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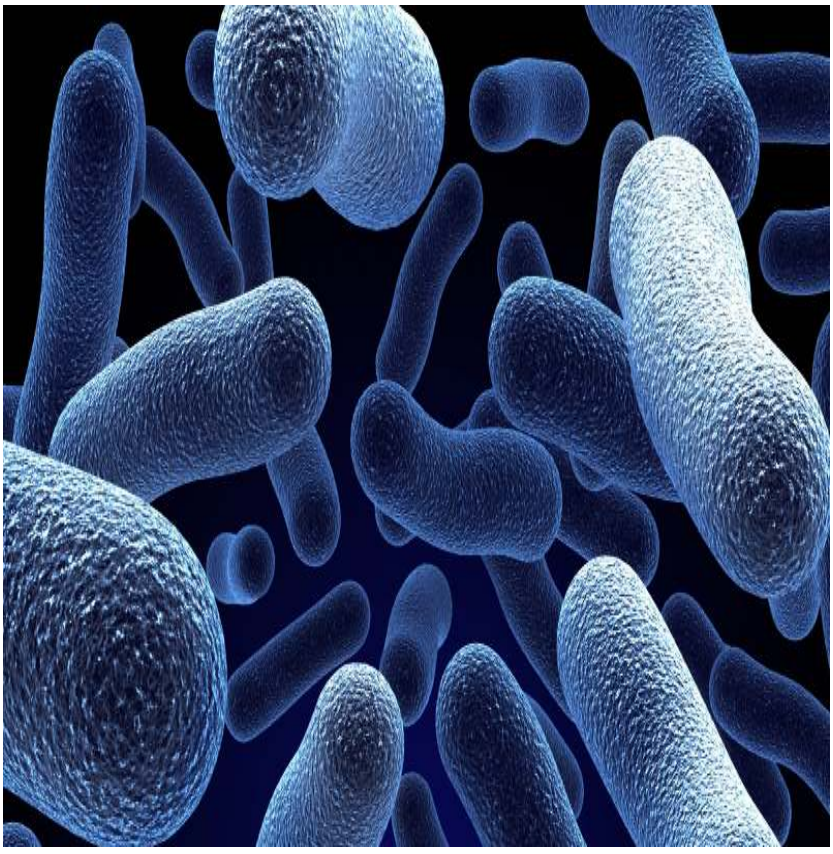
ANTIMICROBIAL ACTIVITY OF Cr-DEPOSITED LASER IRRADIATED MARAGING STEEL AGAINST WATER BORNE BACTERIA

Sambhaji Dagadu Shinde
Department of Physics and Microbiology Research Center,
Mahatma Phule Mahavidyalaya, Pimpri Pune. India.

Short Profile

Sambhaji Dagadu Shinde is a H.O.D. at Department of Physics and Microbiology Research Center in Mahatma Phule Mahavidyalaya, Pimpri Pune. India. He has completed M.Sc., M.Phil., Ph.D. He has published 18 research papers in various journals.



ABSTRACT:
As received and Cr-deposited laser irradiated samples of maraging steel were evaluated for their antimicrobial activity. The Kirby and Bauer method was used with modifications. In order to evaluate antibacterial property, the samples were placed in the microbial lawns and the diameter of inhibition zone was measured. The surface morphology of the samples was studied using Scanning Electron Microscopy technique. It was observed that both maraging steel samples show antibacterial activities against water borne bacteria viz. Legionella pneumophila, Shigella sonnei, Campylobacter jejuni and Aeromonas hydrophila. The antibacterial activity of Cr-deposited laser irradiated samples was found to be greater than that of as received samples for L. pneumophila and A. hydrophila but smaller for S. sonnei and C. jejuni. This was due to the changes in surface morphology and rearrangement of the constituents at the surface. The chromium ions at the surface of the samples are responsible for increase in the antimicrobial activity.

KEYWORDS
Cr-deposition, Laser irradiation, water borne bacteria, antimicrobial activity .

INTRODUCTION:

The effects of laser irradiation on the surface of metals and alloys have been studied by many researchers. If the high power laser is incident on the surface of the metal then characteristic properties of the base material are changed. The melting of the surface occurs in less than a second but base metal remains cool and acts as heat sink. At the boundary of the melted region and base metal, a very high temperature gradient was observed. This caused rapid quenching and resolidification [Draper et al., 1984]. If the chromium is deposited on the surface of maraging steel and lasers are incidents on it, then the new alloy formation results. The properties of base material for application are changed. The alloy formed was observed to be corrosion resistant [Shinde (1) et al., 2013] [Shinde (2) et al., 2013]. The antimicrobial activity of metals, alloys and different chemicals was studied by many researchers. The antimicrobial activity study and corrosion inhibition of 18% Ni 350 grade maraging steel have been investigated [Henrich,2011]. The catalytic antimicrobial coatings which may capture and kill pathogens have been studied [David et al., 2009]. The nano-crystallized 18% Ni maraging steel related with antibacterial activity was analyzed using X-ray diffractometer. The investigation of Bauer et al. about the antibiotic susceptibility testing is very important and can be studied by modified standard single disc method [Bauer et al., 1966] [Jiang et al., 2007] [Sheretz et al., 1989].

It was observed that there is no any literature available related with antibacterial activity of Cr-deposited laser irradiated maraging steel. Especially, the study of antibacterial activity of selected sample in the drinking water supply was found to be not available. Hence, I selected this problem.

EXPERIMENTAL: MATERIALS AND METHODS

Preparation of samples:

The samples having thickness 1 mm were cut in the form of circular discs with 8 mm diameter. The due care was taken to avoid scratches and other defects. Samples were polished on both sides by using the various grade emery papers. They were cleaned and washed using distilled water and dried. They were cleaned and washed with the double distilled water and were dried.

The Cr-deposited samples were prepared by depositing chromium over layer with thickness 25 nano-meters on to the as-received maraging steel specimens. The care was taken to ensure clean evaporation environment and at background pressure of 10-6 torr, the evaporation was carried out. The evaporation unit was equipped with fore line traps (molecular sleeves) and liquid nitrogen traps to avoid contamination due to oil mist. The coatings were produced without generation of hydrogen, which can embrittle the substrate material. The care was taken to remain chromium depositions free from pores and inclusions [Shinde (1) et al., 2013].

In the intervals between polishing and surface modification and between surface modification and subsequent use, the specimens were stored in desiccators.

Cr-deposited laser irradiated samples

Laser irradiation was carried out using JK laser Model Sr. No. 5696 (Q-switched ruby laser, wave length 694 nm, pulse duration of 30 ns). Both as-received and Cr-deposited specimens were laser irradiated by varying laser energy from 0.6 J to 1.8 J. The samples were called as Cr-deposited laser irradiated samples. The laser irradiation method used was very advantageous [Shinde (2) et al, 2013]

[Cottrell et al., 1998] [McCafferty et al., 1982].

ANTIMICROBIAL ACTIVITY:

The antimicrobial activity was evaluated using the modified susceptibility disc diffusion technique [Baueret al., 1966] [Holt et al., 1994] [Shinde (1), 2013] [Shinde (3), 2014].

PROCEDURE:

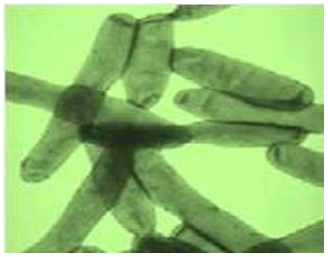
1.The sterile swab was dipped in to the inoculums and sterilization of the agar plates was accomplished. The required inoculums selection was done by pressing and rotating the swab above the level of liquid [Jogensen et al., 2007].
2.The streaking of swab was done by rotating the plates through 60 0 angle each time. At room temperature, the inoculation was dried by closing the lid of plates. The swab was allowed to pass round the agar surface edge.
3.The inoculation was dried for a few minutes, at room temperature, with the lid closed.
4.The standard antibiotic discs (Ampicilin and Tetracycline) and Maraging steel discs (control, Cr deposited laser irradiated) were placed in inoculated plates.
5.These plates were placed in refrigerator by maintaining the temperature of 4 0C and time interval of 15 minutes. Then the ready plates were kept in the incubator.
6.The temperature of 37 0C was maintained constant for the 48 hours' time [Shinde (3), 2014].
7.After this 48 hours' time span, the inhibition zone diameter was measured and recorded in mm. The measurement was accomplished with a ruler from the bottom side of plate. During measurement, the lid of the plate was not opened.
8.The measurement and experimental procedure was repeated three times for each specimen.
The results were tabulated in a systematic form.

RESULTS AND DISCUSSION:

Results of the microbial analysis of water samples are presented in TABLE.1, TABLE.2 and FIG.1. A water sample (Manchar village drinking water supply) examined in this study showed pH 6.7. It was observed that water sample contained four different bacterial isolates viz. Legionella pneumophila, Shigella sonnei, Campylobacter jejuni and Aeromonas hydrophila. These isolates were identified and confirmed, based on the selective, differential media and biochemical tests [Shind e (3), 2013] [Shinde (4), 2014].

TABLE.1: BACTERIA IDENTIFIED IN DRINKING WATER SUPPLY

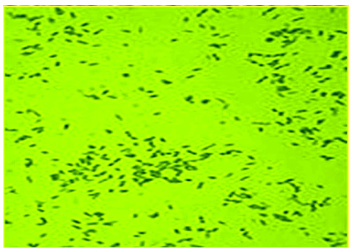
Sr. No.	Isolate No.	Bacteria – Species Name
1	Isolate 1	*Legionella pneumophila*
2	Isolate 2	*Shigella sonnei*
3	Isolate 3	*Campylobacter jejuni*
4	Isolate 4	*Aeromonas hydrophila*



a. ***L. pneumophila***

b. ***S. sonnai***

c. ***Campilobacter jejuni***



d. ***A. hydrophila***

FIG.1: IDENTIFICATION OF BACTERIA PHOTOGRAPHS AFTER MAGNIFICATION : a. L. pneumophila b. S. sonnai c. Campilobacter jejuni d. A. hydrophila

ANTIBACTERIAL ACTIVITY FOR DIFFERENT BACTERIAL ISOLATES (Cr-DEPOSITED LASER IRRADIATED SAMPLES):

Antibacterial activity of as received laser irradiated samples for different bacterial isolates has been given in TABLE.2 and shown in FIG.3.

It was observed that the diameter of inhibitory zone diameters for Cr-deposited laser irradiated samples for different bacterial isolates were higher (10, 6, 4, 5 mm) than that for as received (8, 8, 6 and 4 mm) and Cr-deposited samples for all four isolates (2, 6, 4 and 2 mm). Particularly for energy density 1.8 J/cm2, diameter of zones was found to be highest (10mm) for Legionella pneumophila isolate. As like as received laser irradiated samples, these samples also showed the increase in diameter of inhibitory zones with increase in corrosion resistance. Thus from both the observations it is clear that the antibacterial activity depends on the corrosion resistance of the maraging steel samples for laser treatment.

TABLE.2: Antibacterial activity of Cr-deposited laser irradiated samples (inhibitory zone in mm diameter by subtracting disc diameters)

Sr. No.	Name of Organism/ Isolate No.	Antibiotics		Control sample (as received)	Cr-deposited sample	Cr-deposited laser irradiated samples (energy density in J/cm^2)			
		Ampicillin	Tetracycline			0.6	1.0	1.3	**1.8**
		Diameter in mm							
1	Isolate 1	0	12	8	2	3	6	7	**10**
2	Isolate 2	12	2	8	6	2	5	6	**6**
3	Isolate 3	11	3	6	4	2	2	5	**4**
4	Isolate 4	9	8	4	2	5	4	3	**5**

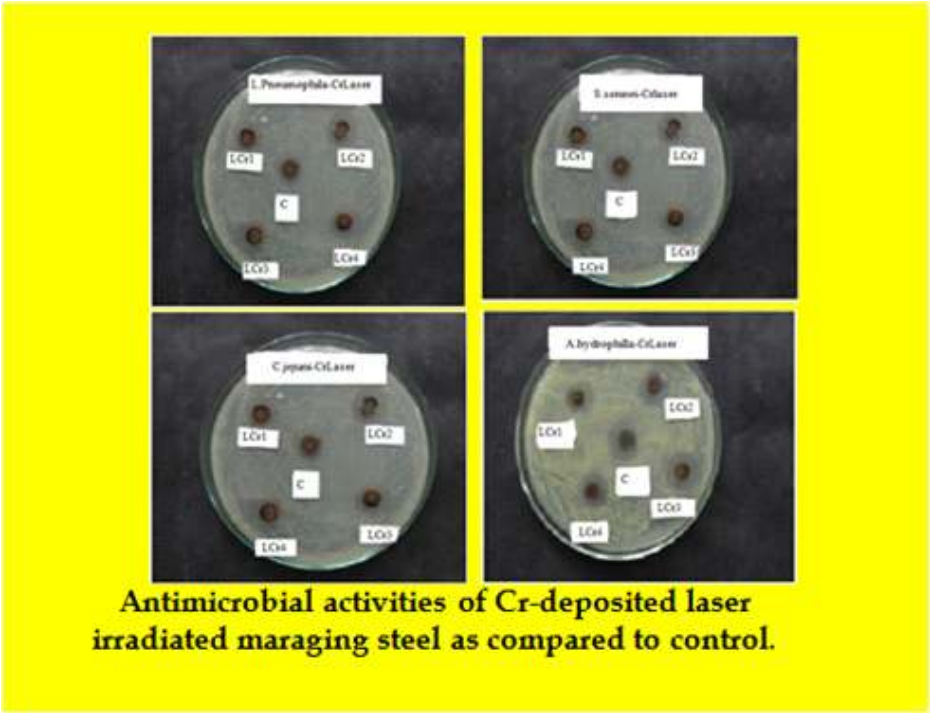
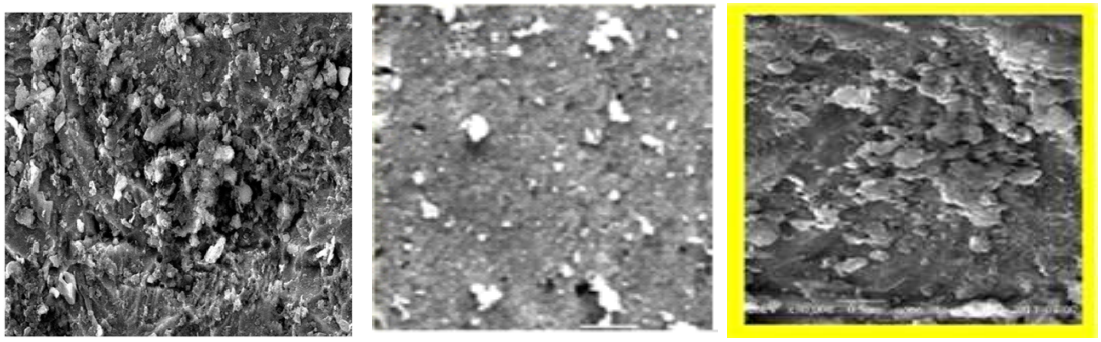


FIG.2: ANTIMICROBIAL ACTIVITIES OF Cr-DEPOSITED LASER IRRADIATED MARAGING STEEL: For different isolates

a. L. pneumophila b. S. sonnai

c. Campilobacter jejuni d. A. hydrophila



(a) (b) (c)

FIG.3: SEM OF (a) AS RECEIVED (b) Cr-DEPOSITED (c) Cr-DEPOSITED LASER TREATED (1.8 J/ cm^2) SPECIMEN

Thus from both the observations it is clear that the antimicrobial activity is dependent on the corrosion resistance of the maraging steel samples for laser treatment [Shinde (1), 2013] [Shinde (4), 2014]. The increased activity may be related to the Chromium amount present at the surface of the sample. The activity of both samples was less compared with antibiotics samples viz. Ampicillin and Tetracycline.

The susceptible nature of bacteria to these samples may be dependent on the corrosion resistance and due to the redistribution of the elements at the surface.

CONCLUSIONS:

From all the above results, it was concluded that

1.Maraging steel basically consists of nickel (18%). It was observed for as received maraging steel samples, chromium deposited samples, and Cr deposited laser irradiated samples are found to possess antibacterial activity. The degree of measure will be different at different places and timings or seasons. The environment conditions also affect the activity changes. If there is change in temperature, then also activity will change.

Thus it can be concluded that as-received and or chromium deposited, irradiated 18% Ni Maraging steel can kill/destroy microorganism viz. Legionella pneumophila, Shigella sonnei, Campylobacter jejuni and Aeromonas hydrophila. Conversely, it can be concluded that these bacteria can eliminate the toxic effects of nickel and chromium elements. We can conclude such effect to be selfi in the both metals.

2.As received laser irradiated samples at dose rate of 1.8 J/cm2 showed significant antibacterial activity. The effect of increased antibacterial activity may be associated with Cr ions clinged at the surface of sample.

3.Antimicrobial activity of as received as well as irradiated samples exhibited significant results. The findings showed that as-received maraging steel as well as surface modified can be utilized for water purification systems. The chromium (Cr) deposited (thickness 25 nm) and as- received samples of maraging steel 250 grade were evaluated for their antimicrobial activity. Diameters of inhibition zones for all isolates of as-received maraging steel are found to be larger than that of Cr-deposited samples. It was concluded that this effect may be due to the morphological changes at the surface of the sample of

maraging steel.

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