

**Study the Bioactive Peptides Extracted
from Medicinal Plants and its Effect on
Bacterial Isolated of Food Spoilage**

Abstract

The mechanisms by which bioactive peptides from medicinal plants inhibit pathogenic microorganisms causing food spoilage are very important, and the world is currently moving towards their use. Medicinal plants include Fenugreek (*Trigonella foenum graecum*), Chia (*Salvia hispanica*), Quinoa (*Chenopodium quinoa*), and Colocynth (*Citrullus colocynthis*) fruit, a promising natural source of bioactive peptides responsible for antibacterial activity. One of the main causes of food waste as well as the harmful effects of food production and processing on environmental sustainability was microbial food spoiling. In This study used peptide isolated from specific medicinal plants to generate bioactive peptides with antibacterial properties. From November 2023 to January 2024, bacteria were isolated and identified from spoiled foods in the laboratories of AL-Muthanna University's College of Science using various culture media and biochemical tests. Proximate analysis showed the sample's moisture, ash, crude protein, and lipid content. Amino acid analyzer was used to identify amino acids in the samples, while High-Performance Liquid Chromatography (HPLC) was employed to identify and quantify the sample's phenolic and flavonoid compounds and fatty acids. The bioactivity of these peptides was evaluated against bacterial pathogens and spoiling food bacteria, using various antibacterial assays.

The results isolated and identified four types of food spoilage and pathogen bacteria, including (*Staphylococcus aureus*, *Bacillus cereus*, *Pseudomonas aeruginosa*, and *Escherichia coli*), using colony shape characteristics and biochemical tests. Proximate composition analysis showed that Fenugreek, Chia, Colocynth fruit, and Quinoa had a moisture content of (3.32, 5.57, 3.24, and 2.47%), ash content of (3.94, 5.14, 3.52, and 2.61%), crude protein of (28.17, 20.76, 24.02, and 12.98%), and crude lipid of (7.86, 15.63, 3.92, and 6.28%) respectively. Study plants contain sixteen amino acids, tin

phenolic, and flavonoid compounds, including Asparagine, Lysine Serine, Threonine, Isoleucine, Alanine, Valine, Tyrosine, Arginine, Cysteine, Methionine, Proline, Histidine, Lucien, Phenylalanine, and Glycine. Phenolic compounds include (Gallic acid, Rutin, Apigenin, Quercetin, Coumaric acid, Ferulic acid, Caffeic acid, Chlorogenic acid, Kaempferol, Syringic acid). Six fatty acids including Palmitic acid, Stearic acid, Linolenic acid omega-3, Oleic acid, Linoleic acid omega-6, Arachidonic acid. Peptides extracted from the protein of the study plants after 24 hours showed high antibacterial activity.

Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC) tests were employed to determine the lowest concentration of these peptides that inhibit or kill the bacteria. Additionally, agar disc diffusion assays were performed to measure antibacterial activity by the formation of inhibition zones. The higher antibacterial assay percentage was found in Feungreek seeds to be at 96.53 % against *B. cereus* while the lowest percentage in Colocynth fruit was at 50.28 %. Based on the result obtained for MIC, the concentration of 3 mg/mL exhibited inhibitory against all four types of bacteria for Chia, Quinoa, Fenugreek, and Colocynth. When the concentration of peptides decreases, the antibacterial activity decreases. The disc assay demonstrated clear inhibition zones; In conclusion, Chia, Quinoa, Fenugreek, and Colocynth are a good source of antibacterial agents since it contain metabolites (bioactive peptides). Thus, it can be used to improve the microbial quality of food and extend the shelf life of food.