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Biocidal Efficiency of Green Inhibitor in Neutral Medium

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Abstract : The inhibiting action of *Citrus medica* [CM] and biocides towards corrosion of carbon steel in open recirculating cooling systems has been studied in presence of Mn^{2+} by weight loss method. Open recirculating cooling water systems are commonly used for industrial cooling purposes to dissipate heat. The main problems associated with this system are scaling, corrosion, fouling and microbiological growth which if left untreated can lead to various problems. Microbiologically influenced corrosion is emerging as a serious problem in cooling systems. Once a biofilm forms, the local environment at the metal /biofilm interface undergoes drastic changes in terms of pH, dissolved oxygen content and concentration of the ionic species. To eliminate the threat of such potential problems, a suitable biocide must be added. A biocide must successfully control a broad spectrum of microbial contamination, provide cost-effective performance and prove compatible with other system components, while at the same time meeting stringent environmental, health and safety standards.This paper is concerned with the study of biocidal efficiencies of *Ocimum tenuiflorum*, *Solanum trilobatum* and *Azadirachta indica* in the presence and absence of the inhibitor *Citrus medica* in carbon steel.

Keywords: Biocide, Carbon steel, *Ocimum tenuiflorum* [OT], *Solanum trilobatum* [ST] and *Azadirachta indica* [AI].

1. Introduction

In open recirculating cooling water system (ORCS) of a petroleum refinery, carbon steel corrosion is a major problem. The makeup water used in (ORCS) are either from river, sea or underground sources. The presence of large amount of total dissolved solids, total suspended solids, total hardness, microorganism and dissolved gases, such as O2 and CO2 in the makeup water are responsible for scaling, fouling, under deposit and microbiological corrosion in the various equipments of cooling water system. Due to such corrosion the tubes of heat exchanger, an important part of ORCS get damaged easily, which affect the productivity and profitability. To minimize the adverse effect of corrosion, various productive methods have been adopted, one of the frequency used measure is the use of organic compounds containing nitrogen and sulphur atoms [1-3]. These compounds either can form strong co-ordination bond with metal atom or form passive film on the surface [4]. There is still a continuous search for better inhibitors or blend of inhibitors to meet the demand of the industry. The selection criteria for various inhibitors include low concentration, stability in recirculation, cost effectiveness and low operational hazard. Many studies have also been done for preparation of corrosion inhibitors having biocide properties. Microbiologically induced corrosion (MIC) has been identified as one of the major causes of corrosion of cooling water systems [5]. Technically, biocide is any substance that is poisonous to organisms and can inhibit their metabolism or annihilate them. The objective of this work seeks to investigate the efficacy of biocide in the treatment of internal corrosion of mild steel in cooling water pipelines [6].

2. Experimental

2.1. Preparation of the specimen

The carbon steel specimens were chosen from the same sheet of the following composition: 0.0267% S, 0.06% P, 0.4% Mn, 0.1% C and the rest iron. Carbon steel specimens of the dimensions 1.0 x 4.0 x 0.2 cm were polished to mirror finish, degreased with trichloroethylene and used for weight-loss and surface examination studies.

2.2. Weight Loss Method

Three carbon steel specimens were immersed in 100ml of the solutions containing DD water and various concentrations of the inhibitor in the absence and presence of Mn^{2+} for one day. The corrosion product is cleaned with Clark's solution [7]. The weights of the specimens before and after immersion were determined using a Shimadzu balance AY62. Inhibition efficiency (IE) was calculated from the relationship:

$IE = (1-W_2/W_1) \times 100\ \%$
where
W_1 = corrosion rate in the absence of inhibitor and
W_2 = corrosion rate in the presence of inhibitor.

2.3. Plant materials

The plant leaves used in this study were collected in and around Trichy District. The disease free plant part (leaf) was spread out and dried in the laboratory at room temperature for 5-8 days until they were easily broken by hand. Once completely dried, leaves of the plant were grounded to a fine powder using an electronic blender. Plants were stored in a closed container at room temperature until required.

2.4. Preparation of Solvent Extracts

Fifty grams of the dried and powdered plant material (leaves) were soaked separately with 500 ml of the solvent ethanol, in a soxhlet apparatus for 48 hrs at 31°C until complete extraction of the materials. At the end of 48 hrs, each extract was filtered through Whatman No.1 filter paper and filtrates were concentrated at room temperature in order to reduce the volume. The sample was concentrated using rotary evaporator and freeze dried to powdered form. The paste like extract was stored in pre-weighed screw scrapped bottle and the yield of extracts was weighed. These screw scrapped bottle was kept in refrigerator at 4°C. The extract was reconstituted using minimal amounts of the extracting solvent prior to use.

2.5. Zobell medium

Zobell medium was prepared by dissolving 5g of peptone, 1g of yeast extract, 0.1g of potassium dihydrogen phosphate and 15g of agar-agar in 1litre of double distilled water. The medium was sterilized by applying 15 pounds per square inch for 15 minutes in an autoclave.

2.6. Influence of *Ocimum tenuiflorum*

The corrosion rates of carbon steel in ground water containing Mn^{2+}+ CM inhibitor formulation for various concentrations of *Ocimum tenuiflorum* [OT] are tabulated in Table 1 and the CR as a function of the concentrations of *Ocimum tenuiflorum* is given in Figure 1.

It is seen from the Table 1 that the addition of *Ocimum tenuiflorum* to the inhibitor formulation drastically reduces the IE from 81.5% to 47%. This is due to the precipitation of CM by *Ocimum tenuiflorum*. However, further addition of *Ocimum tenuiflorum* increases the IE and reaches 61.6% at 200 ppm, above which the IE decreases. The gradual increase in IE above 50 ppm is due to the corrosion inhibition property of *Ocimum tenuiflorum*. However, after micelle formation due to the aggregation of monomers to form micelles, the IE decrease beyond 200 ppm.

Table 1. Influence of Ocimum tenuiflorum on the IE of CM + Mn^{2+} system

Corrosion rates of carbon steel in ground water in the presence and absence of the inhibitor system and the inhibition efficiencies obtained by the mass-loss method.

Inhibitor system: CM + Mn^{2+}+ *Ocimum tenuiflorum*

S.No	CM (ppm)	Mn^{2+} (ppm)	*Ocimum tenuiflorum* (ppm)	Corrosion Rate (mdd)	Inhibition Efficiency (%)
1	0	0	0	38	-
2	300	20	0	7.0	81.5
3	300	20	50	20.12	47
4	300	20	100	17.65	53.5
5	300	20	150	16.69	56
6	300	20	200	14.58	61.6
7	300	20	250	15.42	59.4
8	300	20	300	15.23	59.9

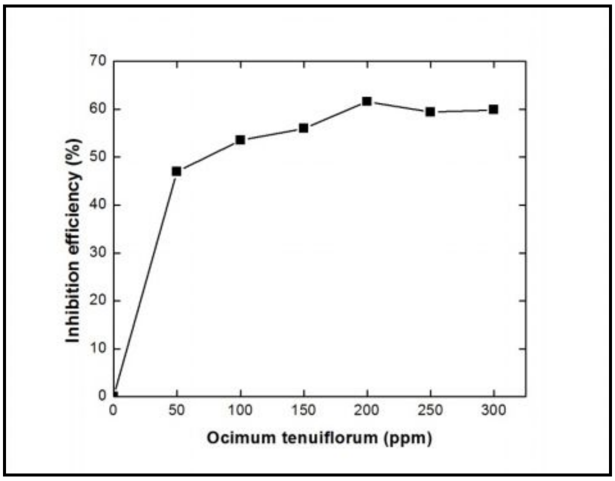

Figure 1. Influence of OT on the CR of carbon steel in the presence of CM+Mn^{2+} system

2.7. Influence of *Solanum trilobatum*

The corrosion rates of carbon steel in ground water containing Mn^{2+}+ CM inhibitor formulation for various concentrations of *Solanum trilobatum* [ST] are tabulated in Table 2. The CR as a function of the concentrations of *Solanum trilobatum* is given in Figure 2.

A steep fall in IE of the formulation is observed on introducing 50 ppm of *Solanum trilobatum* to the inhibitor system. This is due to the precipitation of CM by *Solanum trilobatum*. Further addition of *Solanum trilobatum* does not alter the IE much.

Table 2. Influence of Solanum trilobatum on the IE of CM + Mn^{2+} system

Corrosion rates of carbon steel in ground water in the presence and absence of the inhibitor system and the inhibition efficiencies obtained by the mass-loss method.

Inhibitor system: CA + Mn^{2+}+ *Solanum trilobatum*

S.No	CM (ppm)	Mn^{2+} (ppm)	*Solanum trilobatum* (ppm)	Corrosion Rate (mdd)	Inhibition Efficiency (%)
1	0	0	0	38	-
2	300	20	0	7.0	81.5
3	300	20	50	13.98	63.21
4	300	20	100	13.58	64.26
5	300	20	150	13.12	65.47
6	300	20	200	12.89	66
7	300	20	250	12.52	67
8	300	20	300	11.59	69.5

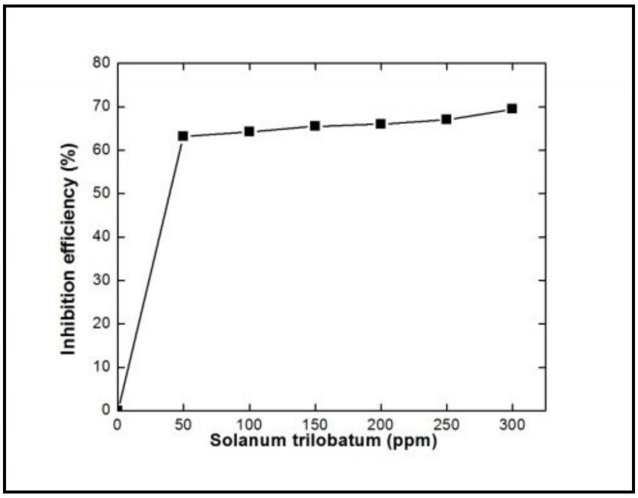

Figure 2. Influence of ST on CR of carbon steel in the presence of CM+Mn^{2+} system

2.8. Influence of *Azadirachta indica*

The corrosion rates of carbon steel in ground water containing Mn^{2+}+ CM inhibitor formulation for various concentrations of *Azadirachta indica* [AI] is tabulated in Table 3.

The CR as a function of the concentrations of *Azadirachta indica* is given in Figure 3. Addition of 50 ppm of *Azadirachta indica* to 300 ppm of CM and 20 ppm of Mn^{2+} reduces the IE to a small extent, (from 81.5% to 77.89%). However, the IE does not alter much by further addition of *Azadirachta indica*. Hence, it is concluded that *Azadirachta indica* does not alter the corrosion behaviour of steel in the presence of inhibitor system.

Table 3. Influence of *Azadirachta indica* (AI) on the IE of CM + Mn^{2+}system

Corrosion rates of carbon steel in ground water in the presence and absence of the inhibitor system and the inhibition efficiencies obtained by the mass-loss method.

Inhibitor system: CM + Mn^{2+}+ *Azadirachta indica*

S.No	CM (ppm)	Zn^{2+} (ppm)	*Azadirachta indica (*ppm)	Corrosion Rate (mdd)	Inhibition Efficiency (%)
1	0	0	0	38	-
2	300	20	0	7.0	81.5
3	300	20	50	8.40	77.89
4	300	20	100	8.31	78.13
5	300	20	150	8.12	78.63
6	300	20	200	8.10	78.68
7	300	20	250	7.92	79.15
8	300	20	300	7.90	79.21

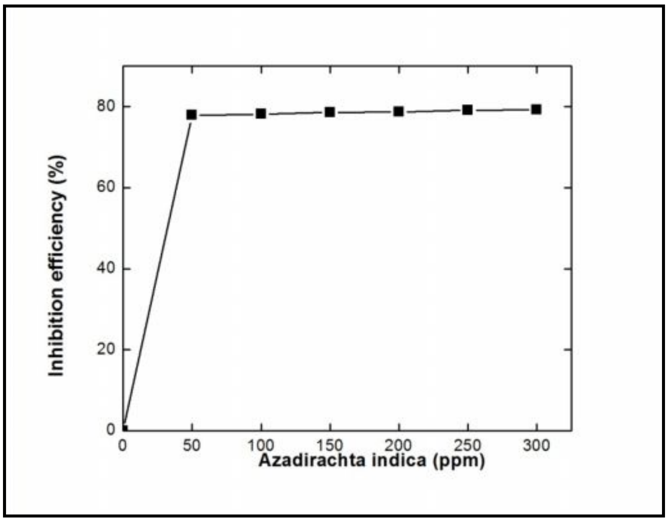

Figure 3. Influence of ST on CR of carbon steel in the presence of CM+Mn^{2+} system

2.9. Determination of biocidal efficiency of the system

Inhibitor-Zn2+ formulation that offered the best corrosion inhibition efficiency was selected. The biocidal efficiency of biocides such as *Ocimum tenuiflorum* [OT], *Solanum trilobatum* [ST] and *Azadirachta indica* [AI] was determined. Various concentrations of OT, ST and AI namely 50 ppm, 100 ppm, 150 ppm, 200 ppm, 250 ppm were added to the formulation consisting of the inhibitor system. Polished and degreased carbon steel specimens in duplicate were immersed in these environments for one day. After one day one ml each of test solutions from the environments was pipetted out into sterile petri dishes containing about 20 ml of the sterilized Zobell medium kept in a sterilized environment inside the Laminar flow system. Then the Petri dishes were kept in an incubator at 30ºC in an inverted position for 2 days. Distinct colonies of bacteria were developed. Such colonies may be easily isolated. The number of colony forming units (CFU/mL) present in the above formulations were determined by Step dilution technique [8]. Each organism grows and reproduces itself. To determine the number of colonies, plate count technique [9] was applied.

2.8. Biocidal efficiencies of biocides in DD **water**

Biocidal efficiencies of various concentrations of *Ocimum tenuiflorum* with and without the presence of inhibitor formulations are tabulated in the Table 4. The visuals of the bacterial colonies formed in DD water in the presence and absence of the inhibitor system and the biocide after immersing the specimens for three days are shown in Figures 4 and 5

Table 4. Biocidal efficiencies of *Ocimum tenuiflorum* in the presence and absence of CM + Mn^{2+} system in DD water

S.No	*Citrus medica* (ppm)	Mn^{2+} (ppm)	*Ocimum tenuiflorum* (ppm)	Colony Forming (units/ml)	Biocidal Efficiency (%)
1	0	0	0	1374	-
2	0	0	10	6	99.56
3	0	0	20	0	100
4	0	0	50	0	100
5	300	20	0	753	45.19
6	300	20	10	680	50.5
7	300	20	50	598	56.48
8	300	20	100	0	100

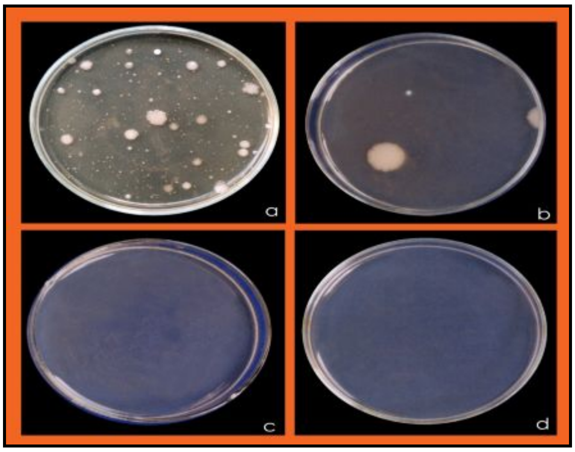

Figure 4. Bacterial colonies formed in ground water in the presence of various concentration of OT (a) 0 ppm, (b) 10 ppm, (c) 20 ppm, (d) 50 ppm

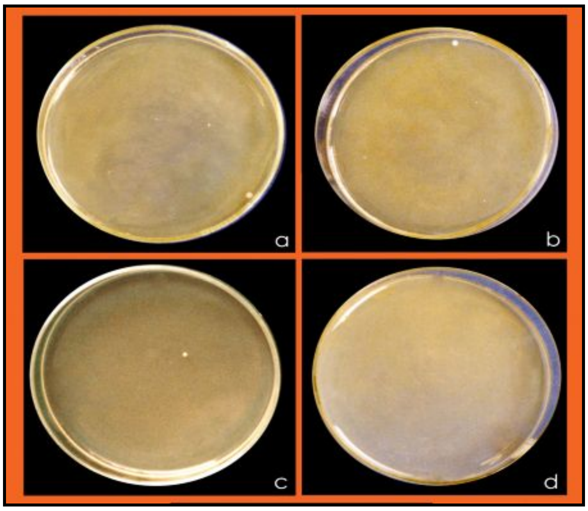

Figure 5. Bacterial colonies formed in DD water containing 20 ppm Mn^{2+} and 300 ppm of CM in the presence of various concentration of OT
(a) 0 ppm, (b) 10 ppm, (c) 50 ppm, (d) 100 ppm

The analysis of the Table 4 shows that in the absence of inhibitor formulation, 20 ppm of *Ocimum tenuiflorum* is sufficient to achieve 100 % BE and in the absence of *Ocimum tenuiflorum*, the Mn^{2+}/ CM formulation offers 45.19% BE. Though, the addition of 10 ppm of *Ocimum tenuiflorum* to the DD water in the presence of inhibitor formulation increases the BE, 100 ppm of *Ocimum tenuiflorum* gives 100 % BE. However addition of *Ocimum tenuiflorum*, reduces the IE drastically and hence it is inferred that *Ocimum tenuiflorum* is not a good additive as biocide for the Mn^{2+}/ CM formulation.

Table 5. Biocidal efficiencies of *Solanum trilobatum* in the presence and absence of CM + Mn^{2+} system in DD water

S.No	*Citrus medica* (ppm)	Mn^{2+} (ppm)	*Solanum trilobatum* (ppm)	Colony Forming (units/ml)	Biocidal Efficiency (%)
1	0	0	0	1374	-
2	0	0	10	3	99.78
3	0	0	20	0	100
4	0	0	50	0	100
5	300	20	0	753	18.92
6	300	20	10	1114	99.48
7	300	20	50	0	100
8	300	20	100	0	100

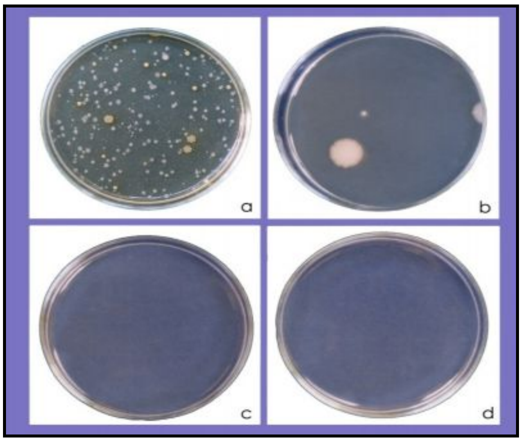

Figure 6. Bacterial colonies formed in ground water in the presence of various concentration of ST
(a) 0 ppm, (b) 10 ppm, (c) 20 ppm, (d) 50 ppm
(b)

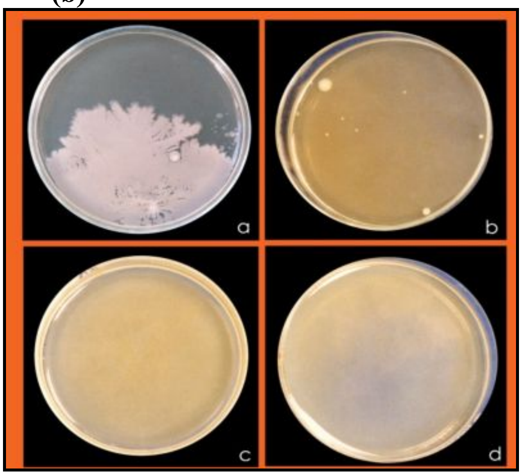

Figure 7. Bacterial colonies formed in DD water containing 20 ppm Mn^{2+} and 300 ppm of CM in the presence of various concentration of ST
(a) 0 ppm, (b) 10 ppm, (c) 50 ppm, (d) 100 ppm

The biocidal efficiencies of various concentrations of *Solanum trilobatum* with and without the presence of inhibitor formulations are tabulated in the Table 5. The visuals of the bacterial colonies formed in DD water in the presence and absence of the inhibitor system and the biocide after immersing the specimens for one day are shown in Figures. 6 and 7

The analysis of the Table 5 shows that in the absence of inhibitor formulation, 20 ppm of *Solanum trilobatum* is sufficient to achieve 100 % BE and in the absence of *Solanum trilobatum*, the Mn^{2+}/ CM formulation offers 43.19% BE. However, the addition of 20 ppm of *Solanum trilobatum* to the DD water in the presence of inhibitor formulation does not give 100 % BE. This is due to the precipitation of *Solanum trilobatum* by *Citrus medica*. Hence a higher concentration of *Solanum trilobatum* is required to achieve 100 % BE. As a steep fall in the IE is observed on the addition of *Solanum trilobatum*, it is concluded that *Solanum trilobatum* is not compatible with the inhibitor Mn^{2+}/ CM formulation.

The biocidal efficiencies of various concentrations of *Azadirachta indica* with and without the presence of inhibitor formulations are tabulated in the Table 6. The visuals of the bacterial colonies formed in DD water in the presence and absence of the inhibitor system and the biocide after immersing the specimens for three days are shown in Figures 8 and 9. 100 ppm of *Azadirachta indica* in the presence and absence of the inhibitor system controls the microbial activity in DD water. As addition of *Azadirachta indica* is not affecting the corrosion inhibition efficiency much, *Azadirachta indica* is a suitable biocide for the corrosion and microbial growth control in DD water.

Table 6. Biocidal efficiencies of *Azadirachta indica* in the presence and absence of CM + Mn^{2+} system in DD water

S.No	*Citrus medica* (ppm)	Mn^{2+} (ppm)	*Azadirachta indica* (ppm)	Colony Forming (units/ml)	Biocidal Efficiency (%)
1	0	0	0	1374	-
2	0	0	10	1100	19.94
3	0	0	50	2	99.85
4	0	0	100	0	100
5	0	0	150	0	100
6	300	20	0	753	45.19
7	300	20	10	750	45.4
8	300	20	50	1	99.9
9	300	20	100	0	100
10	300	20	150	0	100

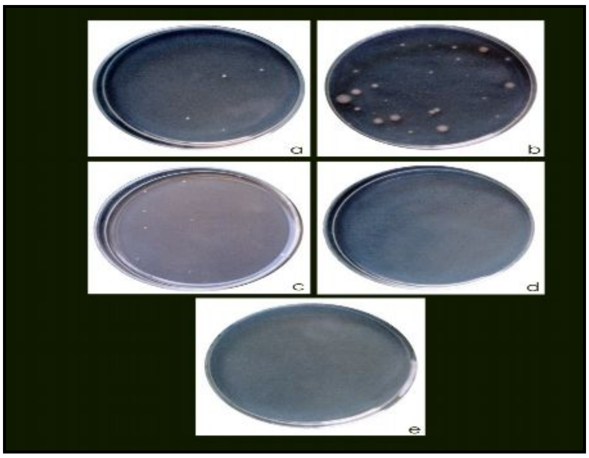

Figure 8. Bacterial colonies formed in ground water in the presence of various concentration of AI

(a) 0 ppm, (b) 10 ppm, (c) 50 ppm, (d) 100 ppm (e) 150 ppm

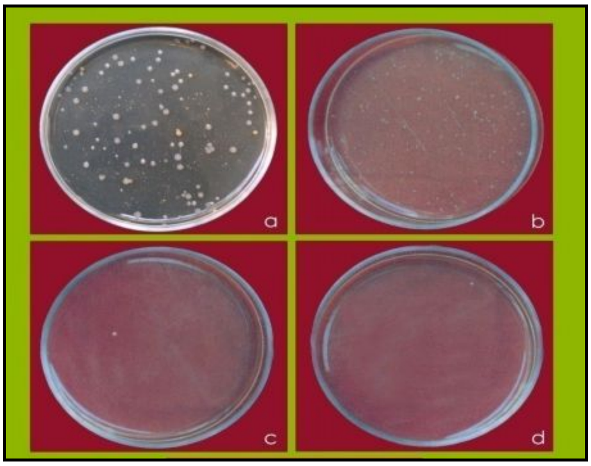

Figure 9. Bacterial colonies formed in DD water containing 20 ppm Mn^{2+} and 300 ppm of CM in the presence of various concentration of AI
(a) 0 ppm, (b) 10 ppm, (c) 50 ppm, (d) 100 ppm

3. Conclusion

Among the three biocides, ST offers 99.78% BE when 10 ppm of ST was added to DD water in which the test specimens were immersed for one day. The inhibitor formulation gives 18.92% BE in the absence of ST. The inhibitor formulation consisting of 300 ppm of CM, 20 ppm of Mn^{2+} ions and 100 ppm of ST gives 100 % BE. Therefore, a minimum of 100 ppm of SDS is required for the complete eradication of microbes. As the combination of 300 ppm of CM, 20 ppm of Mn^{2+} ions and 100 ppm of SDS offer 100 % BE and 64.26% IE, it is ideal to use this combination for cooling water system to control corrosion as well as microbial growth.

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