CHEMICAL CHARACTERIZATION AND BIOLOGICAL EVALUATION OF ALGAL PEPTIDES

By

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B.Sc. Agric. Sci. (Biochemistry) Fac. Agric., Ain Shams Univ., 2002
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THESIS

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APPROVAL SHEET

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Title of Thesis: Chemical Characterization and Biological Evaluation of Algal Peptides
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ABSTRACT

This study was conducted to study the production of peptides from *Spirulina plarensis* and *Scenedesmus obliquus*, their biological activities as antioxidant, antiviral, and anticancer were also investigated.

Protein was extracted from *Spirulina platensis* by three different extraction methods (Sp1, Sp2 and Sp4). Protein extracts and hydrolysates were assessed by gel electrophoresis (SDS–PAGE) of samples and marker. Algae cells and protein extracts were rich in Arg, Lys, Asn, Ala and His. Antioxidant activity of protein hydrolysates from Spirulina was investigated. Protein hydrolyzed by trypsin (Sp2Try) and (Sp4Try) showed the highest antioxidant activity against 1, 1-diphenyl-2-picryl-hydrazyl (DPPH) radical-scavenging (32.27% and 30.11%,respectively) and on 2, 2'-azinobis 3-ethyl- Benzothiazoline-6-sulphonate(ABTS) radical (56.39% and 34.26%, respectively) at sample concentration (150 µg/ml).

Protein was extracted from *S. obliquus* by three different extraction methods. Protein extracts and hydrolysates were assessed from stained gels following SDS–PAGE of samples. Antioxidant activity of protein hydrolysates was investigated. Protein hydrolyzed by papain (Sd1pa) and protein hydrolyzed by trypsin (Sd2Try) induced highest antioxidant activity based on 1,1-diphenyl-2-picryl-hydrazyl (DPPH) radical- scavenging (41.41% and 40.62%) respectively, and on 2,20-azinobis 3-ethyl- benzothiazoline-6-sulphonate (ABTS) radical (87.03% and 45.12%) respectively, at 150 mg/ml. The inhibitory effect and mode of action of protein hydrolysates were evaluated against Coxsackie B3 virus (CVB3). Protein hydrolyzed by papain (Sd2pa) and protein hydrolyzed by pepsin (Sd1pep) at 100 mg/ml exhibited antiviral activity (66.2% and 57.6%, respectively), against (CVB3) from all protein hydrolysates. *S. platensis* and *S. obliquus* protein hydrolysates can be used as nutritional food with antioxidant properties. *S. obliquus* protein hydrolysates have a potential as antioxidative neutraceutical ingredients and a potential therapeutic agent against CVB3.

The sequence of peptides obtained from *Spirulina* and *Scenedesmus* were determined by using MALDI-Triple TOF method.

Key words: *Spirulina platensis*, *Scenedesmus obliquus*, algal peptides, antioxidant activities, antiviral activities**,** cytotoxic activities, MALDI-TOF.

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LIST OF ABBREVIATIONS

GLA: Gamma linolenic acid.

FDA: Food and Drug Administration.

DNA: Deoxyribonucleic acid.

NaCl: Sodium chloride.

SCPs: Single cell protein.

NR: Nitrate reductase.

NiR: Nitrite reductase.

GS: Glutamine synthetase.

EAA: Essential amino acids.

GRAS: Generally Recognized as Safe.

ROS: Reactive oxygen species.

$O2^{\bullet-}$: Superoxide anion radicals.

$OH^{\bullet}$: Hydroxyl radicals.

$NO^{\bullet}$: Nitroxide radicals.

H_2O_2: Hydrogen peroxide.

$ROO^{\bullet-}$: peroxyl radicals.

$ONOO^{-}$: Peroxynitrite anion.

GSH: Glytathione.

GSSG: Glutathione disulfide.

SOD: Superoxide dismutase.

CAT: Catalase.

GPX: Glutathione peroxidase.

PrxR: Peroxiredoxin.

TOH: Tecopherols.

AA: Ascorbic acid.

DPPH: 1, 1- diphenyl-2-pricrylhydrazyl.

ESR: Electron spin resonance.

EP: Ethanol-soluble proteins.

ACPs: Anticancer peptides.

AMPs: Antimicrobial peptides.

ACE-I: Angiotensin converting enzyme I.

RAAS: Renin angiotensin aldosterone system .

HIV-1: human immunodeficiency virus.

HSV-1: herpes simplex virus types 1.

HSV-2: herpes simplex virus types 2.

PAF-AH: Platelet activating factor acetylhydrolase.

RP-HPLC: Reversed-Phase High-Performance Liquid Chromatography

POP: Prolyl oligopeptidase.

PEP: Prolyl endopeptidase.

GABA: Gamma-Aminobutyric acid.

GABAergic: Any chemical that modifies the effects of GABA in the body or brain.

MS: Multiple Sclerosis.

AD: Alzheimer's disease.

GIP: Gastric inhibitory polypeptides.

DPP-IV: Dipeptidyl peptidase-4.

T2D: Type 2 Diabetes.

ABTS: 2, 2'-azino-bis (3-ethylbenzothiazoline-6-sulphonic acid.

BHT: Butylated hydroxytoluene.

Trolox: 6-hydroxy-2, 5, 7, 8-tetramethylchroman-2-carboxylic acid.

AFP: Alpha-fetoprotein.

BH3: Interacting-domain death agonist.

BCL-2: B-cell lymphoma 2.

ABH3: Artificial BCL-2 homology 3 peptides.

BR2: Cancer Specific Cell-Penetrating Peptide.

GI: gastrointestinal.

pH: the decimal logarithm of the reciprocal of the hydrogen ion activity.

SP: *Spirulina Platensis*

BBM: Bold basal medium.

Lux: Unit of illuminance.

SGR: Specific growth rate.

OD: Optical density.

NaOH: Sodium hydroxide.

HCl: Hydrochloric acid.

TCA: Trichloroacetic acid.

PBS: Phosphate buffer solution.

K_2SO_4: Potassium sulfate.

BSA: Bovine serum albumin.