CHAPTER 1

INTRODUCTION

1.1 Background

The benefits of consuming fresh fruits and vegetables and minimally processed fruits and vegetables have long been known and largely advocated. Among the benefits are high moisture and nutrients contents such as vitamins, minerals, fibres, and antioxidants (Mendoza et al., 2022). However, the open nature of fresh produce production makes it susceptible to contamination from multiple sources (Murray et al., 2017). The more steps in the handling and processing of a product, the more obvious physiological changes will be in that product, leading to shortened product shelf-life and enhance the growth and survival of microbes associated with the product (Matthews, 2014).

Food safety is of great concern as the impact brought about by the consumption of unsafe foods affects not only human health but also socioeconomic development (Havelaar et al., 2015). Food may be contaminated with many foodborne causative agents, such as pathogens, biotoxins, man-made physical and chemical toxicants (e.g., pesticides and metals) during any point in the food supply chain such as production, processing, transport, and storage (S. Li et al., 2020).

Water spinach (*Ipomoea aquatica* Forssk.) is a leafy vegetable from the Convolvulaceae family (USDA, 2019). It is a staple vegetable in Southeast Asia and parts of China (Ebert, 2011). In terms of nutritional contents, water spinach contains carbohydrates and minerals. The investigation of mineral contents found high content

of potassium, magnesium, sodium, and phosphorus in the leaves and stems of *Ipomoea aquatica* Forssk. (Ndamitso et al., 2015). Bioactive compounds such as flavonoids and phenols provide anti-oxidative properties and anti-inflammatory properties (Mariani et al., 2019). When kept post-harvest in cold room with temperature between 12-14°C, the shelf life is between 10-12 days with RH values between 90-95% (Chen & Paull, 2015).

Postharvest treatment of vegetables is expected to effectively reduce microbial load, extends shelf-life by preserving the texture, visual appearance, flavour, and nutritional value (L.-Z. Deng et al., 2017). Ultrasonic treatment is one of the postharvest treatments to decontaminate fresh produce. The high-frequency waves induce acoustic cavitation, that destroys the microbial cell walls and damages the DNA via free radical production (Gallo et al., 2018; Gao et al., 2014; Mendoza et al., 2022). Physical contaminants such as soil and residues of food are also removed by localised chemical and mechanical action of the cavitation (São José et al., 2014). Inhibition of peroxidase and polyphenol oxidase (PPO) by ultrasonic treatment is the key to prolong shelf-life of fruits and vegetables (W. Zhou et al., 2022).

1.2 Problem Statement

Water spinach is the second most produced vegetables in Malaysia in 2023 (Jabatan Pertanian Malaysia, 2023). Despite the high consumption by Malaysians, studies on improving the quality and safety of water spinach post-harvesting are scarcely available. Therefore, this study aims to utilise the potential of ultrasonic treatment to reduce and eliminate pesticide residues, physical and microbial contaminants while preserving the quality of water spinach.

Pesticides residues are the most common chemical contamination found on vegetables, with up to 97.6% of Malaysian farmers preferring its use in pest control

(Halimatunsadiah et al., 2016). Ingestion of chemical pesticide residue by human may lead to diseases such as respiratory disease, neurodevelopmental disorders, cardiovascular diseases and cancer (Chiu et al., 2019; Hertz-Picciotto et al., 2018).To remove pesticide contaminants, usually vegetables are washed with water, however most pesticides are hydrophobic and not easily removed by this method alone (Qi et al., 2017). Postharvest chemical treatment can be the solution to this problem because of the absence of physical handling. However, the disadvantages include absorption of accumulated harmful byproducts such as chlorates and trihalomethanes (Rodrigues Gadelha et al., 2019), mould growth from residual moisture post-treatment (L.-Z. Deng et al., 2017) , reduced efficacy in presence of multiple organic compounds in the washing liquid (Nguyen-the & Carlin, 1994) and consumers' hostility towards synthetic chemicals (Mesías et al., 2021; Rico et al., 2007). A study by Md Sa'at (2020) to determine presence of organophosphate pesticide residue on fresh vegetables sold in hypermarkets in Negeri Sembilan, Malaysia highlighted presence of diazinon residue in water spinach. Therefore, the ability to remove organophosphate pesticide residue, particularly diazinon from water spinach by applying ultrasonic treatment needs to be studied.

Leaves with low cuticle thickness and categorised as non-succulent such as spinach, mustard leaves, and water spinach are especially fragile (M. Zhang et al., 2013). Any excessive handling of these vegetables may damage the physical structure. Physical treatments on leafy vegetables to remove or reduce contamination level involving heat such as sterilisation and blanching usually causes physiological effects such as tissue softening or alteration in biochemical compositions, namely chlorophyll degradation and loss of nutrients (L. Deng et al., 2019). Therefore, non-thermal physical decontamination treatment such as ultrasonic treatment is gaining interest. However,

due to the broad definition of leafy vegetables, often succulent leafy vegetables and tender green leafy vegetables are treated with the same parameters although there are variations in succulence, cuticle thickness and leaf cell types (Radovich, 2018).

Ultrasonic treatment has been shown to delay the deterioration of the physical quality of some vegetables such as lettuce (S. M. R. Azam et al., 2021), tomato and pak choi (Jiang et al., 2020). However, there is lack of information on water spinach quality preservation in the existing literature to conclude that using ultrasonic treatment for making the vegetable safer for consumption will not affect the physical quality negatively. Therefore, suitable parameters for postharvest ultrasonic treatment on tender green leafy vegetables such as water spinach and herbs must be determined to effectively remove contaminants while preserving the physical quality.

Agricultural factors (pre-harvest) and postharvest factors can introduce microorganisms to leafy vegetables and cause contamination (Luna-Guevara et al., 2019). Microbial contamination of vegetables has yet to cause foodborne illness outbreak in Malaysia, however a study by Kuan et al. (2017) found that from both organic and conventional vegetables sampled in Malaysia, mesophilic aerobic bacteria count ranged from 5 to 7 $\log_{10}$ CFU/g while total coliforms counts were recorded between 1 to 7 $\log_{10}$ CFU/g. *Salmonella spp.* prevalence in raw vegetables was 21.5%, with 8 samples detected the presence out of 26 water spinach samples (31.0%) (Abatcha et al., 2018). During washing, some microorganisms will be removed from the product, however, there is still risk of spread from contaminated parts to non-contaminated ones (Nousiainen et al., 2016). With no proper postharvest treatment, high microbial availability in vegetables may be the silent cause of foodborne illness either through direct consumption or cross-contamination with other food types. Hence, postharvest

ultrasonic treatment could potentially reduce microbial availability in vegetables prior to consumption and in turn reduces the possibility of foodborne illness due to pathogenic contamination from vegetables.

1.3 Objectives of Study

The objectives of this study are:

1) To quantitate pesticide (diazinon) concentration on *Ipomoea aquatica* Forssk. in the pre-treatment (fresh (F)), post-treatment (immersed (C) and ultrasonic treatment (U)) at 1 minute, 7 minutes and 15 minutes duration.
2) To evaluate the physical quality of *Ipomoea aquatica* Forssk. treated with ultrasonic treatment based on the photographic comparison, colour, and texture.
3) To evaluate the differences in microbial availability between the pre-treated and post-treated *Ipomoea aquatica* Forssk. using Total Plate Count (TPC) method.

1.4 Significance of Study

There is currently no known study done on ultrasonic postharvest treatment on water spinach. This research aspires to provide data on how water spinach responds to ultrasonic treatment in terms of pesticide reduction level, physical quality and microbial availability. In addition, very little number of studies are published on leafy vegetables' postharvest treatment by ultrasonic treatment. This study seeks to fill in the gaps in the literature knowledge and provide information and possibilities for further expansion in research.

1.5 Scope of Study

This research focuses on the removal of selected chemical contaminant (diazinon pesticide residue) and microbial contaminants from leafy vegetable, which is water spinach by ultrasonic treatment. The effects of ultrasonic treatment on physical qualities of water spinach such as physical appearance, colour and texture are also studied.

Water spinach was purchased from selected hypermarkets in Nilai, Negeri Sembilan. Purchased water spinach was treated with ultrasonic waves in water bath using different times (1, 7 and 15 minutes), immersion in water without ultrasonic waves (positive control) and untreated (negative control). QuEChERS (quick, easy, cheap, efficient, rugged and safe) extraction method was executed, and the pesticide residue level determination was done by chromatographic analysis by gas chromatography flame ionization detector (GC-FID). Physical appearance was compared by digital imaging while colour changes were measured by colorimetric analysis. Texture was represented as leaf firmness measured by texture analyser. Microbial availability was measured by total plate count (TPC).