Identification of bell pepper (*Capsicum annum* L.) microbiota and assessment of antibiotic resistance of foodborne bacteria.

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Abstract of contents

Antibiotic resistance among foodborne bacteria has emerged as a significant public health concern in recent years. The widespread use of antibiotics in agriculture, animal husbandry, and food production has led to the selection and proliferation of antibiotic-resistant strains of bacteria in the food supply chain. These resistant bacteria pose a threat to human health, as they can contaminate food products and cause infections that are difficult to treat with conventional antibiotics. Understanding antibiotic resistance in foodborne bacteria is crucial for planning effective strategies to control its spread and safeguard public health. Additionally, ongoing exploration of emerging pathogens in food production and their response to commonly used antibiotics is essential. The aim of this study was to isolate and identify microorganisms from the phyllosphere of the bell pepper plant (*Capsicum annuum* L.) and examine their antibiotic resistance patterns to determine the MIC (Minimum Inhibitory Concentration) value. Also, samples from raw pork meat and raw milk ere were consider into de assessment of the antibiotic resistance as part of a PhD research. The antibiotic used was Oxytetracycline-hydrochloride.

This research describes the identified microbiota of bell pepper phyllosphere and investigates the antibiotic resistance of bacteria that have been isolated from raw pork meat, raw milk, and bell pepper phyllosphere; Food samples were collected and processed to isolate bacteria, followed by microscopic investigations and biochemical identification methods (Oxidase, Catalase, and Potassium hydroxide tests) to characterize the bacterial strains. The bacteria selected were identified in genus and species level using MALDI-TOF-MS. The isolates were subjected to antibiotic susceptibility testing using the

diffusion test methods with Oxytetracycline-hydrochloride antibiotic at four different concentrations between 0,375 µg/µl – 3,00 µg/µl to assess their resistance profiles and microdilution tested by Spectroscopy UV/Vis (Multiskan Ascent Microplate Reader) to find the MIC (Minimum Inhibitory Concentration) value.

The findings reveals that the microbiome of bell pepper phyllosphere is diverse and that the incidence of these microbes depends in several factors like the environment, fertilization, source of irrigation water, coworker handling and source of compost. Among the microorganisms found in the bell pepper phyllosphere are *Brachybacterium conglomeratum, Staphylococcus saprophyticus, Pseudomonas extremorientalis* and *Pseudomonas oryzihabitants* in the fruit*; Staphylococcus warneri, Micrococcus luteus, Moraxella osloensis, Bacillus cereus* and *Staphyloccoccus hominis* in the bottom leaves*; Microbacterium arborescens, Pseudomonas flavescens* and *Pluralibacter pyrinus* in top leaves; *Bacillus cereus, Microbacterium arborescens, Pseudonomas antarctica, Pseudomonas fulva* and *Pseudonomas oryzihabitants* in flowers. Some of the microorganisms mentioned before are naturally occurred as commensals in the human body and that haven been reported as causing agents of many infections among the immunocompromised people, which become a concern in the public health, because those microorganisms are not commonly reported in food sources, and now should be considered as part of the surveillance emerging pathogens to prevent foodborne diseases.

Regarding to the assessment of antibiotic resistance, two microorganisms from each food source were selected *Micrococcus luteus and Pseudomonas antarctica from bell pepper phyllosphere; Kocuria salsicia and Macrococcus caseolyticus from raw milk; Buttiauxella gaviniae and Pseudomonas lundensis from raw pork meat;* Which highlighting the diverse microbial populations present in the food samples, due to the different environments that the food is exposed along the production chain. Antibiotic susceptibility testing revealed that *Macrococcus caseolyticus* was the most resistant bacteria among the others it showed minor inhibition zones (8-9 mm) at different concentrations of the antibiotic, in contrast to *Micrococcus luteus* which was the most sensitive microorganism with large inhibition zones (29-43 mm) at all applied concentrations. *Pseudomonas antarctica, Kocuria salsicia, Buttiauxella gaviniae* and *Pseudomonas lundensis* showed similar resistance with inhibition zones of 19-37 mm. Based on the microdilution test carried out by Multiskan, the growth modelling curves revealed that all the isolates were susceptible to the antibiotic oxytetracycline-hydrochloride.

These findings underscore the importance of intensify the vigilance of the good agriculture practices to prevent the transmission of potential pathogens to the produces. Also, monitoring antibiotic resistance in foodborne bacteria and implementing measures to mitigate its spread should be part of the prevention practices, such as prudent antibiotic use in agriculture and food production, as well as surveillance programs to track resistance trends. Continued research in this area is vital for safeguarding public health and ensuring the efficacy of antibiotic treatment in foodborne illnesses, encouraging to continue with this study with Multiskan using lower antibiotic concentrations.