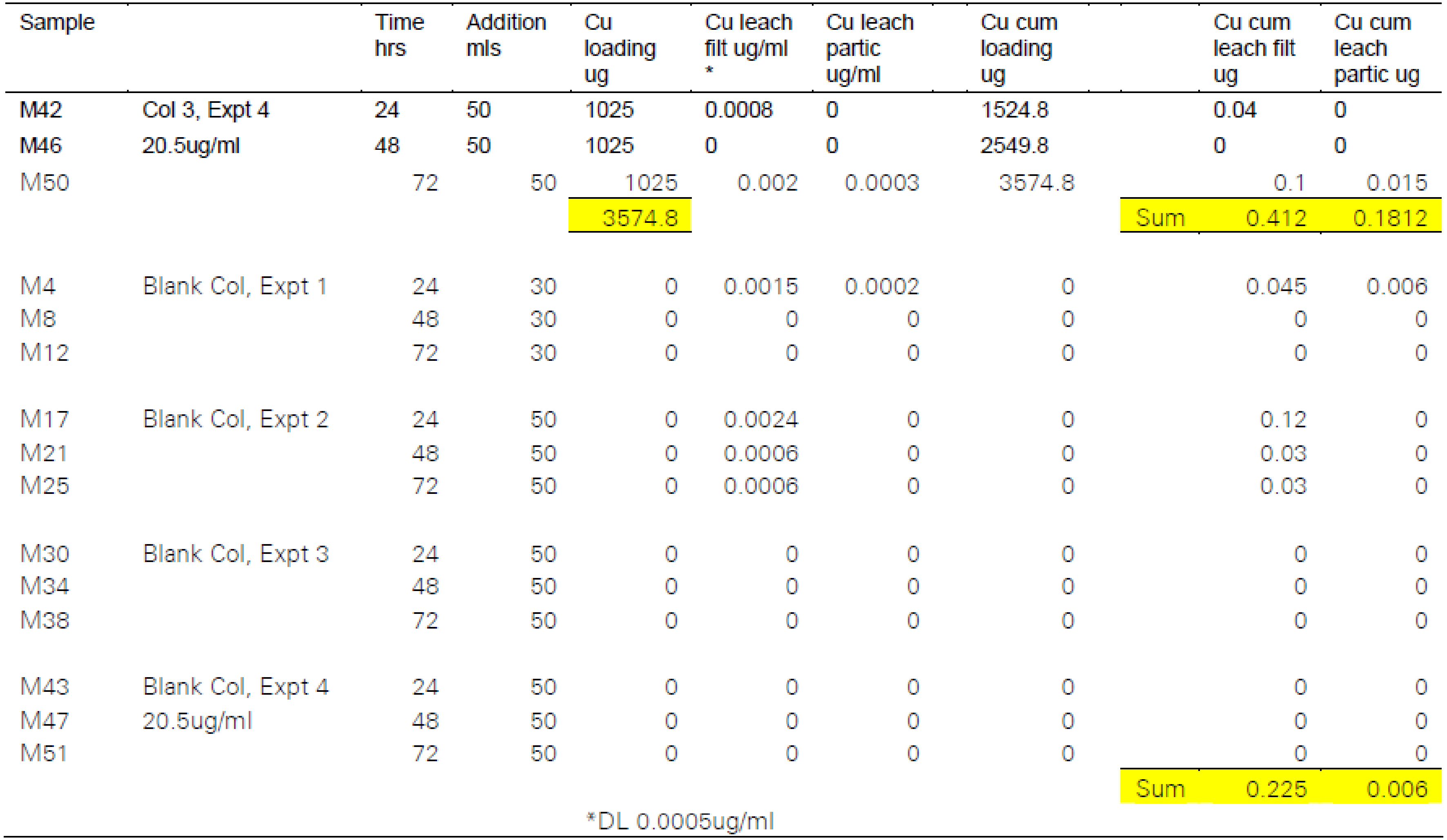
Sample		Time hrs	Addition mls	Zn loading	Zn leach filt ug/ml *	Zn leach partic ug/ml	Volume mls	Zn cum loading ug		Zn cum leach filt ug	Zn cum leach partic ug
M28	Col 2, Expt 3	24	50	1045	0	0	278	1951.078		0	0
M32		48	50	1045	0	0	328	2996.078		0	0
M36		72	50	1045	0.002	0	378	4041.078		0.1	0
M41	Col 2, Expt 4	24	50	4965	0.564	0.014	428	9006.078		28.2	0.7
M45		48	50	4965	1.09	0.2	478	13971.08		54.5	10
M49		72	50	4965	1	0.02	528	18936.08		50	1
				18936.08					Sum	133.43	11.7
M3	Col 3, Expt 1	24	26	13.026	0.003	0	26	13.026		0.078	0
M7		48	26	13.026	0.002	0.008	52	26.052		0.052	0.208
M11		72	26	13.026	0.006	0	78	39.078		0.156	0
M16	Col 3, Expt 2	24	50	289	0.002	0	128	328.078		0.1	0
M20		48	50	289	0.002	0	178	617.078		0.1	0
M24		72	50	289	0.002	0	228	906.078		0.1	0
M29	Col 3, Expt 3	24	50	1045	0	0	278	1951.078		0	0
M33		48	50	1045	0	0	328	2996.078		0	0
M37		72	50	1045	0.001	0	378	4041.078		0.05	0
M42	Col 3, Expt 4	24	50	4965	0.363	0.004	428	9006.078		18.15	0.2
M46	99.3ug/ml	48	50	4965	1.77	0.12	478	13971.08		88.5	6
M50		72	50	4965	4.52	0.1	528	18936.08		226	5
				18936.08					Sum	333.286	11.408

Sample		Time hrs	Addition mls	Zn loading	Zn leach filt ug/ml *	Zn leach partic ug/ml	Volume mls	Zn cum loading ug		Zn cum leach filt ug	Zn cum leach partic ug
M4	Blank Col, Expt 1	24	30	0	0.002	0.008		0		0.06	0.24
M8		48	30	0	0.006	0		0		0.18	0
M12		72	30	0	0.002	0		0		0.06	0
M17	Blank Col, Expt 2	24	50	0	0.019	0		0		0.95	0
M21		48	50	0	0.005	0		0		0.25	0
M25		72	50	0	0.006	0		0		0.3	0
M30	Blank Col, Expt 3	24	50	0	0	0		0		0	0
M34		48	50	0	0	0		0		0	0
M38		72	50	0	0	0		0		0	0
M43	Blank Col, Expt 4	24	50	0	0.01	0		0		0.5	0
M47		48	50	0	0.002	0		0		0.1	0
M51		72	50	0	0.002	0		0		0.1	0
				0					Sum	2.5	0.24
			*DL 0.001ug/ml								

Copper

Sample		Time hrs	Addition mls	Cu loading ug	Cu leach filt ug/ml *	Cu leach partic ug/ml	Cu cum loading ug		Cu cum leach filt ug	Cu cum leach partic ug
M1	Col 1, Expt 1	24	30	3	0.0014	0.001	3		0.042	0.03
M5	0.100ug/ml	48	30	3	0.002	0	6		0.06	0
M9		72	30	3	0.0036	0.0004	9		0.108	0.012
M14	Col 1, Expt 2	24	50	60.5	0.001	0	69.5		0.05	0
M18	1.21ug/ml	48	50	60.5	0.0006	0	130		0.03	0
M22		72	50	60.5	0.0007	0	190.5		0.035	0
M27	Col 1, Expt 3	24	50	103.5	0	0.0006	294		0	0.03
M31	2.07ug/ml	48	50	103.5	0	0	397.5		0	0
M35		72	50	103.5	0	0	501		0	0
M40	Col 1, Expt 4	24	50	1025	0	0.0005	1526		0	0.025
M44	20.5ug/ml	48	50	1025	0	0	2551		0	0
M48		72	50	1025	0.0007	0.0002	3576		0.035	0.01
				3576				Sum	0.36	0.107
M2	Col 2, Expt 1	24	26	2.6	0.0015	0.0001	2.6		0.039	0.0026
M6	0.100ug/ml	48	26	2.6	0.0017	0.0001	5.2		0.0442	0.0026
M10		72	26	2.6	0.004	0.0002	7.8		0.104	0.0052

Sample		Time hrs	Addition mls	Cu loading ug	Cu leach filt ug/ml *	Cu leach partic ug/ml	Cu cum loading ug		Cu cum leach filt ug	Cu cum leach partic ug
M15	Col 2, Expt 2	24	50	60.5	0.0043	0.007	68.3		0.215	0.35
M19	1.21ug/ml	48	50	60.5	0.0008	0.002	128.8		0.04	0.1
M23		72	50	60.5	0.0008	0.001	189.3		0.04	0.05
M28	Col 2, Expt 3	24	50	103.5	0.0009	0	292.8		0.045	0
M32	2.07ug/ml	48	50	103.5	0	0	396.3		0	0
M36		72	50	103.5	0	0	499.8		0	0
M41	Col 2, Expt 4	24	50	1025	0	0.0009	1524.8		0	0.045
M45	20.5ug/ml	48	50	1025	0	0	2549.8		0	0
M49		72	50	1025	0	0	3574.8		0	0
				3574.8				Sum	0.5272	0.5554
M3	Col 3, Expt 1	24	26	2.6	0.0014	0	2.6		0.0364	0
M7	0.100ug/ml	48	26	2.6	0.0015	0.0002	5.2		0.039	0.0052
M11		72	26	2.6	0.0016	0.006	7.8		0.0416	0.156
M16	Col 3, Expt 2	24	50	60.5	0.0009	0.0001	68.3		0.045	0.005
M20	1.21ug/ml	48	50	60.5	0.0007	0	128.8		0.035	0
M24		72	50	60.5	0.0009	0	189.3		0.045	0
M29	Col 3, Expt 3	24	50	103.5	0.0006	0	292.8		0.03	0
M33	2.07ug/ml	48	50	103.5	0	0	396.3		0	0
M37		72	50	103.5	0	0	499.8		0	0



Sample		Time hrs	Addition mls	Cu loading ug	Cu leach filt ug/ml *	Cu leach partic ug/ml	Cu cum loading ug		Cu cum leach filt ug	Cu cum leach partic ug
M42	Col 3, Expt 4	24	50	1025	0.0008	0	1524.8		0.04	0
M46	20.5ug/ml	48	50	1025	0	0	2549.8		0	0
M50		72	50	1025	0.002	0.0003	3574.8		0.1	0.015
				3574.8				Sum	0.412	0.1812
M4	Blank Col, Expt 1	24	30	0	0.0015	0.0002	0		0.045	0.006
M8		48	30	0	0	0	0		0	0
M12		72	30	0	0	0	0		0	0
M17	Blank Col, Expt 2	24	50	0	0.0024	0	0		0.12	0
M21		48	50	0	0.0006	0	0		0.03	0
M25		72	50	0	0.0006	0	0		0.03	0
M30	Blank Col, Expt 3	24	50	0	0	0	0		0	0
M34		48	50	0	0	0	0		0	0
M38		72	50	0	0	0	0		0	0
M43	Blank Col, Expt 4	24	50	0	0	0	0		0	0
M47	20.5ug/ml	48	50	0	0	0	0		0	0
M51		72	50	0	0	0	0		0	0
								Sum	0.225	0.006

*DL 0.0005ug/ml

11 Appendix 3: Column experiment two

Table 1: Results for column 1, with shell particle size range 500-2000 µm.

* Inadvertent use of 20 mg l^{-1} solution rather than client-brief value of 25 mg l^{-1}.

** Where $[Zn]_{out}$ exceeds the chosen critical value of 0.05 mg l^{-1}, results shown in red

$[Zn]_{in}$/ppm	$[Zn]_{out}$/ppm	$[Ca]_{out}$/ppm	V/ml	ΣZn,µg/g shell	$[Zn]_{out}$/µMol/L	$[Zn]_{in}$, mMol/L	$[Ca]_{out}$/mMol/L	$[Zn]_{in}$ +$[Ca]_{out/mMol/L}$	Sample ID	Lab ID
0	<0.0011	4.8					0.12	0.12	A_0	631530.33
2	0.0041	5.2	250	3.3	0.06	0.03	0.13	0.15	1-2	631530.1
5	0.0032	6.7	250	12	0.05	0.076	0.17	0.20	1-5	631530.2
10	0.0078	10	250	28	0.12	0.15	0.25	0.27	1-10	631530.3
20*	0.0021	20	250	62	0.03	0.306	0.50	0.43	1-20	631530.4
50	0.0013	40	250	145	0.02	0.765	1.00	0.88	1-50	631530.5
100	0.44**	79	250	312	6.73	1.53	1.98	1.65	1-100	631530.6
200	2.8	130	100	445	42.83	3.06	3.25	3.18	1-200-1	631530.7
200	15	160	100	578	229.43	3.06	4.00	3.18	1-200-2	631530.8
50	0.023	33	200	645	0.35	0.76	0.83	0.88	1-50-2	633255.5
50	0.048	46	200	712	0.73	0.76	1.15	0.88	1-50-4	633255.6
50	0.095	49	200	778	1.45	0.76	1.23	0.88	1-50-6	633255.7
2	0.018	11	100	780	0.28	0.03	0.28	0.15	1-2-1	633255.20
2	0.017	11	100	781	0.26	0.03	0.28	0.15	1-2-2	633255.24
50	0.041	48	500	945	0.63	0.76	1.20	0.88	1-50-11	635208.1
50	0.17	49	500	1112	2.60	0.76	1.23	0.88	1-50-16	635208.2
50	0.28	49	500	1278	4.28	0.76	1.23	0.88	1-50-21	635208.3

50	0.48	53	500	1445	7.34	0.76	1.33	0.88	1-50-26	635208.4
2	0.029	13	200	1448	0.44	0.03	0.33	0.15	1-2-4	635208.20

Table 2: Results for column 2, with shell particle size range 250-2000 µm.

$[Zn]_{in}$/ppm	$[Zn]_{out}$/ppm	$[Ca]_{out}$/ppm	V/ml	ΣZn,µg/g shell	$[Zn]_{out}$/µmol/L	$[Zn]_{in}$, mmol/L	$[Ca]_{out}$/mmol/L	$[Zn]_{in}$ +$[Ca]_{0/mmol/L}$	Sample ID	Lab ID
2	<0.0011	6.5	250	3.3	0.02	0.03	0.1625	0.15	2-2	631530.9
5	<0.0011	7.2	250	12	0.02	0.08	0.18	0.20	2-5	631530.10
10	0.0018	10	250	28	0.28	0.15	0.25	0.27	2-10	631530.11
20	0.0016	18	250	62	0.24	0.31	0.45	0.43	2-20	631530.12
50	0.0074	36	250	145	0.11	0.76	0.9	0.88	2-50	631530.13
100	0.0036	72	250	312	0.06	1.53	1.8	1.65	2-100	631530.14
200	0.14	120	100	445	2.14	3.06	3.0	3.18	2-200-1	631530.15
200	3.1	150	100	578	47.42	3.06	3.75	3.18	2-200-2	631530.16
100	0.018	37	100	645	0.28	1.53	0.925	1.65	2-100-1	633255.8
100	0.028	63	100	712	0.43	1.53	1.575	1.65	2-100-2	633255.9
100	0.044	82	100	778	0.67	1.53	2.05	1.65	2-100-3	633255.10
100	0.11	86	100	845	1.68	1.53	2.15	1.65	2-100-4	633255.11
2	0.012	16	100	846	0.18	0.03	0.4	0.15	2-2-1	633255.21
2	0.015	15	100	848	0.23	0.03	0.375	0.15	2-2-2	633255.25
100	0.16	71	200	978	2.45	1.53	1.775	1.65	2-100-6	635208.5

100	0.66	86	300	1178	10.09	1.53	2.15	1.65	2-100-9	635208.6
100	1.1	91	500	1512	16.82	1.53	2.275	1.65	2-100-14	635208.7
100	1.5	93	500	1845	22.94	1.53	2.325	1.65	2-100-19	635208.8
100	1.5	99	500	2178	22.94	1.53	2.475	1.65	2-100-24	635208.9
2	0.075	19	200	2181	1.15	0.03	0.475	0.15	2-2-4	635208.21

Table 3: Results for column 3, with shell particle size range 125-2000 µm.

$[Zn]_{in/}$ppm	$[Zn]_{out}$/ppm	$[Ca]_{out}$/ppm	V/ml	ΣZn,µg/g shell	$[Zn]_{out}$/µmol/L	$[Zn]_{in,}$ mmol/L	$[Ca]_{out}$/mmol/L	$[Zn]_{in}$ +$[Ca]_{0/mmol/L}$	Sample ID	Lab ID
2	0.0015	6.9	250	3.3	0.02	0.03	0.1725	0.15	3-2	631530.17
5	0.0033	8	250	12	0.05	0.08	0.2	0.20	3-5	631530.18
10	0.0033	10	250	28	0.05	0.15	0.25	0.27	3-10	631530.19
20	0.0051	18	250	62	0.08	0.31	0.45	0.43	3-20	631530.20
50	0.0039	34	250	145	0.06	0.76	0.85	0.88	3-50	631530.21
100	0.0019	67	250	312	0.03	1.53	1.675	1.65	3-100	631530.22
200	0.011	120	100	445	0.17	3.06	3	3.18	3-200-1	631530.23
200	0.56	150	100	578	8.57	3.06	3.75	3.18	3-200-2	631530.24
100	0.0099	31	100	645	0.15	1.53	0.775	1.65	3-100-1	633255.12
100	0.0089	61	100	712	0.14	1.53	1.525	1.65	3-100-2	633255.13
100	0.017	76	100	778	0.26	1.53	1.9	1.65	3-100-3	633255.14
100	0.033	82	100	845	0.50	1.53	2.05	1.65	3-100-4	633255.15
2	0.013	18	100	846	0.20	0.03	0.45	0.15	3-2-1	633255.22

2	0.017	16	100	848	0.26	0.03	0.4	0.15	3-2-2	633255.26
100	0.006	65	200	978	0.09	1.53	1.625	1.65	3-100-6	635208.10
100	0.022	82	300	1178	0.34	1.53	2.05	1.65	3-100-9	635208.11
100	0.055	91	500	1512	0.84	1.53	2.275	1.65	3-100-14	635208.12
100	0.19	88	500	1845	2.91	1.53	2.2	1.65	3-100-19	635208.13
100	0.48	96	500	2178	7.34	1.53	2.4	1.65	3-100-24	635208.14
2	0.016	20	200	2181	0.24	0.03	0.5	0.15	3-2-4	635208.22

Table 4: Results for column 4, with shell particle size range 63-2000 µm.

[Zn]$_{in}$/ppm	[Zn]$_{out}$/ppm	[Ca]$_{out}$/ppm	V/ml	ΣZn,µg/g shell	[Zn]$_{out}$/µmol/L	[Zn]$_{in}$, mmol/L	[Ca]$_{out}$/mmol/L	[Zn]$_{in}$ +[Ca]$_0$/mmol/L	Sample ID	Lab ID
2	0.0029	6.2	250	3.3	0.04	0.03	0.155	0.15	4-2	631530.25
5	<0.0011	7.4	250	12	0.02	0.08	0.185	0.20	4-5	631530.26
10	0.0066	11	250	28	0.10	0.15	0.275	0.27	4-10	631530.27
20	<0.0011	16	250	62	0.02	0.31	0.4	0.43	4-20	631530.28
50	<0.0011	32	250	145	0.02	0.76	0.8	0.88	4-50	631530.29
100	0.029	71	250	312	0.44	1.53	1.775	1.65	4-100	631530.30
200	1.6	120	100	445	24.47	3.06	3	3.18	4-200-1	631530.31
200	4.3	150	100	578	65.77	3.06	3.75	3.18	4-200-2	631530.32
100	0.011	42	100	645	0.17	1.53	1.05	1.65	4-100-1	633255.16
100	0.04	67	100	712	0.61	1.53	1.675	1.65	4-100-2	633255.17
100	0.065	78	100	778	0.99	1.53	1.95	1.65	4-100-3	633255.18
100	0.099	84	100	845	1.51	1.53	2.1	1.65	4-100-4	633255.19
2	0.0091	19	100	846	0.14	0.03	0.475	0.15	4-2-1	633255.23

2	0.012	17	100	848	0.18	0.03	0.425	0.15	4-2-2	633255.27
100	0.0098	69	200	978	0.15	1.53	1.725	1.65	4-100-6	635208.15
100	0.044	85	300	1178	0.67	1.53	2.125	1.65	4-100-9	635208.16
100	0.16	94	500	1512	2.45	1.53	2.35	1.65	4-100-14	635208.17
100	0.51	93	500	1845	7.80	1.53	2.325	1.65	4-100-19	635208.18
100	0.71	97	500	2178	10.86	1.53	2.425	1.65	4-100-24	635208.19
2	0.06	24	200	2181	0.92	0.03	0.6	0.15	4-2-4	635208.23