

DEDICATION

This work is dedicated to

My father's soul, Mother,
Sisters,
Family,
And friends.

ACKNOWLEDGEMENTS

Thanks to Allah who gave me the health, strength and patience to complete this study.

I'm grateful to my supervisor Dr. Mohammed Ahmed Hadad for his kind supervision, guidance, suggestions, valuable advices and patience.

I would like to thank Dr.Mohammed Ahmed Salih,Dr. Mohammed Ahmed Adlan, Prof. Abdallah Elhagwa from the Agricultural research corporation for their support .

My appreciation is extended to Prof. Ahmed Eltigani ,Dr. Awad Galal, Dr. Tilal sayed and Elsamawal Merghani .

Special thanks to my M.Sc teaching staff and to all my colleagues.

Finally I would like to thank all my friends for their help and encouragement.

ABSTRACT

The root environment of leguminous crops undergoes constant changes as a result of chemical reactions taking place within the soil. Rhizobia produces root nodules is subject to changes in the soil environmental conditions including salt concentrations, pH and temperature.

In this study, four *Rhizobium* strains have been used for screening their stress tolerating ability. The cultural, microscopic and, biochemical characteristics were studied for all the isolates.

Growth of pure *Rhizobium* strains on Yeast Extract Mannitol Agar (YEMA) medium having variable range of pH (6.0, 8.0 and 10), different concentrations of NaCl (0%, 0.1%, 0.5%, 1 % and 2%) and different incubation temperatures (20⁰C, 30⁰C and 40⁰C) was studied .

The results revealed that the optimum growth for the tested strains was under the low concentrations of NaCl (0% and 0.1% NaCl) .pH 8 was the best pH level for the tested strains The highest population growth for the tested isolates was at 30⁰C.

From the general results under the different stresses strain USDA 3386 was selected as the superior strain as it had the best performance at the different stresses therefore, it can be utilized in the stressed environments.

المستخلص

إن بيئة جذور النباتات البقولية تمر بتغيرات ثابتة كنتيجة للتفاعلات الكيميائية التي تحدث داخل التربة. تخضع البكتريا المنتجة للعقد الجذرية لمختلف التغيرات في بيئة التربة والتي تشمل الملوحة و درجة تفاعل التربة و درجة الحرارة.

في هذه الدراسة تم إستخدام أربعة سلالات من البكتريا لإختبار قدرة تحملها لمختلف أنواع الإجهاد. تمت الخصائص المزرعية, و المجهرية, و الكيموحيوية لكل السلالات.

تمت دراسة العزلات النقية لسلاسل الرايزوبيا في بيئة أجار مستخلص الخميرة و المانيتول بأرقام أس هيدروجيني مختلفة (6 و 8 و 10) و تراكيز مختلفة من كلوريد الصوديوم (0% و 0.1% و 0.5% و 1% و 2%) و مختلف درجات حرارة مختلفة (20 و 30) و 40 درجة مئوية

أوضحت الدراسة أن النمو الأمثل للسلاسل كان تحت التركيزات المنخفضة لملح كلوريد الصوديوم (0.0%) و (0.1%) و أن رقم الأس الهيدروجيني 8 كان الأفضل للسلاسل المختبرة. و كان النمو الأعلى للسلاسل المختيرة في درجة حرارة 30 درجة مئوية

من النتائج العامة و تحت ظروف الإجهاد التي تمت دراستها تم إختيار سلالة البكتيريا لأنها الأكثر امتيازاً تحت الظروف المختلفة و أنها المثلى للتلقيح في USDA3386 الظروف البيئية المجهدة.

LIST OF CONTENTS

No	LIST OF CONTENTS	Page . No
	Dedication	I
	Acknowledgements	II
	Abstract	III
	Arabic Abstract	IV
	List of Contents	V
	List of Tables	VI
	List of Figures	VI
	List of Photos	VII
	CHAPTER ONE: INTRODUCTION	1
	CHAPTER TWO: LITERATURE REVIEW	3
2.1	Rhizobia	3
2.2	Importance of Rhizobia in Agriculture	3
2.3	Nitrogen Fixation	4
2.3.1	Non Biological Nitrogen Fixation	4
2.3.2	Biological Nitrogen Fixation(BNF)	4
2.4	Tolerance of Rhizobia to pH	6
2.5	The Mechanism of pH Tolerance in Rhizobia	8
2.6	Tolerance of Rhizobia to Salinity	9
2.7	The Mechanism of Salt Tolerance in Rhizobia	14
2.8	Tolerance of Rhizobia to Temperature	17
2.9	The Mechanism of Temperature Tolerance in	20

	Rhizobia	
	CHAPTER THREE: MATERIALS AND METHODS	22
3.1	Sources of Materials	22
3.2	Cultural Characteristics	22
3.3	Microscopical Characteristics	22
3.3.1	Gram Staining	22
3.3.2	Motility Test	23
3.4	Biochemical Characteristics	23
3.4.1	Growth on YEMA Containing Congo Red	23
3.4.2	Catalase Test	23
3.4.3	Growth at Different NaCl Concentrations	24
3.4.4	Growth at Different pH Levels	24
3.4.5	Growth at Different Incubation Temperatures	24
	CHAPTER FOUR: RESULTS AND DISCUSSIONS	25
4.1	Morphological, Microscopical and Biochemical Characteristics of the tested <i>Rhizobium</i> Strains	25
4.2	<i>Rhizobium</i> Strains Tolerance to Salt	25
4.3	<i>Rhizobium</i> Strains Tolerance to pH	30
4.4	<i>Rhizobium</i> Strains Tolerance to Temperature	33
4.5	Conclusions	37
	REFERENCES	39
	APPENDIXES	5

List of Tables

No.	List of Tables	Page No.
4.1	Morphological, Microscopic and Biochemical Characteristics of the Tested <i>Rhizobium</i> Strains	25
4.2	Effect of Different Salt Concentrations on The Tested <i>Rhizobium</i> Strains	27
4.3	Effect of Different pH Levels on The Tested <i>Rhizobium</i> Strains	31
4.4	Effect of Different Temperatures on The Tested <i>Rhizobium</i> Strains	35

List of plates

No.	List of plates	Page No.
4.1	Effect of Different Salt Concentrations on The Tested <i>Rhizobium</i> Strains	29
4.2	Effect of Different pH Levels on The Tested <i>Rhizobium</i> Strains	33
4.3	Effect of Different Temperatures on The Tested <i>Rhizobium</i> Strains	37