



# Study on the microbial corrosion of Carbon Steel and its inhibition with some natural extracts

A Thesis
Submitted for partial fulfillment of Master degree in
Microbiology

## By

## Basma Ahmed Ali Omran

B. Sc. Microbiology – Biochemistry (2007)

## **Supervisors**

#### Dr. Mohamed Abd-Elmontaser Abouzeid

Dr. Nour Shafik El-Gendy

Professor of Microbiology Microbiology Department, Faculty of Science, Ain Shams University

Assistant Professor of Environmental
Biotechnology
Head of Petroleum Biotechnology Lab,
Egyptian Petroleum Research Institute
(EPRI)

### **Dr. Einas Hamed El-Shatoury**

Lecturer of Microbiology Microbiology Department, Faculty of Science, Ain Shams University

Microbiology Department Faculty of Science Ain Shams University 2013





Student name: Basma Ahmed Ali Omran

**Thesis Title:** 

Study on the microbial corrosion of carbon steel and its inhibition with some natural extracts

**Degree:** Master Degree of Science (Microbiology)

**Supervised by:** 

Dr. Mohamed Abd-Elmontaser Abouzeid

Professor of Microbiology Microbiology Department, Faculty of Science, Ain Shams University

### Dr. Nour Shafik El-Gendy

Assistant Professor of Environmental
Biotechnology
Head of Petroleum Biotechnology Lab,
Egyptian Petroleum Research Institute
(EPRI)

### **Dr. Einas Hamed El-Shatoury**

Lecturer of Microbiology Microbiology Department, Faculty of Science, Ain Shams University

Microbiology Department Faculty of Science Ain-Shams University 2013





# Approval sheet Study on the microbial corrosion of carbon steel and its inhibition with some natural extracts

## By Basma Ahmed Ali Omran

B. Sc. Microbiology – Biochemistry

Ain Shams University

(2007)

## This thesis for M.Sc degree has been approved by:

Prof.	Dr.	Ahn	ned Ibrahir	n EL-Diwany				
Profes	sor	of	Microbiolo	gy, Microbia	and and	Natura	ıl produ	cts
Depar	tme	nt, N	ational Res	earch Centre.				
		,						
•		,						
Prof.	Dr.	ŕ	na El-Zhar	aa Hanafy As	shour			
		Fatr		raa Hanafy As Engineering				

**Date of Examination:** 7/10/2013

# Acknowledgment

First, thanks to Allah, the most merciful and the most graceful.

This thesis would not have been possible without the guidance and the help of several individuals who in one way or another contributed and extended their valuable assistance in the preparation and completion of this study.

I am truly indebted and grateful to **Dr. Mohamed Abd-Elmontaser Abouzeid**, Professor of Microbiology, Microbiology Department, Faculty of

Science, Ain Shams University for his valuable assistance and supervision.

I offer my sincerest gratitude to my supervisor **Dr. Einas Hamed El-Shatoury**, lecturer of Microbiology, Faculty of Science, Ain Shams University for her valuable guidance.

I would like to express my deepest appreciation to **Dr. Nour Shafik El-Gendy**, Assistant Professor of Environmental Biotechnology, Head of
Petroleum Biotechnology Lab, Egyptian Petroleum Research Institute for
her supervision and helping in completing this work.

I would also like to acknowledge with much appreciation the role of **Dr. Nesreen Abd-Elhamid**, Researcher in Marine Biotechnology, Petroleum Research Institute for her continuous support during the work.

I would like to thank and express my gratitude to **Dr. Amal Hamdy**, Assistant Professor in Egyptian Petroleum Research Institute, for her valuable efforts in guiding me throughout the work. One simply could not wish for better or friendlier supervisors.

Special thanks go to the staff and members of Petroleum Biotechnology lab for their encouragement and continuous help.

My gratitude is extended to Head of Microbiology Department, my colleagues in Microbiology Department, Faculty of Science, Ain Shams University.

Special thanks go to Egyptian Petroleum Research Institute (EPRI) which provided me with the support and equipment I have needed to produce and complete this thesis.

Basma Ahmed Ali Omran

# **DEDICATION**

I Dedicate This Work to: My Mother. This is for you, Mom.

Thanks for always being there for me in each step of the way. I lovingly dedicate this thesis to my husband, Mohamed Omar.

Thanks for your love, patience and endless support when I needed you. Finally, to my sister, brother and my new coming baby.

# **Declaration**

This thesis has not been previously submitted for this or other degree in this or any other university.

Basma Ahmed Ali Omran

# **CONTENTS**

Title	Page
List of Tables	
List of Figures	
List of Abbreviations	
Abstract	1
Introduction	3
Aim of the Work	7
Review of Literature	
1.1 Microbially Influenced Corrosion	8
1.1.1 Economic impact of MIC	11
1.1.2Microorganisms and MIC	12
1.1.2.1 Aerobic microbial corrosion	13
a. Sulfur oxidizing bacteria	13
b. Metal oxidizing bacteria	14
c. Slime producing bacteria	15
d. Fungi	16
e. Microalgae	16
1.1.2.2 Anaerobic microbial corrosion	17
a. Anaerobic corrosion by sulfate-reducing bacteria (SRB)	17
b. Anaerobic corrosion by microorganisms other than SRI	B <b>20</b>
i. Corrosion by methanogenic archaea	20
ii. Corrosion by Fe (III)-reducing bacteria	20
iii. Corrosion by nitrate-reducing bacteria	21

1.1.3 MIC Mechanisms	21
1.1.3.1 General mechanisms	21
i. Differential respiration aeration cells	21
ii. Corrosive metabolites	23
iii. Microbial alteration of passive films and corrosion	
products	24
1.1.3.2 Sulfate reducing bacteria corrosion mechanisms	24
i. Mechanism of cathodic depolarization	24
ii. Corrosion by H <sub>2</sub> S	25
iii. Mechanism of acid corrosion	26
iv. Mechanism of fixed anode region	26
v. Extracellular polymeric substances (EPS) production by	
SRB	26
1.1.4 Biofilm and its role in MIC	26
1.1.4.1 Biofilm structure and architecture	26
1.1.4.2 Biofilm formation and its stages	29
1.1.4.3 Multiple roles of biofilm in MIC	33
1.1.5 Analytical techniques in testing MIC	34
1.1.5.1. Detective methods	34
1.1.5.2. Electrochemical methods	34
1.1.5.3. Surface analysis methods	35
1.2 Macrobiofouling	35
1.2.1. Macrobiofouling community	37
1.2.2. Macrobiofouling impacts	37
1.3 Electrocemical Corrosion	38
1.3.1 Corrosion forms	39

1.4 Prevention and Control	41
1.4.1 Biofouling control methods	41
1.4.1.1 Physical control methods	41
1.4.1.2 Chemical control methods	41
1.4.1.3 Mechanical control methods	42
1.4.1.4 Thermal control methods	43
1.4.1.5 Coatings	43
1.4.1.6 Biological control methods	43
1.4.2 Prevention and control of chemical corrosion	44
1.5 Green Biocides & Inhibitors as a New Promising Tren	d of
Control	47
Materials and Methods	
2.1 Materials	51
2.1.1 Plant extracts	51
2.1.2 Coupons	51
2.1.3 Microorganisms	51
2.1.4 Culture media	52
2.1.5 Mussels	53
2.1.6 Non-target sea organisms	54
2.2 Methods	54
2.2.1 Extraction procedure	54
2.2.2 Identification of the chemical composition of each extract	55
2.2.3 Surface activity of the extracts	56
2.2.3.1 Surface tension measurement	56
2.2.3.2 Critical Micelle Concentration (CMC)	56
2.2.4 Preparation of coupons	57

0-	4 -	4
เก	nte	nts

2.2.5SRB bioassay	57
2.2.5.1 Enumeration of planktonic SRB	57
2.2.5.2 Weight loss measurement for biocorrosion	
Experiment	58
2.2.6 Macrobial biofouling assay	<b>59</b>
2.2.6.1 Mussels	59
2.2.7 Toxicity against non-target sea organisms	60
2.2.8 Electrochemical corrosion study	60
2.2.8.1 Solutions preparation	60
2.2.8.2 Potentiodynamic polarization measurements	61
2.2.8.3 Weight loss measurements	62
2.2.8.4 Surface analysis	63
2.2.8.4.1 SEM and EDX analyses	63
2.2.8.5 Corrosion products analysis	64
2.2.8.5.1 X-Ray diffraction (XRD) analysis	64
Results	65
3.1 Percentage yield of each extract	65
3.2 Chemical composition of the extracts	<b>67</b>
3.2.1 Gas chromatographic analysis of the extract of waste bitt	er
water of Egyptian lupine	<b>67</b>
3.2.2 Gas chromatographic analysis of orange peels hot	water
extract	68
3.2.3 Gas chromatographic analysis of mandarin peels hot	water
extract	71
3.3 Surface activity of the extracts	77
3.4 Biocidal activity against SRB	<b>79</b>

3.4.1 Biocidal activity against planktoic SRB cultures	<b>79</b>
3.4.2 Biocidal activity against sessile SRB cultures grown on C-	
steel coupons	
3.5 Biocidal Activity against Brachidontes variabilis	91
3.6 Toxicity against non-target sea organisms	94
3.7 Electrochemical Corrosion Control	95
3.7.1 Corrosion inhibition of C- steel in acidic media	95
3.7.1.1 Weight loss measurements	95
a- Effect of time	95
b- Effect of Concentration	97
c- Effect of temperature	100
3.7.1.2 Potentiodynamic polarization measurements	101
3.7.1.3 X-Ray Diffraction Analysis	106
3.7.1.4 Scanning electron microscope (SEM) observations of	f the
surface	107
3.7.1.5 Energy dispersive X-ray analysis (EDX)	109
3.7.2 Corrosion inhibition of C- steel in saline solution	111
3.7.2.1 Weight loss study	111
a- Effect of time	111
b- Effect of concentration	113
c- Effect of temperature	116
3.7.2.2 Potentiodynamic polarization measurements	117
3.7.2.3 X-Ray diffraction analysis	121
3.7.2.4 Scanning electron microscopy (SEM)	122
3.7.2.5 Energy dispersive X-ray (EDX)	124
Discussion	126

Contents
----------

Conclusion	141
English Summary	143
References	146
<b>Arabic Summary</b>	

# LIST OF TABLES

Table (	(1):Oxidation reactions of <i>Thiobacilli</i> (Videla,
1	1996)14
Table (	(2):Corrosive metabolites of microbes inducing MIC
(	(Sand, 1997 and 2003)
Table (	(3):Cathodic depolarization reactions (Von Wolzogen
]	Kuhr and Van der Vlugt, 1934)25
Table (	(4):Chemical composition of C- steel alloy 51
Table (	(5):Chemical composition of the Egyptian lupine bitter
7	water extract as identified by GC/MS67
Table (	(6):Orange peels hot water extract, chemical constituents
8	and their mass fragmentations sorted according to
1	retention time (RT) of their appearance
Table (	(7):Mandarin peels hot water extract; chemical
(	constituents and their fragmentations sorted according to
1	retention time of their appearance73
Table (	(8):Biocidal activity of the extracts against planktonic
1	non-halotolerant Desulfovibrio sapovorans ATCC 33892
8	and the low salinity mixed cultures SRB1 and
9	SRB2 <b>82</b>

Table	(9):Biocidal activity of different extracts against
	planktonic halophilic Desulfovibrio halophilus ATCC
	51179 and the high salinity mixed culture
	SRB3 <b>85</b>
Table	(10):Corrosion rates of C- steel coupons immersed in
	control cultures of ATCC 33892 and in cultures treated
	with each of the three tested extracts
Table	(11):Corrosion rates of C- steel coupons immersed in
	control cultures of ATCC 51179 and in cultures treated
	with each of the three tested extracts
Table	(12): Mortality percentage of Brachidontes variabilis at
	different concentrations of L, O and M extracts92
Table	(13): The safe concentrations of each of L, O and M
	extracts against the tested non-target sea organism94
Table	(14):C- steel weight loss data in 1 M HCl without and
	with different concentrations of tested extracts98
Table	(15):Electrochemical parameters obtained from
	polarization of C-steel in 1M HCl solution with and
	without different concentrations of inhibitors105
Table	(16):C-steel weight loss data in 40,000 mg/L saline
	solution without and with different concentrations of
	tested extracts114