ANTIOXIDANT POTENTIAL OF *CUCURBITA MAXIMA* EXTRACT AS AFFECTED BY PRETREATMENT, SOLVENT EXTRACTION AND DRYING PROCESS

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Abstract

Pumpkin (*Cucurbita maxima*) is one of the classified fruits that contains a lot of beneficial properties that could contribute to health. However, most usage of pumpkin from its fresh form is prone to microorganisms' spoilage. The main purpose of this study was systematically to evaluate the Total Phenolic Contents (TPC) of pumpkin extracts as affected by pre-treatment, solvent extraction and drying process. Therefore, four different objectives were applied. The first objective is conducted by oven-drying with different temperatures (0°C, 40°C and 80°C). The dried extracts of 40°C and 80°C are compared to untreated or fresh pumpkin (0°C). The second objective is carried out by using different methods of drying (oven-drying, sun-drying and shade-drying). The TPC of these dried pumpkins were evaluated. The third objective is conducted by using different green solvents (ethanol, aqueous ethanol and methanol). The concentration of extracted bioactive compounds is greatly influenced by the solvent used to extract bioactive compounds from the pumpkin since the compounds have different properties and polarities. The last objective is carried out by using different blanching methods (blanching, non- blanching). Blanching method was applied to preserve the bioactive compounds in the pumpkin and the results were monitored. Results obtained for every objective respectively; in the first objective, oven-drying at 40°C increased the value of TPC. However, at 80°C the TPC were enhanced significantly compared to the fresh ones. In the second objective, all three drying methods caused loss of TPC. However, the least reduction of TPC can be seen by oven-drying at certain temperature. Next, in the third objective, aqueous ethanol has the highest TPC compared to ethanol and methanol. The last objective shows that blanching method helps in preserving the bioactive compounds. This can be proved by the result in which blanching have the highest TPC compared to non-blanching. Through the ANOVA statistical analysis, objective 1 has the highest F-value with 70883.93 which shows that temperature gave significant effects of TPC compared to solvent extraction with F-value of 3700.5 and followed by different drying methods with F-value of 56.43. Pre-treatment has the least effects on TPC with the lowest F-value which is 5.09.

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1. Background of Study

1.1 Literature Review

Pumpkin typically refers to Cucurbitaceous family and botanically, it is in the category of squash fruit, most frequently exist orange in colour when ripe, and worldwide used both as human and animal feed (Adiletta et al., 2018). The colour of the orange fruit is due to the abundance of beta carotene. Pumpkins, as the other squash, originated in north eastern Mexico and southern United States. Beyond its exquisite taste, pumpkin is nutritious and related to many health benefits as it is considered as full of nutrition. For health benefits, pumpkin has an impressive nutrient profile. It is classified as highly nutritious and particularly rich in vitamin A (*9 Impressive Health Benefits of Pumpkin*, n.d.), where this vitamin is very beneficial for eye protection from night blindness (nyctalopia) and develop high immune system (Helen West, 2018). Moreover, pumpkin fruit also able to soothe viral infections and inflammations and widely used in food technology as well reported by (Kulczyński et al., 2020). In cooking applications, pumpkins are very versatile where they are eaten mashed and making its way into soups and purées. Sometimes, the seeds are roasted and eaten as snacks. The leaves of the pumpkin plant are served in soups or as cooked vegetable. In addition, raw pumpkin can be fed to poultry during the winter to help maintain egg production. Pumpkin fruits acquire high amount of beta carotene, which also help to improve memory and cognitive function (Kulczyński et al., 2020). From these lists of health promoting behaviour of pumpkin, the benefits gained is because of Total Phenolic Content (TPC). TPC activity is a method to determine the amount of phenolic content in the samples in antioxidant behaviour (Razali et al., 2019). Phenolic compounds found in plants where it provides redox effects and can serve as antioxidants because of their properties. In a variety of ways, phenolics are able to serve as antioxidants. Phenolic hydroxyl groups are labelled as great hydrogen donors. In a termination reaction, hydrogen-donating antioxidants can react with reactive oxygen and reactive nitrogen species, which breaks the cycle of new radicals being produced. A radical form of the antioxidant is generated following to the chemical reaction with the initial reactive species, providing a much greater chemical stability than the initial radical. The interaction between hydroxyl groups of phenolics and benzene ring's electrons provides special properties to the molecules, most notably the ability to produce free radicals where delocalization stabilises the radical. Radical-mediated oxidation processes can be modified by the formation of these relatively long-lived radicals (Pereira et al., 2009). Thus, the formation of antioxidant in plants are formed. The existence of antioxidants such as phenolic has been studied on

herbal plants, vegetables and fruits where it has free radical scavenging capabilities to combat serious illness (Saeed et al., 2012). Antioxidant activity depends on the type of solvent used in the extraction process and on the pumpkin part as well as on the drying and blanching method used to the plant parts. Antioxidants emerge as easily as extractable compounds, soluble, and as the residue of the extract. This is why it is hard to identify and categorize the contribution of different compounds and the effect of different factors to the total antioxidant capacity in the pumpkin. The pumpkin can be considered as an important source of natural antioxidants which can be used in alternative medicine or food formulations. Pumpkin also has strong antioxidative properties; it will be significant for human health care. It also helps to protect the human body from harmful effect of free radicals, and it will indirectly reduce the risk of diseases. Several researchers have analysed the antioxidant potential of plant materials from few affected factors such as different temperature of oven drying (Adiletta et al., 2018), drying processing (Naqvi et al., 2020); (Zhang et al., 2009); (Mehta et al., 2017), solvent extraction (Kulczyński et al., 2020) and also pre-treatment (Choo et al., 2014a).

Drying is a type of conventional method to conserve food for a long period. To maintain the phenolic content is a particular issue during drying process because phenolic complexes play vital role in plant cells. Different temperature of oven drying (40°C and 80°C) have different results on TPC compared to untreated fresh pumpkin (0°C) reported by (Adiletta et al., 2018). Temperature of 0°C is considered in the state of fresh pumpkin as there is no treatment applied on the plant. There is a study done by (Djendoubi Mrad et al., 2012) reported that longer drying times resulted in reduction of TPC. Moreover, the continuous prolonged process may affects to low moisture content and resulted to degradation of phenolics to the plant. According to (Mbondo et al., 2018), the retention rate of total phenolics increases as temperature increases. This observation was conducted to extreme temperature for example 80°C where immediate inactivation of polyphenol oxidase enzymes occur. Higher drying temperatures lead to inactivation or inhabitation of polyphenol oxidase (PPO)-mediated oxidation of phenolics. Hence, the identification of suitable temperature need to be done to obtain the highest TPC as possible.

The application of different drying methods has its own challenges to acquire antioxidant at the highest value which includes time-consuming results, daytime temperature that cannot be controlled and low space utilization. Generally, drying process refers to the removal of moisture or water from the product in order to increase the shelf life. Some drying method could enhance the bioactive compound. However, unsuitable drying method may reduce the