EFFECT OF INORGANIC SALTS, SOAPS AND DETERGENTS ON DISSOLUTION AND LARVICIDAL ACTIVITY OF ALGINATE FORMULATION OF *BACILLUS SPHAERICUS*

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Abstract. Various inorganic salts and commonly used soaps and detergents were tested in the laboratory for their effect on the dissolution and larvicidal and residual activity of a slow-release alginate encapsulated granular formulation of *Bacillus sphaericus*. Fluoride, chloride and sulphate salts and a detergent powder affected the residual activity of this formulation drastically by rupturing it but did not effect its larvicidal activity. Nitrates and phosphates of sodium and potassium also had the same effect but to a moderate level. The safest concentration of these water impurities for effective functioning of the alginate encapsulated *B. sphaericus* formulation have been determined.

INTRODUCTION

A variety of factors associated with the aqua of the mosquito breeding habitats have a marked influence on the activity of biopesticides (Lacey and Undeen, 1986). Of these, the ionic composition is more important (Lacey, 1985; Sutherland and Khoo, 1987). Biopesticides applied in the form of alginate encapsulated formulations have been reported to be effective in controlling mosquito breeding (Kuppusamy *et al,* 1987; Desai and Shethna, 1989). The mode of release of the active ingredients from such formulations has been found to be by a gradual dissolution of the alginate matrix due to the action of water (Bashan, 1986). Depending on the quality of water in the treated habitats, the duration of residual activity of these formulations have been reported to vary from 51 to 107 days (Pradeepkumar *et al,* 1988; Arunachalam *et al,* 1991a,b; Rajendran *et al,* 1991). However, no direct information is available on the effect of various inorganic salts, soaps and detergents present in different mosquito breeding habitats on the properties and larvicidal activity of such formulations. Therefore, a laboratory study was undertaken to understand the effect of such chemicals on the dissolution and larvicidal activity of a slow-release alginate encapsulated granular formulation of *Bacillus sphaericus.*

MATERIALS AND METHODS

Fourteen different inorganic salts, $CaCO_3$, $MgSO_4$, NaCl, $NaHCO_3$, $NaNO_3$, NaH_2PO_4, KCl, KNO_3, KH_2PO_4, $(NH_4)_2SO_4$, $FeCl_3$, $FeSO_4$, $MnCl_2$ and NaF, cosmetic soaps with the trade names Liril, Hamam, Lifebuoy and Sandoor, washing soaps with the trade names Rin, Wheel and Trilo and a detergent powder with the trade name Surf were tested. The preliminary testing was carried out at 250 and 500 ppm concentrations, in distilled water. To disposable plastic cups containing 250 ml of various salt solutions were individually added 50 III instar *Culex quinquefasciatus* larvae. A perforated polypropylene sachet containing 50 mg of a slow-release granular alginate formulation of *B. sphaericus,* developed at the Vector Control Research Centre, Pondicherry (Kuppusamy *et al,* 1987), was then placed in each cup. Parallel cups containing distilled water but without the formulation served as controls. The experiment was replicated twice. The cups were held at 28°C and added with sterile yeast powder and dog biscuit, as larval food. Larval mortality was scored after 24 hours, corrected to control mortality, if any, and the percentage mortality was calculated using Abbott's formula (Abbott, 1925). After every day's observation, the sachets were transferred to a fresh set of cups containing healthy larvae as stated above. The experiment was continued until the formulation was dissolved completely. The mortality profile obtained with each treatment was compared with that of distilled water control and the safest level (*ie,* the concentration at which the residual activity of the formulation and/or its effectiveness are not affected) of each one was determined as detailed above with varying concentrations in quadruplicate.

RESULTS AND DISCUSSION

Fourteen different inorganic salts which are commonly encountered in different aquatic habitats and 4 each of cosmetic and washing soaps including a detergent powder which find their way into domestic effeuents were tested for their effect on the dissolution and larvicidal activity of an alginate encapsulated granular formulation of *B. sphaericus* (Kuppusamy *et al*, 1987), which releases the active ingredients in a sustained manner by getting dissolved in water gradually, leading to prolonged larvicidal activity. The results obtained indicate that all the materials tested had no adverse effect on the larvicidal activity of the formulation even at the highest concentration of 500 ppm (Table 1). The

Table 1

Effect of inorganic salts, soaps and detergents on larvicidal and residual activity of alginate formulation of *B. sphaericus*.

Chemical	% Mortality of III instar *Cx. quinquefasciatus* larvae at different concentration of chemicals[a]					
	50	100	150	200	250	500 ppm
Distilled H_2O (Control)	89					
Salts:						
$(NH_4)_2 SO_4$	91	89	98[b]	99[b]	100[c]	100[c]
$CaCO_3$	89	86	90	92	90	89
$FeCl_3$	86	90	89[b]	97[b]	99[b]	100[c]
$FeSO_4$	86	89	99	99[b]	100[c]	100[c]
$MgSO_4$	92	88	89	98[b]	98[b]	100[c]
$MnCl_2$	90	91	88	89	90	90
KCl	92	88	89	99[b]	97[b]	100[c]
KH_2PO_4	91	93	92	86	99[b]	97[b]
KNO_3	85	90	88	91	99[b]	99[b]
$NaHCO_3$	90	86	84	93	91	90
NaCl	86	92	88	98[b]	98[b]	100[c]
NaH_2PO_4	88	90	86	85	97[b]	99[b]
NaF	88	98[b]	99[b]	100[c]	100[c]	100[c]
NaNO	93	91	90	89	98[b]	98[b]
Soaps:						
Hamam	89	90	91	87	84	89
Lifebuoy	90	91	90	88	87	92
Liril	93	84	87	92	90	91
Sandoor	91	89	93	88	85	88
Detergents:						
Rin	92	91	83	87	91	87
Surf (powder)	84	89	99[b]	97[b]	100[c]	100[c]
Trilo	86	92	88	89	87	91
Wheel	88	88	93	86	90	89

a - mean value of 32 observations recorded over 2 months
b - mean value of 16 observations recorded over 4 weeks
c - mean value of 8 observations recorded over 2 weeks

larvicidal activity of the formulation observed with all the test solutions were almost comparable to that of distilled water. However, some of the salts and the detergent powder caused rapid dissolution of the formulation and thus reduced the duration of residual activity (Table 2). The salt, NaF ruptured the formulation completely within 2 weeks at the concentration of 250 ppm. And in contrast to this, the formulation remained intact for more than 2 months in distilled water. The detergent powder, Surf as well as chloride and sulphate salts such as NaCl, KCl and $FeCl_3$ and $(NH_4)_2SO_4$, $FeSO_4$ and $MgSO_4$ also caused a significant fragility in the formulation within a period of a week at 250 ppm. Nitrates and phosphates of sodium and potassium slightly increased the size of the formulation when compared to that of distilled water. The other salts, $CaCO_3$, $MnCl_2$ and $NaHCO_3$ and cosmetic and washing soaps did not affect the texture of the formulation even at the concentration of 500 ppm.

The safest concentration (critical level) of all these materials as determined in the present study are presented in Table 3. The data indicate that the critical level of sodium fluoride, Surf, chlorides

Table 2

Effect of inorganic salts, soaps and detergents on dissolution of alginate formulation of *B. sphaericus*.

Chemical	Nature of the formulation at different concentration of chemicals					
	50	100	150	200	250	500 ppm
Salts:						
$(NH_4)_2 SO_4$	N	N	F	F	R	R
$CaCO_3$	N	N	N	N	N	N
$FeCl_3$	N	N	N	F	F	R
$FeSO_4$	N	N	F	F	R	R
$MgSO_4$	N	N	N	F	F	R
$MnCl_2$	N	N	N	N	N	N
KCl	N	N	N	F	F	R
KH_2PO_4	N	N	N	N	F	F
KNO_3	N	N	N	N	F	F
$NaHCO_3$	N	N	N	N	N	N
NaCl	N	N	N	F	F	R
NaH_2PO_4	N	N	N	N	F	F
NaF	N	F	F	R	R	R
$NaNO_3$	N	N	N	N	F	F
Soaps:						
Hamam	N	N	N	N	N	N
Lifebuoy	N	N	N	N	N	N
Liril	N	N	N	N	N	N
Sandoor	N	N	N	N	N	N
Detergents:						
Rin	N	N	N	N	N	N
Surf (powder)	N	N	F	F	R	R
Trilo	N	N	N	N	N	N
Wheel	N	N	N	N	N	N

N - Normal; F - Fragile; R - Ruptured.

Table 3

Level of inorganic salts, soaps and detergents safest for effective functioning of alginate formulation of *B. sphaericus*.

Chemical	Concentration, ppm
Salts:	
$(NH_4)_2 SO_4$	100
$CaCO_3$	500
$FeCl_3$	150
$FeSO_4$	100
$MgSO_4$	150
$MnCl_2$	500
KCl	150
KH_2PO_4	200
KNO_3	200
$NaHCO_3$	500
NaCl	150
NaH_2PO_4	200
NaF	50
$NaNO_3$	200
Soaps:	
Hamam	500
Lifebuoy	500
Liril	500
Sandoor	500
Detergents:	
Rin	500
Surf (powder)	100
Trilo	500
Wheel	500

(except $MnCl_2$) and sulphates is very low (50-150 ppm) and hence it is likely that the alginate encapsulated formulation of *B. sphaericus* may not have prolonged residual activity if applied to habitats in which these chemicals are abundant. When compared to these chemicals, the presence of phosphates and nitrates (critical level: 200 ppm) in such habitats is likely to moderately affect the duration of residual activity whereas the presence of cosmetic and washing soaps (critical level : 500 ppm) is likely to be quite safe. Thus, the critical level of all these materials shuld be taken note of while evaluating alginate based formulations of *B. sphaericus* or any other bacterial agent for prolonged residual activity.

Habitats like back waters, coir pits and casuarina pits have been reported to contain high level of chloride (400-58,000 ppm) and sulphates up to 1,300 ppm (Geetha Bai *et al*, 1984; Reuben *et al*, 1984) and therefore, in such habitats the life span of alginate based formulation is likely to be shortened. Cess pools and cess pits, the predominant breeding sites of *Cx. quinquefasciatus* mosquitos, receiving domestic sullage will also reduce the life span of such formulations because they are likely to contain higher concentrations of surf as indicated in the present study.

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