

## ABSTRACT

**Manal Abd El Mottela. Quality attributes based on activities of selected enzymes and Inhibitors in some processed food. Unpublished Doctor of philosophy Dissertation. Ain Shams University, Faculty of Agriculture, Food Science Department, 2002.**

It was aimed through the scope of the study to look forward about the effect of some food additives on the activity of oxidoreductase and hydrolase enzymes in some processed food stuffs. The investigated enzymes namely polyphenolase and peroxidase representing the former group, while the pectin methyl esterase was chosen to represent the latter one; Also, Alfa amylase, protease and lipase were determined in other experiments as a measure factor affecting the quality of some processed food as a result of adding different food additives.

The processed food being under test within the research at hand were potatoes, garlic, artichoke. These aforementioned samples were treated with different concentration (100, 200, 400 ppm) of organic acid (ascorbic, sorbic and citric) for different periods (15,45, 90min) then blanched at 95°C for 3 min.and frozen at -18°C. The activitiesof these enzymes were studied after the previous treatment.

Also, processed mayonnaise was made and treated with BHA (200 ppm), or BHT (100 ppm) or mixture of both and kept for 2 months at -18°C and the activity of lipase and protease enzymes was followed.

Orange,guava and pineapple juices, and apple yoghurt were prepared and the sucrose was replaced by sweeteners (Aspartame, Saccharine and Acessulfam-k) in a concentration of (500, 300, 500 ppm) in order to check out their effects on the activity of pectin methyl esterase(PME).

On the other hand, strawberry jam was prepared after adding 300 ppm (Aspartame, Saccharine and Acesulfam-k) then kept for one week at -4°C and the enzyme activity was determined to investigate the regeneration pattern of the enzyme.

Frying of potatoes and fish was performed in oil containing antioxidants BHA and BHT to prove the role of antioxidants on the enzyme activities of these fried samples which compared with a control sample.

Other treatments were tried within the scope of the study including:

- The effect of adding BHA & BHT(300 ppm) to minced frozen fish and shrimp on the activity of protease & lipase.
- The effect of probionic acid (2 or 3 g / kg flour) on the activity of the  $\alpha$ -amylase enzyme in some fermented pizza dough was investigated .
- The effect of adding sodium nitrite (150 ppm) on the protease and lipase activities in beef burger was also considered.
- The effect of benzoic and sorbic acids on the activities of PME enzyme in strawberry prepared Juice was studied.

**Key words :** BHA, BHT, (PPO), (POD), Protease, Lipase, Alpha Amylase, Pectin Methyl Esterase (PME), Potato, Artichoke, Garlic, Sodium nitrite, Benzoic acid, Sweeteners, Saccharine, Aspartame, Acesulfam-k, Citric acid, ascorbic, Sorbic, Juices, Pizza dough , Strawberry Jam, Beef burger, Fish, Shrimp and Mayonnaise.

# CONTENTS

	page
LIST of TABLES	V
LIST of FIGURES	X
LIST of ABBREVIATIONS	XII
INTRODUCTION	1
REVIEW of LITERATURE	5
<b>Part I :Inhibition Patterns of Nitrite, Nitrate and Propionic acid.</b>	5
1-Nitrite and nitrate.	5
2-Propionic acid:	9
<b>Part II : Inhibition Pattern of the Antioxidants (BHA and BHT).</b>	10
1-Mayonnaise.	10
2- Frying oil.	12
3- Fish and Shrimp.	15
<b>Part III : Inhibition Pattern of The Investegated Organic Acids:</b>	21
1-Potato	21
2-Artichoke.	31
3-Garlic.	32
<b>Part IV : The Sorbic and Benzoic Acids:</b>	35
1-Strawberry Juice.	35
<b>Part V : The Sweeteners.</b>	37
<b>Materials and Methods</b>	45
<b>I-Materials</b>	45
<b>II -Analytical Methods.</b>	52
II-1 -Major chemical constituents	52
II-2- Minerals analysis	52
II-3-Amino acids	52
II-4-Antioxidants (BHA and BHT).	53
II-5-Determination of acesulfame- k ,aspartame and saccharine	54
II-6-Propionic acid	55
II-7-Ascorbic acid determination	55
II-8-Benzoic and sorbic acid	56

II-9-Determination of nitrites .	56
II-10- Determination of Citric acid:	57
II-11- Measuring of Enzymes activities	57
II-11-1-Lipase .	57
II-11-2- Protease.	57
II-11-3- Activity of pectin methyl esterase (PME)	57
II-11-4- polyphenol oxidaes (PPO) and peroxidase (POD)	58
II-11-5- Alpha Amylase	59
<b>RESULT AND DISCUSSION</b>	60
<b>Part I: Activity of protease, lipase and alpha amylase in beef burger and pizza samples .</b>	60
-1- Activity of lipase and protease enzymes in beef burger untreated and treated with nitrite salt.	60
-2- Activity of Alpha amylase enzyme in Pizza dough untreated and treated with propionic acid.	66
<b>Part II: Activity of protease and lipase in mayonnaise, fish, and shrimp samples.</b>	72
II-1-Activity of lipase and protease in mayonnaise samples untreated and treated with BHA and BHT.	72
II-2-Quality of french fried potatoes and fried Trout fish as influenced by corn oil treated with BHA and BHT.	77
II-3-Changes in amino acids pattern in Salmon, Trout and shrimp samples in relation to the addition of antioxidants BHA and BHT.	89
3-1-Salmon, Trout fish and shrimp samples	90
3-1-1-Salmon samples under investigation	90
3-1-2-Identified amino acids in the untreated salmon samples	91
3-1-3- Salmon samples treated with 300 ppm of BHA	95
1-3- Salmon treated with 300 ppm of BHT	100
1-2-2- Trout samples treated with and without 300 ppm of BHA or BHT	
1-2-2-a- Untreated Trout (control)	106
1-2-2-b-Trout samples treated with 300 ppm of BHA	106
	107

1-2-2-C-Trout samples treated with 300 ppm of BHT :	109
3-1- Shrimp samples treated with and without 300 ppm of BHA and BHT:	112
3-1-a- Shrimp samples treated with 300 ppm of BHA:	113
3-2- Shrimp samples treated with 300 ppm of BHT:	114
3-3-Activities of lipase and protease	118
<b>Part - III: Chemical Constituents, Activity of PME, POD and PPO in potato, artichoke and garlic samples.</b>	121
A-Potato :	121
A-1-Chemical constituents:	121
A-2-Amino acids.	123
A-3- Soaking in organic acids:	125
A-4-Activity of PME,POD and PPO:	134
B-Artichoke:	142
B-1-Chemical constituents.	142
B-3- Soaking in organic acids:	146
B-4-Activity of PME,POD and PPO:	151
C- Garlic samples under test:	160
C-1- Chemical constituents.	160
C-2-Soaking in organic acids	164
C-3- Amino acids:	166
C-3-Enzymes activity:	
<b>Part-IV: Treatment of fruit products with organic acids or sweeteners.</b>	174
IV:A-Treatment of strawberry Jam with 500 ppm of benzoic and sorbic acids.	174
IV-A-1- Residual amounts of benzoic and sorbic acids:	175
IV-A- Activity of pectin methyl esterase(PME)	177
IV:B-Treatment of fruit products with sweeteners (Saccharine, Aspartame and Acesulfam-k) .	179
IV-B-1- Chemical constituents of fruit juices	181
IV-B-2-Treatments with sweeteners	183

- SUMMARY AND CONCLUSION.	189
- REFERENCES.	202
- APPENDIX 1.	220
- APPENDIX 2.	232
- ARABIC SUMMARY.	

## LIST OF ABBREVIATIONS

AOAC	association of official analytical chemists
AA	Amino acid
aw	Water activity
BHA	butylated hydroxyanisole
BHT	butylated hydroxytoluene
BV	biological value
CCMP	Cooked crude meat pigment
Cs	Chemical score
°C	degree Celsius(centigrade)
E	enzyme
EAAI	Essential amino acid index
EC	enzyme commission
Enz.con	enzyme concentration
EPR	electron paramagnetic resonance
ES	enzyme substrate complex
F	Fahrenheit.
FAO	food and agriculture organization.
FDA	food and drug administration.
Fig.	figure
GATT	general agreement on tariffs and trade.
GMP	good manufacturing practices
GRAS	general recognized as safe
HPLC	high-performance liquid chromatography
HACCP	hazard analysis and critical control point
J	joule
JECFA	joint FAO/WHO Expert committee on Food Additives.
MIN	minutes
NAS	National Academy of Sciences
N.B.	note bene
Nm	nanometer( $10^{-9}$ )

NO.	number
NOC	N-nitros Compounds
NPN	Non Protein Compounds
PME	pectin methylesterase
PPO	poly phenol oxidase
POD	peroxidase
QE	Quality enhancement
S.D	standard deviation
S.E.	standard error
SEC.	second
SPE	Solid phase extraction
T	absolute temperature
TDS	Total dite study
$\mu$	micron(0.001 mm)
$\mu$ G	micro gram (10-6g)
$\mu$ L	microlitre (10-6L)
$\mu$ V	ultraviolet
ppm	part per million
VOL.	volume
WHO	world health organization