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Efficacy of sodium hypochlorite in controlling viral and bacterial diseases in muga silkworm, Antheraea assamensis Helfer.

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ABSTRACT

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INTRODUCTION 1.

The muga silkworm Antheraea assamensis Helfer is a holometabolus sericigenous insect endemic to North East region of India only. This silkworm produces the unique golden colour silk which is more durable and has high demand in the global market. This silkworm is polyphagous in nature and feeds on a wide range of host plants [1]. Among the food plants, 'som' Persea bombycina Kost and 'soalu' Litsea monopetala Juss, are the two major primary food plants. Out of these two primary food plants 'soalu' Litsea monopetala Juss is semi-deciduous in nature while the other is evergreen. The muga silkworm is multivoltine in nature and is generally reared six crops in a year. Annually, India produces about 126 tons of muga raw silk. Due to outdoor nature of rearing muga silk worms are exposed to various rigors of changing environment of the varied topography of the region and thus the silkworm is prone to various diseases leading to heavy loss in production [2, 3]. Outbreak of various diseases viz., pebrine, flacherie, muscurdine and grasserie are the major constrains encountered in muga industry. Out of these diseases, flacherie and grasserie caused by virus and bacteria occurred mostly in every season and thereby causing death of considerable number of larvae which ultimately affects the cocoon silk production. The pathogens primarily infect the alimentary tract of the silk worm, but high infection may affect

The incidence of the viral and bacterial diseases viz. flacherie and grasserie caused large mortality to the muga silkworm, Antheraea assamensis Helfer thereby affected the cocoon silk production. It was observed that the application of 0.01 % of NaOCl to the silkworms at larval instars, reduced the mortality due to bacterial and viral diseases quite significantly (P < 0.1 & P < 0.05) as compared to the control without affecting the quality of cocoons during different seasons at the different locations. The values of Effective Rate of Rearing (E.R.R. %) in the treated lots was significantly higher (P < 0.1 & P < 0.05) than that of control in all the crop seasons at the different locations. The results of the present study deduced that the application of 0.01 % of Sodium hypochlorite during the rearing of muga silkworm, Antheraea assamensis Helfer can effectively control the larval mortality due to bacterial and viral diseases thereby enhance the cocoon production.

> blood cells, hypodermis and fat bodies too. Even after providing quality foliage and suitable environmental conditions during the rearing, in every crop about 14-40% crop loss normally occurred due to incidence of such diseases [4]. Disinfection and maintenance of hygienic condition during rearing are essential factors for preventing occurrence of diseases. Various disinfectants namely, formalin [5], bleaching powder [6, 7], Calcium hydroxide [8] and Chlorine dioxide [9] have been effectively used to minimize disease incidences. Besides, different antibiotics viz. Triazoles, Triadime, Streptomycine sulphate, Gentamycine, Cloxacillin, Kanamycine, Chloram-phenicol, Tetracycline and Aureofungin were used for control of diseases in mulberry silkworm, Bombyx mori [10,11,12,13]. Sodium hypochlorite as egg surface sterilant was also reported in B. mori [14]. The efficacy of Sodium hypochlorite in controlling mortality due to bacterial and viral diseases has been reported in Antheraea mylitta [15]. Considering the effectiveness of Sodium hypochlorite as disinfectant and control of certain diseases in seri-culture, the present study was taken up to observe the efficacy of this chemical against viral and bacterial diseases in A. assamensis Helfer.

MATERIALS AND METHODS

Disease free layings (dfls) of Muga Silkworm Antheraea assamensis Helfer were obtained from the Silkworm Improvement Division, Central Muga Eri Research & Training Institute, Jorhat, Assam (India). The rearing was conducted in outdoor on the foliages of som, Persea bombycina Kost. and Soalu, Litsea monopetala Juss

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at two separate locations viz. Germplasm Conservation Centre (GCC), Chenijan, Assam and Farm No. 03, Central Muga Eri and 10. Research & Training Institute, Lahdoigarh, Assam (India) for three seasons viz. Aherua (June-July), Jarua (January-February) and Chatua (February-March) during 2011 and 2012. During these seasons, higher mortality of worms usually occurred due to incidence of viral and bacterial diseases. Four different concentrations of NaOCl solution viz. 0.001 %, 0.01 %, 0.05 % and 0.1 % were sprayed with the help of the foot sprayer on the foliages once before 2-3 days of rearing, then once each at 2^{nd} , 3^{rd} during 2 and 4^{th} instars and twice at 5^{th} instars on the body of the larvae along with the foliages during the rearing. The silkworm larvae was recerted with the treated leaves. The concentration 0.01 % was not so effective against the control of diseases. The concentration 0.01

% were found to be corrosive to the leaves while 0.001 % was not so effective against the control of diseases. The concentration 0.01 % was found to be more effective in controlling the bacterial and viral diseases and more safe to the leaves. Therefore, 0.01 % of NaOCl was sprayed on the body of the larvae and foliages during the rearing. Each treatment comprised of 4 replications with 500 worms in each replication which were hatched on the same day. The experiment was conducted in a completely randomised design in two different locations for two consecutive years. In the control lot also same numbers of replications with same number of worms were maintained and only distilled water was sprayed on them. Data of mortality of worms due to diseases, effective rate of rearing (ERR %), cocoon weight and shell weight were recorded.

Effective Rate of Rearing (ERR %) was calculated as follows;

ERR % =
$$\frac{\text{Number of larvae reared}}{\text{Number of Cocoons formed}} \times 10^{-10}$$

The recorded data were analyzed statistically and significant differences of the treatment means were compared by using the statistical package SSP (Version 1.0.0.0.).

RESULTS AND DISCUSSION

Among the different concentrations of NaOCl viz., 0.01 %, 0.05% and 0.1 %, the first concentration (0.01%) was found more effective in controlling the mortality due to bacterial and viral diseases (Table I). However, the last two concentrations have shown corrosive action to the leaves and the treated larvae at these concentrations produce low quality cocoons. The larvae treated with concentration of 0.001 % do not show much reduction of mortality as compared to the control. Therefore, 0.01 % of NaOCl was selected for multilocational testing of its efficacy in controlling the mortality due to bacterial and viral diseases in the golden Indian muga silkworm, *Antheraea assamensis* Helfer since it did not show any adverse effect on the quality of the cocoons.

It was observed that when 0.01 % of NaOCl was applied to the silkworms, the mortality due to bacterial and viral diseases reduced quite significantly (P < 0.1 & P < 0.05) as compared to the control without affecting the quality of cocoons during different seasons at the different locations (Table II and III). During Jarua Crop (December-January), the mortality in the NaOCl treated lots at GCC, Chenijan were recorded as 12.50 % during 2011 and 8.55 % during 2012 while at farm No. 3, the

mortality during this season were recorded as 10.23 % during 2011 and 10.25 % during 2012 which were significantly lower as compared to the control lots. Significant difference on the quality of cocoon and cocoon shell were not observed between treated and control lots as shown in Table II and III. Larval mortality due to Flacherie disease accounts for major crop loss in muga culture. The larval mortality due to Flacherie during Jarua crop was recorded as high as 92. 66 % [16]. During Chatua Crop (February-March), the mortality in the treated lots were recorded as 8.54 % during 2011 and 9.75 % during 2012 at GCC, Chenijan while it was recorded as 5.62 % and 5.55 % respectively at Farm No. 3, Lahdoigarh which were significantly (P < 0.1 & P < 0.05) lower as compared to the control lots. It is evident from Table II & III that there was no adverse effect on the quality of cocoon and cocoon shell due to the treatment as compared to the control. Similarly, during Aherua Crop (June-July), the mortality in the treated lots were significantly (P < 0.1 & P < 0.05) reduced to 4.82 % against 24.64 % of control during 2011 and 12.44 % against 30.22 % of control during 2012 at GCC, Chenijan while at Farm No. 3, mortality was recorded as 4.52 % against 30.88 % of control and 7.83 % against 34.82 % in control respectively during the two years showing significant (P < 0.1 & P < 0.05) reduction of the mortality after the application of NaOCl solution. The values of E.R.R. % in the treated lots were significantly (P < 0.1 & P < 0.05) higher than that of control in all the crop seasons and years at the different locations (Table-II & III). As evident from Table-II & III, the E.R.R. during crop seasons ranged from 50.52 % to 64.87 % in the treated lots at the different locations while in the control lots, it ranged from 38.62 % to 47.34 % which indicated the efficacy of NaOCl solution in increasing the survivability in muga silkworms. Sodium hypochlorite acts as a leave surface disinfectant. Sodium hypochlorite is effective against bacteria, viruses and fungi and it disinfects the same way as chlorine does. When sodium hypochlorite dissolves in water, two substances viz. hypochlorous (HOCl) and the less active hypochlorite ion (OCI) are formed, which play a role for oxidation and disinfection. The bactericidal and virucidal effect of active chlorine and NaOCl has been reported in the larvae of cabbage looper [14] and Bombyx mori [17]. The application of NaOCl (0.05-0.5 %) was found to effectively reduce the viability and infectivity of Bacillus thuringiensis, Beauvaria bassiana and nuclear polyhedrosis virus (NPV) in cabbage looper [18]. It has been observed that the bactericidal and virucidal action of NaOCl is mainly caused by the release of hypochlorous acid (HOCl). The hypochlorite ion OCI may be a contributing factor since alkaline hypochlorite solutions show some antibacterial potency. The application of NaOCl disrupted the normal protoplasmic balance in the living cell which eventually caused the death of the pathogens and reduced the mortality of the host silkworm. From the present study it is deduced that the application of 0.01 % of Sodium hypochlorite during the rearing of muga silkworm, Antheraea assamensis Helfer can effectively control the mortality due to bacterial and viral diseases thereby will enhance the cocoon production.

Table. I: Effect of different	concentrations of NaOCl	on the ERR % of A.	assamensis.
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Concentration -	Crop season			
Concentration	Jarua (December-January)	Chatua (February-March)	Aherua (June-July)	
0.001%	42.60	47.25	55.65	
0.01%	60.40	64.65	66.55	
0.05%	60.58	62.45	65.45	
0.1%	55.64	58.35	54.70	
Control	38.52	44.50	52.40	

Table. II: Effect of NaOCl Flacheri disease and commercial characters of A. assamensis during 2011-12.

Crop/Season	Location	Treatment with NaOCl	Incidence of disease	ERR %	Cocoon weight	Shell weight
		(0.01 %)	(%)		(g)	(g)
Jarua (Dec-Jan)	GCC Chenijan	Control	46.72	40.22	5.65	0.42
		Treatment	12.50	61.71	6.28	0.45
	Farm No: 03	Control	41.82	42.62	5.53	0.40
	Lahdoigarh	Treatment	10.23	51.75	6.12	0.47
	GCC Chenijan	Control	36.75	47.34	6.22	0.52
Chatua		Treatment	8.54	56.81	6.85	0.58
(Feb-March)	Farm No: 03	Control	37.42	51.87	5.56	0.49
	Lahdoigarh	Treatment	5.62	57.77	6.02	0.52
Aherua (June-July)	GCC Chenijan	Control	24.64	55.52	5.54	0.58
		Treatment	4.82	64.87	6.45	0.62
	Farm No: 03	Control	30.88	54.72	5.8	0.52
	Lahdoigarh	Treatment	4.52	66.82	6.54	0.58
CD at 5%		7.604	7.618	0.362	0.089	
	1%		11.357	11.372	0.541	0.131

 Table. III: Effect of NaOCl Flacheri disease and commercial characters of A. assamensis during 2012-13.

Crop/Season	Location	Treatment with NaOCl	Incidence of disease	ERR %	Cocoon weight	Shell weight
		(0.01 %)	(%)		(g)	(g)
Jarua (Dec-Jan)	GCC Chenijan	Control	36.62	44.62	5.55	0.45
		Treatment	8.55	58.45	6.0	0.42
	Farm No: 03	Control	40.20	42.23	5.06	0.45
	Lahdoigarh	Treatment	10.25	50.52	5.58	0.42
Chatua (Feb-March)	GCC Chenijan	Control	46.52	38.62	5.52	0.49
		Treatment	9.75	52.27	6.02	0.52
	Farm No: 03	Control	23.62	42.33	5.50	0.48
	Lahdoigarh	Treatment	5.55	57.62	5.85	5.0
Aherua (June-July)	GCC Chenijan	Control	30.22	40.56	5.98	0.48
		Treatment	12.44	54.56	6.0	0.45
	Farm No: 03	Control	34.82	45.55	5.58	0.48
	Lahdoigarh	Treatment	7.83	57.23	6.02	0.52
CD at 5%			7.432	3.678	0.305	0.086
	1%		11.093	5.491	0.456	0.128

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