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Use of hydrated lime to control *Styela clava* in the PEI mussel farming industry: industry practises and potential effects on non-target invertebrates

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ABSTRACT

Mussel culture in Prince Edward Island must contend with infestations of tunicates particularly the clubbed tunicate, *Styela clava*. Periodically, aquaculture leases are treated with hydrated lime ($Ca(OH)_2$) in order to control these infestations and ensure minimal loss of the mussel crop. Treatment may take several forms but all involve dipping or spraying mussels sleeves with a 4% suspension of hydrated lime. Excess lime from these treatments may be released directly into the surrounding aquatic environment raising questions about the potential biological effects on non-target indigenous species. We herein review the current state of knowledge regarding treatment, chemistry and biological effects of hydrated lime and its post-treatment by-products. Treatment with hydrated lime results in a significant, yet short-lived, increase in pH and the conversion of $Ca(OH)_2$ to carbonate which may precipitate into the environment. Lab-based studies have characterised the hazard associated with elevated pH and the presence of particulate carbonate. Field studies have shown that exposure to hazardous pH or particulate carbonate is unlikely to last more than several minutes and elevated pH is only observed for distances of several metres from the treatment operation. These data lead us to conclude that under current operating conditions lethal effects on non-target organisms resulting from use of hydrated lime are unlikely to occur.

Utilisation de la chaux hydratée pour contrôler le *Styela clava* dans l'industrie mytilicole de l'Î.-P.-É. : pratiques de l'industrie et incidence potentielle sur les invertébrés non ciblés

RÉSUMÉ

La mytiliculture sur l'Île-du-Prince-Édouard est aux prises avec des infestations de tuniciers, particulièrement l'ascidie plissée *Styela clava*. De façon périodique, les concessions aquacoles sont traitées avec de la chaux hydratée ($Ca(OH)_2$) afin de contrôler ces infestations et assurer une perte minimale des récoltes de moules. Ce traitement peut se faire de plusieurs façons, mais implique toujours le trempage des boudins de moules dans une suspension à concentration de 4 % de chaux hydratée, ou l'arrosage avec cette solution. L'excès de chaux produit par ces traitements peut être libéré directement dans le milieu aquatique environnant, ce qui soulève des questions concernant les effets biologiques potentiels sur les espèces indigènes non ciblées. Nous examinons ici l'état actuel des connaissances concernant le traitement à la chaux hydratée, la chimie et les effets biologiques connexes et les sous-produits engendrés par le traitement. Le traitement à la chaux hydratée entraîne une augmentation marquée, mais de courte durée, du pH et de la conversion du $Ca(OH)_2$ en carbonate, ce qui peut former un précipité dans l'environnement. Les études en laboratoire ont permis de caractériser les dangers associés à un pH élevé et à la présence de particules de carbonate. Les études sur le terrain ont démontré qu'il est peu probable que l'exposition à un pH de niveau dangereux ou à des particules de carbonate dure plus de quelques minutes; un pH élevé n'est observé que sur une distance de quelques mètres autour du lieu de traitement. Ces données nous permettent de conclure que dans les conditions d'exploitation actuelles, des effets létaux sur les organismes non ciblés résultant de l'utilisation de la chaux hydratée sont peu probables.

INTRODUCTION

Suspended culture of blue mussels (*Mytilus edulis*) in Prince Edward Island (PEI) produced close to 23,000 tonnes in 2013 with an estimated value of over 30 million Canadian dollars (DFO 2015). The industry, however, has been impacted by infestations of invasive species since 1998 (Government of PEI 2015). Proliferation of invasive tunicates such as the clubbed tunicate (*Styela clava*) in estuaries has necessitated the development of approaches for managing these fouling organisms. The primary goal of tunicate management in PEI is the removal of tunicates from aquaculture infrastructure, including mussel sleeves.

The primary treatment method involves either immersion or spraying with a saturated suspension of hydrated lime. Hydrated lime, chemical formula $Ca(OH)_2$, is a colourless crystal or white powder and is obtained when calcium oxide (quicklime) is slaked with water. It is soluble in water at 0.160 g/100 g (CRC 2005). Treatment with hydrated lime (calcium hydroxide) is effective against tunicates, but, as is the case whenever chemical compounds are applied directly to water, questions arise regarding the potential for deleterious effects to non-target organisms to occur concurrent with, or after treatment. Of particular concern in Malpeque Bay (PEI) is the effects on the American lobster (*Homarus americanus*).

Requests have been made to increase the leases and the production of mussels in Malpeque Bay, Prince Edward Island (DFO 2016). In support of continued consultations on the proposed lease expansion, DFO Aquaculture Management has asked for advice on whether the current use of lime in the mussel aquaculture industry changes significantly the environmental footprint of a mussel lease, specifically in the context of the expanded use of hydrated lime associated with proposed mussel lease expansions in Malpeque Bay.

This review paper was prepared to support the science peer review of the extent to which liming may change the footprint of mussel leases. The terms of reference for this review were:

- What are the current hydrated lime treatment methods (including concentration of lime, timing and duration of treatment) to control *Styela clava* and predators on mussel seed lines?
- What is known regarding changes in water quality after a lime treatment?
- Are the observed changes consistent across treatment methods?
- What is the duration of the changes in water quality parameters?
- What is known of benthic impacts following treatment with hydrated lime, e.g. is sediment chemistry changed and, if yes, for what duration?
- What are the known effects of hydrated lime on marine organisms? This includes lethal, sublethal and behavioural responses.
- What is known of effects of hydrated lime on indigenous species, particularly the American lobster?
- Do current treatment methods result in the potential for concentrations of lime or alterations in water or sediment chemistry to affect indigenous species?

Some research has been carried out in the lab and in the field to determine hazards associated with the use of hydrated lime as a pest control product in PEI mussel culture (Locke et al. 2009; Ramsay et al. 2014). The authors relied heavily on these documents as well as Burridge et al. (2010) wherein the pathways of effects associated with use of hydrated lime was reviewed.

CHEMISTRY AND FATE OF HYDRATED LIME

Canadian Council of the Ministers of the Environment (CCME) have developed water quality guidelines for the protection of aquatic life in marine environments (CCME 1999). Although the factsheet for pH was prepared in 1999 the guideline remains the same in 2016. These guidelines are science-based and, while determined using toxicity data, they recommend "safe" levels that are lower than reported lethal or toxic thresholds. The CCME water quality guideline for pH states:

> "The pH of marine and estuarine waters should fall within the range of 7.0-8.7 units unless it can be demonstrated that such a pH is a result of natural processes. Within this range, pH should not vary by more than 0.2 pH units from the natural pH expected at that time. Where pH is naturally outside that range, human activities should not cause pH to change by more than 0.2 pH units from the natural pH expected at that time, and any change should tend towards the recommended range."

Calcium carbonate ($CaCO_3$) is a common substance in rocks in all parts of the world and the main constituent of both limestone and shells in marine molluscs. When calcium carbonate is heated at high temperatures, carbon dioxide (CO_2) is released forming quicklime (CaO). Quicklime, the product of calcination of limestone, consists of the oxides of calcium (CaO) and magnesium (MgO) and other byproducts and the relative composition of these depends upon the source of the mineral (for example high calcium quicklime versus dolomitic quick lime; Fact Sheet Properties of Lime January 2007; accessed May 31, 2016). While the cation (Mg^{2+}) is different, the chemistry is the same with $MgCO_3$ being formed on addition to seawater (Fricker and Park 2013).

Hydrated lime is produced by "slaking" quicklime. Briefly water is added to quicklime and in this process CO_2 is released from the carbonate and the quicklime picks up hydroxide from water producing hydrated lime ($Ca(OH)_2$). The product is a colourless crystal or white powder. Hydrated lime is easily converted back to calcium carbonate ($CaCO_3$), in the presence of carbon dioxide (CO_2) in either air or in water. When hydrated lime reacts with carbon dioxide in the air the effectiveness of the product is reduced, meaning that hydrated lime has a shelf life (Ramsay et al. 2014). When hydrated lime is mixed in excess with water there is not sufficient CO_2 to convert all the lime. This results in release of hydroxide ions (OH^-), an alteration of the hydrogen ion concentration and therefore a change in pH.

Saturated hydrated lime suspensions have a pH of approximately 12.7. When $Mg(OH)_2$ or $Ca(OH)_2$ is exposed to CO_2 dissolved in seawater, $MgCO_3$ or $CaCO_3$ is also formed. The solubility of these carbonates is much lower than hydrated lime at 0.00066 g/100 g water for $CaCO_3$ (CRC 2005), and therefore has the potential to precipitate out of suspension and settle to the bottom in particulate form as the reaction occurs. This is easily observed during treatment of mussel sleeves (Ramsay et al. 2014). Calcium carbonate is a common substance in rocks in all parts of the world and is the main component in shells of marine organisms, and so is considered innocuous (Locke et al. 2009). Calcium carbonate in marine sediments can also be converted to bicarbonate $2(HCO_3-)$ and ionic calcium (Ca^{2+}) in the presence of dissolved CO_2 (University of Puerto Rico 2015).

LABORATORY BASED TOXICITY STUDIES OF HYDRATED LIME

There is a considerable body of literature regarding effects of lowering pH; in addition to causing direct effects due to increased acidity, it may make other toxicants more available, particularly in freshwater. The consequences of increased pH, particularly in the marine environment, is not as well studied. Long term exposure to increased pH can result in damage to scales and mucous layers with fish but acute lethality, when observed in response to elevated pH as with liming, is likely caused by damage to gills and disruption of cellular membranes. There may also be severe osmotic stress in “unprotected” species such as the tunicate (Loosanoff and Davis 1942).

Locke et al. (2009) conducted laboratory bioassays of hydrated lime with the bacterium *Vibrio fischeri*, sand shrimp (*Crangon septemspinos*a) and threespine stickleback (*Gasterosteus aculeatus*). Results of these lethality studies are shown in Table 1. These authors also reported No Observable Effect Concentrations (NOEC) for the stickleback and shrimp. NOEC refers to the concentration of the compound that results in <= 10% mortality over the duration of the experiment. These results are shown in Table 2. Only in 14 day experiments with shrimp is the NOEC, expressed as pH (8.17), close to reported normal pH as measured in Malpeque Bay.

Lobster larvae are known to be intolerant of quicklime (Loosanoff and Engle 1942) and the susceptibility to hydrated lime was reported in Burridge et al. 2010 (in reference to unpublished data by Doe et al. 2009). Stage III larval lobsters were exposed to a range of concentrations and exposure scenarios, including realistic short-term pulse exposures followed by transfers to clean water. Lethality results are presented in Table 1. Clearly high concentrations of lime and a consequent elevated pH are required to kill Stage III lobsters. In tests with more realistic exposure scenarios, short term or pulsed, higher concentrations and pH are required.

In a lab-based experiment Reebs et al. (2011) investigated responses of the sand shrimp, *Crangon septemspinosa*, when given a choice of settling on sediment that was carbonate-layered as a result of treatment with hydrated lime or untreated sand bottoms. The shrimp showed a preference for the untreated sand. In aquaria where lime treatment had taken place up to 75% of the shrimp died (Reebs et al. 2011). Carbonate was injected using saturated suspension of hydrated lime to mimic “real world” addition. A consequence of the exposure technique was a rapid and significant elevation of pH. The authors attribute mortality to the elevated pH and speculate that, while this work clearly shows a hazard to this marine invertebrate, there are a number of reasons to believe the risk may not be significant (Reebs et al. 2011). The observed precipitate is likely to remain undissolved in systems where there is no turnover or renewal of water as is the case in static lab-based assays. During operational treatments there is constant exposure to seawater as the lime suspension dissipates.

A 48-h exposure to 9.0 pH units was found to be lethal to *Crassostrea virginica* (eastern oyster) and *Mercenaria mercenaria* (hard clams) larvae under laboratory conditions (Calabrese and Davis 1966). Similarly, a 48-h exposure to 8.5 pH units lowered the percentage of *Mulinia lateralis* (coot clam) embryos that developed normally, and a 6-8 day exposure to 9.0 pH units decreased *M. lateralis* survival at the larval stage (Calabrese, 1970).

Doe et al. (2009) (unpublished work reported in Burridge et al. 2010) also reported that lobster (Stage IV) behaviour, as indicated by increased frequency of tail flicks which is a known stress response in lobsters (for example Chiasson et al. 2015), was affected by plumes of hydrated lime. Tail flicks decrease as particles settle to the bottom of the jars. Ramsay et al. (2014) report on unpublished work wherein exposure to $CaCO_3$ alone did not result in an increase in tails flicks but exposure to particles generated by addition of hydrated lime did. The authors suggest it is the “caustic” nature of the particles that the lobster are reacting to, i.e. the tail flicks are probably in response to encountering small particles of undissolved lime. Interestingly, Reebs et al. (2011) report that when they added hydrated lime to water, 80-85% of the resulting particles were magnesium carbonate. It is therefore possible that the Stage IV lobsters were being exposed to two different types of carbonate. As stated earlier, some hydrated lime products contain important proportions magnesium. In addition, magnesium ions are the third most abundant ions in seawater with only sodium and chloride ions being more abundant (Angelis 2005). It is therefore likely that liberated CO_3^{2-}, either from $Mg(OH)_2$ or $Ca(OH)_2$, will combine with available Mg in seawater to produce $MgCO_3$. Magnesium carbonate, like $CaCO_3$ is a white powder and considered to be an inert chemical in terms of biological activity.

TREATMENT METHODOLOGY

Mussel lines are treated with hydrated lime in one of three methods:

- Immersion bath where mussel sleeves, lines and associated infrastructure are pulled through a trough containing a saturated suspension of hydrated lime.
- Spraying a saturated suspension of hydrated lime on mussel sleeves, lines and associated infrastructure in an open system. Lime is pumped through a garden hose and hand-sprayed on the lines, etc. The effluent goes directly into the environment.
- Spraying a saturated suspension of hydrated lime on mussel sleeves, lines and associated infrastructure in a closed system. Lime is pumped through a number of low-pressure nozzles in a shower system. There is a recovery container to capture/recover the waste and the recovered solution is reused in the process.

Treatment is with a 4% suspension: 40 g of hydrated lime per 960 ml (approximately 1 L) of seawater (40,000 mg/L). Ramsay et al. (2014) describe the details of each treatment. While this information is of considerable interest in terms of production, from an ecotoxicology perspective the key parameter is the total input to the surrounding environment. This dictates how much hydrated lime, its by-products, or an "adjustment" of pH a non-target organism may experience, and for how long.

The rate of use of hydrated lime has been estimated at 1 to 2 bags (22.7 kg per bag) of lime used per 600 ft line (400 sleeves). Growers can treat 6-10 lines per day therefore use approximately 203 - 450 kg per day at one site (Ramsay et al. 2014). DFO (2010) report that lime treatment is effective in lowering numbers of invasive tunicates to manageable levels with approximately 90% mortality of treated tunicates.

FIELD STUDIES AND POST-TREATMENT OBSERVATIONS

FATE OF LIME (WATER AND SEDIMENT)

Natural pH levels in Malpeque Bay vary with season and temperature. Ramsay et al. (2014) report a number of pre lime treatment pH measurements from several years and these range from 7.43 to 8.1 pH units. However, there appear to be no studies that have systematically recorded the variations of pH within the Bay. These data would be interesting, particularly if they were collected to coincide the period of lime treatment.

Ramsay et al. (2014) report in situ measurements and observations made during operational lime treatments in PEI (Murray River, Malpeque Bay and Darnley Basin) in 2007, 2008 and 2009. The 2007 work assessed effluent from an immersion treatment while the work in 2008 and 2009 assessed effluent from an open spray operation. The pH of the saturated lime suspension ranged from 12.5 to 13.1. The “pH footprint” was established by pH measurement taken near the point of entry of lime to the aquatic environment. The measured pH readings were back to pre treatment levels at a distance no more than 2 metres from the point of entry. While there are few details regarding methodology, Doe et al. (2009, reported in Burridge et al. 2010) indicate that these were a series of surface measurements with hand-held pH meters.

In two treatments (Darnley Basin and Murray River) divers observed a cloud of lime particles in the water column immediately below the area where the treated sock exited the lime trough or close to the surface spray. The particles drifted to the sediment and appeared to be dissociated. The authors do not provide an indication of the time it takes to sink or to become dissociated (Ramsay et al. 2014). These authors state that in many years of underwater observation of sediment near mussel growing areas personnel from PEI Fisheries, Agriculture and Rural Development (FARD) have never observed lime deposits.

Filgueira et al. (2015) have reviewed the contribution of mussel shells in carbon cycling in bays where mussel culture takes place. It is clear from the discussion that whatever the carbonate input associated with hydrated lime treatment, it is inconsequential relative to biogenic calcification.

Comeau et al. (In review)[1] describe the seawater alkalinity footprint resulting from operational lime treatments in Malpeque Bay in 2013—2015. Factory-calibrated pH sondes (INW TempHion™ pH/ORP, WA, USA) were attached directly to fouled mussel sleeves destined for hydrated lime treatment. The measured pH values at the outside edge of treated mussel sleeves were highest (9.3 to 11.7) immediately after sleeves were returned into the water column (Figure 1a). Thereafter pH rapidly declined and returned to pre-treatment levels within 3.1 ± 0.5 min (std error) (range 0.3 to 10.5 min, n = 31 sleeves). Vertical profiling revealed that the pH footprint was mainly confined to a depth range of 1.0 - 3.0 m, which is consistent with sleeve length. The type of sprayer (manual vs automated) had no significant effect on the time required to meet the water quality guideline. Only the experimental units (boats) significantly altered the duration of the footprint, suggesting that the amount of hydrated lime released into the environment was largely governed by the specific activity (grower) and perhaps the level of tunicate infestation at the time of treatment. In addition, multiple confounding environmental variables may have been at play since the different methodologies were tested at different sites and also on different days.

Comeau et al. (in review)[1] observed increases in pH, measured by sensors deployed 15 cm above the seabed during actual lime treatments (Figure 1b). These events were generally within federal water quality guidelines (pH change less than 0.2 units), with measured changes above ambient levels of 0.02 to 0.48 pH units, with a maximum recorded absolute value of 8.4 pH units. The duration of these episodic events ranged from 2.4 to 126.0 min, with a mean duration of 36.8 ± 8.0 min (1 std error). The application method (manual vs automated) had no significant effect on these near-bottom pH signatures, either in terms of signal amplitude or duration.

Comeau et al. (in review)[1] also deployed a series of green AstroTurf carpets (0.9 m × 0.9 m) in the Malpeque study area. Underwater photos of these carpets were taken before and after treatments with the objective of assessing whether calcium hydroxide aggregates, which are characteristically white in colour, deposited onto the bottom. They found no evidence that limestone particles or flakes were precipitating onto the seabed when hydrated lime was being applied in mussel farms. Only mussel feces were clearly detectable on the photos. The underlying reason for this result was unclear. From laboratory trials, they calculated that lime particles fall slowly (0.36 ± 0.03 cm s^{-1}) compared to the documented velocities (0.27 - 1.81 cm s^{-1}) for mussel feces (Callier et al. 2006). Depositional modelling indicated that the fine particles may have been carried over distances > 100 m and consequently may have been

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[1] Comeau, L.A., Sonier, R., Guyondet, T., Landry, T., Ramsay, A., and Davidson, J. (In review). Behavioural response of bivalve molluscs to calcium hydroxide. Submitted to Aquaculture.

deposited outside the carpeted study area. Therefore the lack of evidence that lime particles are accumulating in the bottom sediments may be due to methodology.

At the Malpeque Bay-scale, Comeau et al. (In review)[1] estimated the collective effort needed to manually-spray all sleeves at approximately 96.5 days (926,212 sleeves (Filgueira et al., 2015) times 400 sleeves h^{-1} (Ramsay et al., 2014) or approximately 6.4 days per leaseholder). Assuming a two-metre horizontal dispersal range (Ramsay et al. 2014) and a water renewal time of approximately 41 days (Bacher et al. 2016), tunicate control measures may briefly (approximately 3.1 min) increase alkalinity in approximately 0.48% of the Malpeque Bay volume. This conservative assessment is consistent with previous mass balance calculations for Malpeque Bay (Locke et al. 2009), which led the authors to conclude that the system has the capacity to absorb pH changes at the current level of aquaculture.

FIELD EXPOSURES OF NON-TARGET ORGANISMS

Ramsay et al. (2014) report on in situ tests wherein non-target organisms were exposed to the effluent from operational lime treatments. A number of fish and invertebrate species were exposed in a variety of cages and placed in the effluent plume from a treatment then subsequently tied to mussel sleeves for post-treatment monitoring. In only one case was there loss of experimental animals coincident with lime treatment. In an exposure conducted in November 2008, 11.1% of the exposed rock crabs (*Cancer irroratus*) were dead two weeks post treatment. The authors do not discuss this result in their report and rock crabs were not used in any other field studies.

Locke (2008) reviewed the scientific literature on the alkaline tolerances of marine and estuarine organisms and concluded that “most pH-related impacts on survival, growth, photosynthesis, feeding and immune response occur at pH levels outside the [CCME] recommended range [7.0 and 8.7 pH units]”. Comeau et al. (In review)[1] postulated that the alkalinity footprints caused by tunicate treatments probably have no or little detrimental effects on drifting planktonic larvae of molluscs or other commercially-important marine invertebrates. High concentrations over extended periods are required to kill such larvae.

Finally, the Comeau et al. paper gives particular attention to the behavioural response of bivalves during hydrated lime exposure. Their premise was that atypical valve activity, such as a tendency towards the closure of valves, is indicative of physiological stress. *Mytilus edulis* (mussel), *Crassostrea virginica* (eastern oyster) and *Argopecten irradians* (bay scallop) challenged to near-bottom like alkalinity fluctuations (≤ 9.2 pH units) consistently responded by completely or partially closing their valves. These experimental bivalves were subjected to slightly higher alkaline exposures (maximum of 9.2 pH units for 3 hours) than were recorded in the benthic environment when the mussel growers were treating infested sleeves (maximum of 8.4 pH units for 2 hours). In their pioneering work on calcium oxide, Loosanoff and Engle (1942) exposed three species (*C. virginica, M. edulis and Mercenaria mercenaria*) to alkaline environments (pH 8.6 to 9.5) for prolonged periods of time, ranging from 2 to 8 days. They reported that shell movements became normal “soon” after the lengthy exposures. Comeau et al. (In review)[1] found that the behavioural response was generally confined to the treatment period and was of short duration, ranging from 0.2 to 4.7 h. No bivalve mortality was detected 14 days following the hydrated lime exposures. Similarly, exposures to concentrated calcium oxide did not kill nor noticeably injure *C. virginica, M. mercenaria, M. edulis* and other mollusks (Needler 1940; Loosanoff and Engle 1942; Loosanoff 1961; Shumway et al. 1988). Turner (1970) stated that, while irritating to soft-shelled clams (*Mya arenaria)*, calcium oxide pellets had no impact on survival rates and normal feeding activity was interrupted for only a “few hours”.

DISCUSSION

Seawater has a significant capacity to buffer pH changes. So pH changes are often transient as is the case with the elevation observed during lime treatment. The risk posed to non-target organisms by use of hydrated lime depends on hazard (toxicity) and exposure. The work by Locke et al. (2009), Reebs et al. (2011) and Doe et al. (reported in Burridge et al. 2010) as well as the field studies reported by Ramsay et al. (2014) all involve exposures that could be argued are not representative of real world scenarios. Even the loss of rock crab after exposure during operational treatments should be considered in context. This species is a benthic invertebrate living on the bottom. As such it is unlikely to ever be exposed to elevated pH or lime concentrations associated with surface treatment. Published results from lab-studies show that pH must be elevated for a matter of hours or days in order for lethality to occur. Data from field measurements clearly show pH elevation to be of short duration (minutes). Therefore it seems unlikely that effects on non-target organisms will occur during operational treatments based on the current use pattern.

There remains some uncertainty regarding the fate of carbonate in sediments. Burridge et al. (2010) stated: To our knowledge there have been no studies in the field to examine in a quantitative manner the deposit and accumulation over time of lime in bottom sediments in the vicinity of mussel aquaculture sites treated with lime for control of tunicates. There have been no reported studies on the toxicity of hydrated lime incorporated in bottom sediments to sediment dwelling organisms. There are no CCME sediment quality guidelines for lime (CCME 1999). To this author's knowledge the situation remains the same in 2015.

Calcium carbonate may be persistent indicating the potential for sediment accumulation with continued use. However, in the presence of CO_2, calcium carbonate may be converted to bi-carbonate and calcium (University of Puerto Rico, electronic citation accessed 2015). Cycling of CO_2, particularly that associated with mussel culture (see, for example, Filgueira et al. 2015) and tidal turnover may serve to maintain dissolved CO_2 and thus reducing the calcium carbonate footprint.

Locke et al. (2009) have stated that use of hydrated lime may serve to counteract any negative effects expected to occur as a result of ocean acidification although given the extent of the pH change estimated to occur in Malpeque Bay during one year (minutes over a fraction of the total volume of water), the benefits of liming to mitigating ocean acidification are minimal. Keppel et al. (2012) have shown that growth of juvenile lobsters is negatively affected by lower pH in seawater. The effect is associated with a reduction of available carbonate. As discussed throughout this paper treatment with hydrated lime results in deposition and dissolution of carbonate indicating that a reduction in available carbonate will not occur as a result of liming operations.

All available lethality data show that the pH must be elevated in order for non-target invertebrates to be affected. The absolute value of pH and the duration of the elevation required for biological effects are higher and much longer than would be experienced in any normal treatment scenario and the risk to non-target organisms is considered negligible. However, an unknown quantity of hydrated lime is being released directly into the marine environment as a result of this practice and concerns may remain in some quarters. There are several relatively simple approaches to reducing the potential risk of effects on non-target organsims and in particular, lobsters:

- Determine the extent to which the areas where mussel aquaculture takes place serve as settlement and nursery areas for young lobsters. These areas should be avoided when considering new leases. Lobster larvae are distributed in coastal PEI waters from early or mid-June to mid-September (Harding et al. 1982; Scarratt 1964), but what proportion of the population occurs inside the estuaries is unknown.

- Preferrably treat mussel lines and sleeves using the treatment booth with low pressure spraying and collection of treatment effluent as this reduces the total amount of lime required.
- Dispose of all unused or excess hydrated lime (solid or solutions) in a land-based facility or an area where the potential interactions with non-target organisms is low.

From a regulatory perspective, public confidence could also be bolstered by a requirement of the industry to report quantities of hydrated lime applied by site or by bay. Complete reporting of this data would aid in assessing the overall risk to the receiving environment posed by the use of this product under current and proposed expansions of the industry.

REFERENCES CITED

Angelis, M.D. 2005. Major ions in Seawater. Water Encyclopedia 4: 159 - 160.

Bacher, C., Filgueira, R., and Guyondet, T. 2016. Probabilistic approach of water residence time and connectivity using Markov chains with application to tidal embayments. J. Mar. Systems 153: 25 – 41.

Burridge, L.E., Doe, K.G., and Ernst, W. 2010. Pathway of effects of chemical inputs from the aquaculture activities in Canada. DFO Can. Sci. Adv. Sec. Res. Doc. 2010/017. vi + 63 p.

Calabrese, A. 1970. The pH tolerance of embryos and larvae of the coot clam, *Mulina lateralis* (Say). Veliger 13: 122 – 126.

Calabrese, A., and Davis, H.C. 1966. The pH tolerance of embryos and larvae of *Mercenaria mercenaria* and *Crassostrea virginica*. Biol. Bull. (Woods Hole) 131: 427 – 436.

Callier, M.D., Weise, A.M., McKindsey, C.W., and Desrosiers, G. 2006. Sedimentation rates in a suspended mussel farm (Great-Entry Lagoon, Canada): biodeposit production and dispersion. Mar. Ecol. Prog. Ser. 322: 129 – 141.

Canadian Council of Ministers of the Environment (CCME). 1999. Canadian water quality guidelines for the protection of aquatic life: pH (marine). In: Canadian environmental quality guidelines, 1999, Canadian Council of Ministers of the Environment, Winnipeg. (accessed May 31, 2016)

Chiasson, M., Miron, G., Daoud, D., and Mallet, M.D. 2015. Effect of Temperature on the Behaviour of Stage IV American Lobster (*Homarus americanus*) Larvae. J. Shell. Res. 34(2): 545 - 554.

Chemical Rubber Publishing Company (CRC). 2005. Handbook of Chemistry and Physics. 86th Edition. CRC Press, Taylor and Francis Group, Boca Raton, FL, USA.

DFO. 2010. Containment and Mitigation of Nuisance Tunicates on Prince Edward Island to Improve Mussel Farm Productivity. Aquaculture Collaborative Research and Development Program (ACRDP) Fact Sheet. Issue 6 May, 2010. (Accessed May 31, 2016)

DFO. 2016. Review of potential impacts of hydrated lime treatments associated with proposed expansion of mussel production in Malpeque Bay, PEI. DFO Can. Sci. Advis. Sec. Sci. Advis. Rep. 2016/014.

Filgueira, R., Byron, C.J., Comeau, L.A., Costa-Pierce, B., Cranford, P.J., Ferreira, G., Grant, J., Guyondet, T., Jansen, H.M., Landry, T., McKindsey, C.W., Petersen, J.K., Reid, G.K., Robinson, S.M.C., Smaal, A., Sonier, R., Strand, Ø., and Strohmeier, T. 2015. An integrated ecosystem approach for assessing the potential role of cultivated bivalve shells as part of the carbon trading system. Mar. Ecol. Prog. Ser. 518: 218 - 287.

Fricker, K.J., and Park, A.-H.A. 2013. Effect of H_2O on $Mg(OH)_2$ carbonation pathways for combined CO_2 capture and storage. Chemical Engineering Science 100: 332 - 341.

Harding, G.C., Vass, W.P., and Drinkwater, K.F. 1982. Aspects of larval American lobster (*Homarus americanus*) ecology in St. Georges Bay, Nova Scotia. Can. J. Fish. Aquat. Sci. 39: 1117 - 1129.

Keppel, E.A, Scrosati, R.A., and Courtenay S.C. 2012. Ocean acidification decreases growth and development in American lobster (*Homarus americanus*) larvae. J. Northwest Atl. Fish. Sci. 4: 61 - 66.

Locke, A. 2008. Tabulated observations of the pH tolerance of marine and estuarine biota. Can. Manus. Rep. Fish. Aquat. Sci. No. 2857, iv + 28 p.

Locke, A., Doe, K.G., Fairchild, W.L., Jackman, P.M., and Reese, E.J. 2009. Preliminary evaluation of effects of invasive tunicate management with acetic acid and calcium hydroxide on non-target marine organisms in Prince Edward Island, Canada. Aquat. Inv. 4(1): 221 - 236.

Loosanoff, V.L. 1961. Biology and methods of controlling the starfish, Asterias forbesi (DESOR). United States Department of the Interior, Bureau of Commercial Fisheries, U.S. Fish and Wildlife Service, Milford, Connecticut, Fishery Leaflet 520.

Loosanoff, V.L., and Engle, J.B. 1942. Use of lime in controlling starfish. U S Fish and Wildlife Service Research Report 2: 1 - 29.

Needler, A.W. 1940. Use of quicklime for killing starfish on oyster grounds. Fisheries Research Board of Canada, Ottawa. Oyster Farming Circular 40-FRB-OFCII.

McKindsey, C.W., Anderson, M.R., Barnes, P., Courtenay, S., Landry, T., and Skinner, M. 2006. Effects of shellfish aquaculture on fish habitat. DFO Can. Sci. Adv. Sec. Res. Doc. 2006/011. viii + 84 p.

Ramsay, A., Gill, K., Morrison, A., and MacNair, N. 2014. Hydrated lime application by the PEI aquaculture industry. PEI Department of Fisheries, Aquaculture and Rural Development, Technical Report 253. iv + 28 p. (accessed May 31, 2016)

Reebs, S.G., Jackman, P.M., Locke A., and Fairchild, W.L. 2011. Avoidance by sand shrimp, *Crangon septemspinosa*, of sandy patches covered by hydrated lime (calcium hydroxide) deposits. Can. Tech. Rep. Fish. Aquat. Sci. No. 2938: iv + 7 p.

Scarratt, D.J. 1964. Abundance and distribution of lobster larvae (*Homarus americanus*) in Northumberland Strait. J. Fish. Res. Board Can. 21: 661 - 680.

Shumway, S.E., Card, D., Getchell, R., and Newell, C.R. 1988. Effects of calcium oxide (quicklime) on non-target organisms in mussel beds. Bull. Env. Cont. and Toxic. 40: 503 – 509.

Turner, R.L. 1970. Quicklime: effects on soft-bodied marine organisms. Science 168: 606 – 607.

TABLES

Table 1. Results of toxicity tests of hydrated lime suspensions on non-target organisms.

Species tested	Endpoint	Threshold mean (95% C.I.) mg·L^{-1}	Threshold mean (95% C.I.) pH	Reference
Vibrio fischeri	15 min IC50	31 (CI :18.8 – 51.4)	9.0 (no CI)	Locke et al. (2009)
Crangon septemspinosa	96 h LC50	158 (CI : 50 – 500) -	9.7 (CI : 9.1 – 10.3	Locke et al. (2009)
Crangon septemspinosa	14 d LC50	53.1 (CI : 48.3 – 58.4)	9.2 (CI : 9.1 – 9.3)	Locke et al. (2009)
Gasterosteus aculeatus	96 h LC50	457 (CI : 262 – 785)	10.47 (CI : 10.26 – 10.52)	Locke et al. (2009)
Homarus americanus Stage III	96 h LC50	121 (CI: 73.5 - 198)	9.7: (CI: 9.5-10.0)	Doe (2009) reported in Burridge et al. (2011)
Homarus americanus Stage IV	96 h LC50	998 (CI: 620 – 1610)	10.3 (CI: 10.0 – 10.5)	P. Jackman (EC, unpubl. data, 2016)
Homarus americanus Stage III	LC50 after one hour pulse, followed by 12 days in clean seawater	965 (CI: 633 – 1,470)	10.6 (CI: 10.2 – 11.0)	Doe (2009) reported in Burridge et al. (2011)
Homarus americanus Stage III	LC50 after a one hour pulse on three consecutive days, followed by 9 days in clean seawater	606 (CI: 336 – 1,090)	10.5 (CI: 10.1 – 10.9)	Doe (2009) reported in Burridge et al. (2011)

Table 2. No Observable Effects Concentrations (NOEC) from Locke et al. (2009).

Species tested	Endpoint	NOEC (mg·L-1)	NOEC (pH)
Crangon septemspinosa	96 h LC50	100	9.54
Crangon septemspinosa	14 d LC50	50	8.58
Gasterosteus aculeatus	96 h LC50	32	8.17

FIGURE

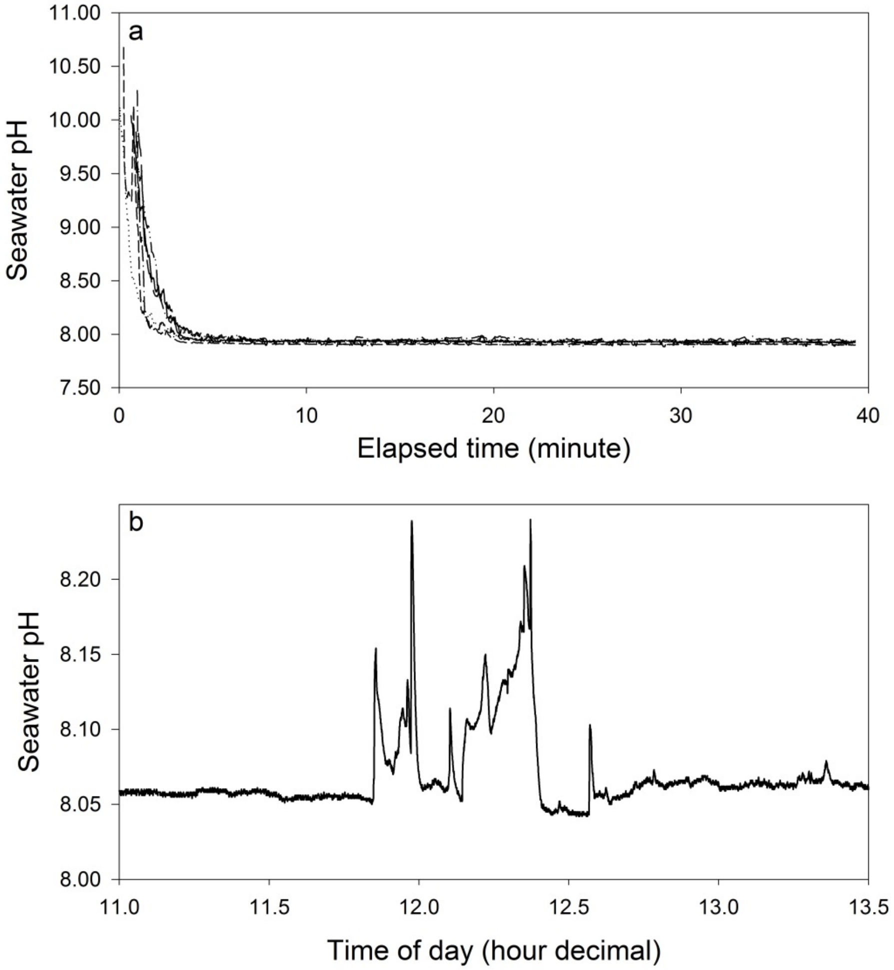

Figure 1. (a – upper panel) Examples of exponential decay of alkalinity at the edge of treated mussel sleeves. (b – lower panel) Example of pH fluctuation above the seabed during the treatment of nearby (< 90 m) mussel sleeves. Taken from Comeau et al. (In Review)[1].