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Extraction of *Pandanus amaryllifiolus Roxb*.: A review of methods and the influencing factors

Qomarudin

Department of Agrotechnology, Faculty of Agriculture, Universitas Wisnuwardhana, Malang, Indonesia

| KEYWORDS | ABSTRACT |
|-----------------------------------|---|
| Pandanus amaryllifiolus Roxb | <i>Pandanus amaryllifiolus Roxb</i> is a tropical plant. Pandan leaves offer potential, especially for their function as a source of natural colors, aroma, and antioxidants. 2-Acetyls-1-pyrroline is the primary fragrance compound in pandan leaves. This scent is essential since it acts as the primary contribution of favorable substances which could be good for physical condition and provide flavor for food products. Its sweetness and flavorful taste are known as a natural origin of flavoring. Some previous studies have carried out standard extraction techniques such as maceration, percolation, reflux, and soxhlet in the extraction process of Pandan leaves. However, the process and results come out differently than the current techniques. The modern technique results in an effective extraction moment, and the solvent used is less than in conventional methods. Hence, this paper discussed several highly good methods, such as ultrasound-assisted extraction (UAE), microwave-assisted extraction (MAE), and supercritical fluid extraction (SFE). The extraction procedure is governed by some aspects that determine the extraction results. The proper extraction method selection also affects the extraction effectiveness. Therefore, enhancing the profitable value of pandan plants can lead to discovering a better-quality extraction technique. |
| Extraction | |
| Ultrasound-assisted extraction | |
| Microwave-assisted extraction | |
| Supercritical fluid extraction | |

Introduction

Pandanus (*Pandanus amaryllifolius Roxb.*) is a tropical flora of the Pandanaceae family in the screw-pine genus. Pandan leaves are frequently called pine threads because they resemble pineapple plants with a long, narrow spiral arrangement and green leaf strings (Wongpornchai, 2006). Although the Pandanaceae family consists of about 600 varieties, there are only two species, *Pandanus amaryllifolius Roxb.* and *Pandanus odoratissimus Linn*, which have aromatic leaves and flowers (Wakte et al., 2007).

The potential of pandan leaves is enormous, especially for their function as a source of natural coloring, unique aroma, and natural antioxidants. Although it is a great source of natural dyes, the green color derived from chlorophyll in pandan leaves has yet to be widely utilized. In addition, as a source of distinctive pandan aromas and natural antioxidants, there have not been studies that explore further about it, nor have many fundamental studies been conducted. Pandan leaves extract comprises several volatile compounds in the group of alcohols, aromatics, carboxylic acids, ketones, aldehydes, esters, hydrocarbons, furans, furanone, and terpenoids. This is possibly due to the distinctive aroma of pandan wangi, which is a derivative compound of the amino acid phenylalanine, namely 2-acetyl-1pyrroline (ACPY) (Faras et al., 2014). ACPY is the main source of the aroma in pandan leaves. In addition to ACPY, other volatile compounds were found in pandan extract, including ethyl formate, 3-4-methylpentanol, 3-hexanone, hexanol. 2hexanone, trans-2-heptanol, \beta-damascenone, 4ethylguaiacol, and 3-methyl-2- (5H)-furanone (Wongpornchai, 2006).

ACPY storage sites have been found in papillae, which are located above the abaxial epidermal cells under pandan leaves (Wakte et al., 2007; Wakte et al., 2010). Papillae are found parallel to the length of the leaf and make the surface of the epidermis lower than the surface of the upper epidermis of pandanus leaves (Wakte et al., 2007). Fresh pandan leaves contain more ACPY than dried one, although they almost have no fragrance. Young leaves have a lower concentration of ACPY than mature pandan leaves (Wakte et al., 2010) since mature leaves take shorter to reach the ACPY of young leaves. In the tissue of fresh leaves, the ACPY is dissolved when the amount of water is higher. When the water content decreases, the leaves will wither and are forced to turn into a gas phase, producing a pleasant odor which continuously released from the withered leaves. (Wongpornchai, 2006).

Pandan leaf flavor is fragrant and pleasant, widely used as a natural fragrance source in Southeast Asian countries, including India, Thailand, Malaysia, and Indonesia. For instance, pandan leaves are used to add flavor. In addition, pandan leaves are also used to scent dishes such as desserts, sweets, coconut jam, and ice cream. With a high chlorophyll content, pandan leaves are a popular source of green leaf color for food. (Loh et al., 2005).

The fragrance of pandan leaf also changes along with the processing steps taken. For example, by drying and heating, the fragrance of pandan leaves may reduce since it undergoes evaporation and several changes due to the reduction of the ACPY aroma. By drying and heating, the color may also experience a significant change from fresh green to pale green due to the effect of the heat imposed on the pandan leaves. Antioxidants from plant sources attract consumers because of their role in maintaining human health (Ghasemzadeh and Jaafar, 2013). Pandan leaves contain antioxidants that can provide good defense and fix destruction caused by free radicals. Native antioxidants and polyphenols in herbaceous plant layers are more secure than synthetic antioxidants (Jimtaisong and Krisdaphong, 2013). Naturally, sources of antioxidants can be found in some vegetables, fresh fruits, plants, and spices that generally contain phenolic compounds. Therefore, exploring natural materials as a source of antioxidants is necessary. Pandan plants are one of the natural ingredients with an enormous potential to be a source of antioxidants, but they have yet to be widely explored for their antioxidant content. Pandan wangi (Pandanus amaryllifolius) is a plant with a chemical value of alkaloids, flavonoids, saponins, tannins, and polyphenols that are antioxidant agents. Therefore, it is crucial to

identify the antioxidant content of pandan leaves, which may be further employed as a natural element to neutralize free radicals.

The main problem with pandan extraction is the longer extraction time and suitability of solvent, which can damage the desired active substance from the extraction process.Extraction is the procedure of selection of the most suitable part one or more compounds from a blend of liquid or solid substances. In the extraction process, a solvent is added to form different phases of the material; therefore, the substances to be separated can appear dissolved in the solvent. The extraction of pandan plants is necessary to obtain suitable extracts for the following research stage. ACPY is the main source of the aroma of pandan leaves. Therefore, the proper extraction may obtain pandan extract, including optimal ACPY. Determining the use of ethanol, temperature, and length of extraction setup in the extraction process is essential for optimizing desired extract acquisition. Selection of the extraction method is crucial since the extraction results will reflect the method's success rate (Garcia-Salas et al., 2010). Conventional extraction (such as maceration, percolation, reflux, and Soxhlet) generally involves a thermal process and takes a considerable amount of time, which consequently causes damage to the phenolic component. Thisreview paper investigates the best extraction methods to be used and their influencing factors.

Benefits of pandan wangi leaf

Pandan wangi is a plant frequently used by the leaves as a food additive, typically a natural coloring and flavor-providing material. ACPY storage sites have been found in papillae where epidermal cells are abaxially in pandan leaves (Wakte et al., 2007; Wakte et al., 2010). Pandan wangi (Pandanus amaryllifiolus Roxb.) is a tropical herbal extensively used as an aroma enhancer in rice and some types of bread. Pandan wangi leaves are widely used in Indonesia for specific purposes (such as a complement to cooking seasonings, a natural dye, or a natural flavor) to improve color and flavor in food products. The presence of amino acid derivative compounds from phenylalanine causes a distinctive aroma of pandan leaves called 2-acetyl-1pyrroline (ACPY) (Faras et al., 2014). ACPY is also a derivative compound of the amino acid phenylalanine, similar with jasmine and basmati rice. However, the

concentration of ACPY in pandan leave is higher (Cheetangdee and Chaiseri, 2006). Moreover, the ACPY content in pandan leaves is ten times that of aromatic rice and one hundred times that of nonaromatic rice (Yahya et al., 2011). The concentration of ACPY in pandan leave varies depending on the isolation method. Distillation using supercritical carbon dioxide as a solvent produced seven point one six ppm of ACPY in pandan wangi (Bhattacharjee et al., 2005). ACPY compounds degrade easily during heating or when cooked (Cheetangdee and Chaiseri, 2006).

Pandan wangi leaves contain essential oils, the primary source of active compounds that benefit human health. In addition to being a spice, pandan wangi leaves are also utilized as a raw ingredient in perfume production. Pandan wangi leaves release a fragrant aroma when squeezed or sliced, often used as a flavoring, aroma enhancer, and green coloring in food (Rahayu and Handayani, 2008). Moreover, pandan wangi leaves are also applied as traditional medicine in various countries, including the Philippines, Thailand, and Indonesia. According to Cheeptham and Towers (2002), pandan wangi leaves are used as a treatment in Southern Ssia to revitalize the body, decrease feverishness, and alleviate stomach ache. Pandan leaves have a potent flavor and are extensively applied as an ingredient for aroma enhancers in Southeast Asia in several food products (i.e., baked products, ice cream, and candy) (Jiang, 1999). The typical aroma of pandan wangi is suspected to be due to the presence of the amino acid derivative compound phenylalanine, namely ACPY (Faras et al., 2014). ACPY is also a derivative compound of the amino acid phenylalanine found in the jasmine plant and basmati rice. However, the concentration of ACPY in pandan wangi is higher (Cheetangdee and Chaiseri, 2006).

Another benefit of pandan wangi leaves, besides their color and aroma, is that they have a high fiber content of 3.5%, especially in their fresh leaves (Adkar and Bhaskar, 2014). Pandan wangi is also reported to have antidiabetic activity in water extract, antioxidants in water and methanol extract, anticancer in ethanol and methanol extract, and antibacterial in ethanol and ethyl acetate (Mardiyaningsih and Aini. extract 2014: Prameswari and Widjanarko, 2013; Ghasemzadeh and Jaafar, 2013; Chong et al., 2012). Those studies also highlighted that selecting suitable solvent in the extraction is an essential factor that affects the therapy's potential. As pandan leaves have been commonly applied as a coloring and flavor enhancer in food, it is very strategic if it is also developed as a food preservative. The high use of synthetic preservatives needs to be balanced with the development of relatively safer natural preservatives. This is due to the ability to suppress development of Escherichia coli and the Staphylococcus aureus bacteria, two types of bacteria for food safety indications (Faras et al., 2014). The compounds of pandan wangi leaves (i.e,. flavonoids, alkaloids, saponins, tannins, polyphenols, and color substances) may contribute to antibacterial activity (Arisandi and Andriani, 2006). Pandan wangi is a flora with a chemical content of alkaloids, flavonoids, saponins, tannins, and polyphenols that function as an antioxidant agents.

Chemical composition of pandan wangi

According to Aini and Mardiyaningsih (2016), the results of the phytochemical screening indicated that pandan wangi have substance tannins, alkaloids, flavonoids, saponins, and polyphenols. There is a difference in the formation of aroma compounds in pandanus compared to rice. The aroma produced by pandan is released during heating process. The fresh pandan wangi has green aroma characteristics. The aroma of the pandan leaves without heating does not contain 2-acetyl-1pyrolline (ACPY), but it contains 73.07% 3methyl-2 (5H)-furanone, 7.09% 3-hexanol, 6.13% 4-methyl-2-pentanol, 2.97% 3-hexanone, 2.65% 2hexanone and several components (Jiang, 1999). However, the fragrance compounds cyclohexanol, cyclohexanone, 2-acetyl-1-pyrolline, 3-methyl-2-(5H)-furanone, N-octanal, nonanal, 2-ethyl-5methyl, and 1-isocyanato-2-methoxy benzene are present in heated pandan wangi leaves (Cheetangdee and Chaiseri, 2006).

According to Ningrum et al. (2015), pandan wangi leaves (P. amaryllifolius Roxb.) contain carotenoid xanthophyll compounds. and Carotenoids, especially a-carotenoids and bcarotene, are precursors of norisoprenoids that also contribute to a solid aroma used in food. Carotenoids have long been known as antioxidant compounds to protect against various degenerative illnesses and diseasees caused by an uncontrolled division of abnormal cells in a part of the body, heart disease, or diseases associated with aging (Perera and Yen, 2007). Therefore, the extract of pandan wangi (Pandanus amaryllifiolus Roxb.) leaves is beneficial for enhancing the appearance of food and benefiting health. Some carotenoid compounds identified in pandan leaves are violaxanthin, neoxanthin, luteinepoxide, acarotene, and b-carotene (Ningrum et al., 2015).

According to Cheetangdee and Chaiseri (2006), other than compounds that produce aromas, fresh pandan wangi leaves contain carbohydrates of 2.38 mg/g fructose, glucose of 1.77 mg/g, and amino acids. Although carbohydrates level found in the pandan wangi leaves is relatively low, the body can use fructose and glucose as energy sources. The pandan wangi leaves contain very strong fibers. Generally, fibers taken from pandan leaves are used to make handicrafts such as hats, bags, and various baskets (Gurmeet and Amrita, 2015).

Besides containing carbohydrates, pandan wangi leaves also have consecutive free amino acids from the highest to the lowest concentration, namely aspartic acid, serine, glutamic acid, glycine, histidine, arginine, threonine, alanine, proline, tyrosine, valine, lysine, isoleucine, leucine, and phenylalanine (Cheetangdee and Chaiseri, 2006).

2-acetyl-1-pyrroline (ACPY)

ACPY is volatile compounds associated with aroma, easterners say pandan and westerners say pop-corn. Various kinds of cereals are found and mostly come from vegetables and animal products (Adams and De Kimpe, 2006; Tulyathan et al., 2008; Sirisoontaralak and Noomhorm, 2006). pPants naturally form ACPY (Sugunya Wongpornchai et al., 2004) and is the main factor forming the characteristics taste of pandan leaves (P. amaryllifolius Roxb.), bread flowers (Vallaris glabra Ktze), and aromatic rice varieties (Oryza sativa L.) such as Jasmine rice and Basmati rice (Buttery et al., 1988; Laohakunjit and Kerdchoechuen, 2007; Wongpornchai et al., 2004; Arikit et al., 2011).

The reaction between two compounds, namely amino acids and sugars on the food surface, produces ACPY, such as rice and bread, at high temperatures (Fuganti et al., 2007; Sugunya Wongpornchai et al., 2004). Reaction at high temperatures between sugar and amino acids is called a non-enzymatic browning reaction or Maillard reaction (Adams and De Kimpe, 2007; Yu et al., 2009). Many scientists have found to synthesize ACPY compounds by the nonenzymatic browning reaction costs, the length of the reaction order, the need for acids and bases for the final stage, and the harmful of being consumed (Fuganti et al., 2007).

Production with microbes of ACPY by *Bacillus cereus* ATCC 27522 has been reported previously (Adams and De Kimpe, 2007). They reported that ACPY making is continued through

enzymatic acetylation of 1-pyrroline, a breakdown product of the amino acid metabolism of ornithine and proline. However, specific B. cereus strains grow on count plates to produce low amounts of ACPY and mainly cannot be reproduced . Furthermore, the synthetic form of ACPY by hydrogenation of 2-acetylpyrrole with rhodium on alumina, followed by oxidation of the amino alcohol produced through an excess of silver carbonate, and adsorbed on Celite in benzene has also been reported by Buttery (1983). However, he stated that the procedure was not feasible because of the expensive and toxic reagents such as benzene and the inability to achieve ACPY on a larger scale. Adams and De Kimpe (2007) also attempted a more effective technique for the production form of ACPY. However, some obstacles are found in the production of ACPY compounds.

ACPY, under the name IUPAC 1-(3,4dihydro-2H-pyrrol-5-yl) ethanone, is a substituted pyrroline and a cyclic imine, the authentic grade ACPY is doughy yellow (Bhattacharjee et al., 2005). These heterocyclic compound usually has a low concentration of nitrogen (Jianming, 2002). Further, 2-Acetyl pyrroline has been recognized as a very volatile hydrophilic compound (at vapor pressure and 25 °C) (Fitzgerald et al., 2009; Hien et al., 2006).

ACPY has a lower odor threshold of 0.1 ppb in water (Buttery et al., 1988) and has a very high solubility in water and alcohol with a log K(o/w) -1.27 value. Therefore, ACPY can be classified as hydrophilic compounds because the value of K (K is the partition coefficient of aroma compounds between octanol and water) is below 2, almost the same as ethyl acetate, ethyl butyrate, 2 hexanone and cis-3-hexanol. These compounds are highly intense, even at small concentrations, and may cover other aroma (Fabra et al., 2009).

Almost identical to another polar compounds, ACPY provide less affinity for oil phase than water (Seuvre et al., 2007), and when subjected to high temperatures, it is easily converted into a gas phase because of its low molecular weight (111.14 gmol-1)(Sriseadka et al., 2006). This compound is thermally labile and is extremely unstable under acidic/basic conditions and at ambient temperature conditions (Laohakunjit and Noomhorm, 2004; Widjaja et al., 1996). Therefore, ACPY reduction in food product may be occurred because of its volatility (Tulyathan et al., 2008).

Pandan Leaf Extraction

Extraction is selectively splitting up one part or more compounds from a blend of liquid or solid substances. In the extraction process, a solvent is added to form different phases of the material; therefore, the substance that wants to be separated can appear dissolved in the solvent (Toledo, 2007). The type of target compound and its chemical content in the material are determined by the solvent selected. In addition, polarity and conditions of use, such as temperature when using solvents, are the most significant factors affecting extraction efficiency and selectivity (Chemat and Vian, 2014).

Before selecting a method, the extraction target needs to be determined first. extraction targets include unidentified bioactive compounds, compounds already present in the organism, and a group of structurally related compounds within an organism (Sarker et al., 2006). The nature of the materials and compounds to be isolated is a major factor in the selection of extraction. Here are some commonly extraction methods used on pandan leaves:

a. Maceration

Classical/conventional extraction techniques useg a small scale to extract bioactive components from several plant materials (Srivastava et al., 2021). These techniques are usually based on different solvent extraction efficiencies used for this purpose. Maceration is the simplest and most widely used method for both small-scale and industrial applications. Due to the reason that the sample and solvent are placed in an enclosed, inert container at room temperature, this approach is seen to be straightforward. The extraction process is stopped if equilibrium between the concentration of the compound in the solvent and the concentration in the plant cell has been achieved. The use of room temperature in this method can avoid damaging thermolabile compounds. (Selvamuthukumara et al., 2017). The maceration procedure has several disadvantages, even though it is considered the most popular method. The main disadvantage of this method is that it takes longer time to extract compounds, requires quite a lot of solvents, and most likely some compounds are difficult to extract at room temperature (Mukhriani, 2014).

The maceration step is used at the homemade level to prepare the resultant liquid a long way before. For small-scale extraction, the maceration process generally consists of several stages. The material is firstly milled or reduced the particle size to enhance the surface area for homogeneous mixing with the chosen solvent. In the second step, the maceration process, select the appropriate solvent and add it to a closed vessel (De Vries et al., 2018). Thirdly, the liquid is filtered, which is the solid residue of this extraction process is pressed to obtain a large amount of clogged solution. The liquid obtained is mixed and separated from the impurities by filtration. Occasional shaking in the maceration process will facilitate extraction in two ways: first, it will increase diffusion, and second, it will remove the concentrated solution from the sample surface to bring a new solvent to the menstruum to achieve more extraction results (Rasul, 2018).

Hydro distilation (HD) is one of the traditional methods used to extract bioactive compounds and essential oils from several plant materials. In this process, organic solvents are not involved and can be removed prior to the dehydration of any plant material. There are three types of HD: water distillation, water and steam distillation, and direct steam distillation (Vankar, 2004). In HD, the plant material is first packed in a stationary compartment; second, water is added in a sufficient amount and then boiled. Alternatively, steam can also be injected directly into the plant sample. Both hot water and steam can act as the main influencing factors to release bioactive compounds from some plant tissues. Indirect cooling by water condenses a mixture of water vapor and oil. The viscous mixture flows from the condenser to the separator, where oil and bioactive compounds are automatically separated from water (Silva et al., 2005). HD involves three main physicochemical processes: 1. hydro diffusion, 2. hydrolysis, and 3. decomposition by heat. At higher extraction temperatures, some of the volatile compounds in the component will disappear. Therefore, this drawback limits its use for extracting various thermolabile compounds from different plant tissues (Selvamuthukumara et al., 2017).

b. Reflux and steam distillation

In the reflux process, the material and the solvent are introduced into a flask connected to a condenser. In this process, the solvent will be boiled to 100 °C. Then the steam condenses and goes back into the flask. Steam distillation has the same process and is usually used for extracting essential oils. During heating, condensed steam and distillate (separated into two parts that do not mix) are housed in a container connected to the condenser. The disadvantage of these two methods is the possibility of degradation in thermolabile compounds (Mukhriani, 2014).

c. Percolation

In the percolation method, the sample is slowly wetted with solvent in a percolator (a circular vessel equipped with a faucet under the tool). The solvent is appended to the sample and permitted to drip slowly on the bottom, which takes up longer time. In this method, the sample is constantly flowed by a new solvent; thus, the solvent needs are quite large. In addition, if the sample in the percolator is not homogeneous, then the solvent will be challenging to reach all parts of the sample, resulting in the extraction has less purity (Mukhriani, 2014)

d. Soxhlet

In the Soxhlet procedure, the material is placed in cellulose sheath in a funnel placed above the flask and below the condenser. Suitable solvents are put into the flask, and the water bath temperature is set below the reflux temperature. Using temperatures at the boiling point may lead to the lapse of thermolabile compounds because of degradation. However, the continuous extraction process where the sample is extracted by pure solvent resulting from condensation causes the use of solvents in this method to be relatively low and take less time (Mukhriani, 2014). Soxhlet extraction techniques have been widely used to remove some substances from several plant ingredients (Manousi et al., 2019). The dry plant material of the sample needs to be stored in a thimble. Then the thimble is placed in the distillation tube when the selective solvent level is reached; siphon sucking thimble holder solution (De Silva et al., 2017). The solution is removed from the siphon into the distillation flask. The solution carries the extracted solute into the bulk liquid. The solute remains in the distillation flask and the solvent returns to the plant solids' place. The procedure repeatedly runs until extraction is complete.

e. Microwave-assisted extraction (MAE)

This MAE method uses microwave assistance and is expected to speed up the extraction process. Using the MAE method can speed up extraction time and effective use of solvent than the conventional method, thus increasing the yield of crude extract (Nawrot et al., 2011). This extraction process that uses microwaves utilizes microwave energy using the frequency of 0.3-300 GHz in the configuration of electromagnetic non-ionizing radiation. Molecular movement in the form of ion migration and dipole rotation is caused by microwave energy. The friction due to the fast movement eventually causes heat energy in the material, so the cell wall or material tissue is disrupted, and the solutes come out (Delazar et al., 2012). The findings of several earlier studies suggest that MAE can improve the successfulness and productiveness of extracting the

favorable components from several kinds of spices, herbaceous plants, and fruits. This is due to microwave radiation which helps to accelerate the most suitable taking out sample heating quickly and maximum productivity (Calinescu et al., 2001; Jain et al., 2009). The apparatus used for the MAE method is shown in Figure 1.

The principle of MAE is that microwave radiation gives heat to the water in the cell to the extent that the water in the cell will evaporate and exert high force on the rigid layer, which results in the expansion of the cell. The force will push the rigid layer from the internal, stretch, and break the smallest structural and functional unit of an organism (Mandal et al., 2007). Active compounds release can be facilitated by destroying the material matrix from within the cell material to the surrounding solvent (Jain et al., 2009). According to Kurniasari (2008), microwave heating occurs through direct interaction between material and microwaves. These direct interactions cause molecules to move, which produces heat, as well as produce friction between molecules. This friction causes the cell walls and tissues of the material to be damaged and make the solute escape; therefore, the longer the friction occurs, the more energy the material absorbs, and more solutes will escape. However, according to Chan et al. (2011), avoid long-term exposure it is necessary to prevent damage to the extraction compoundto. The closed and open type microwave systems are shown in Figure 2.

Extruding thermolabile compounds is very suitable for MAE because it controls temperature better than conventional heating processes. The MAE approaches are widely applicable for extracting different compounds, as well as hotnesssensitive compounds. Furthermore, taller extraction value, little using solvent, and no timeconsuming contrast to regular extraction are other advantages that MAE has (Santos-Buelga et al., 2012). Several materials can be extracted simultaneously using MAE in a shorter time than the Soxhlet method, and the yield results resemble the supercritical fluid extraction result. When applying this method, consideration must be taken when using flammable solvents or extracts that include thermolabile chemicals in solvents with high dissipation factors (Salas et al., 2010).



Figure 1. Microwaveassisted extraction system (Anton-Paar, 2014).



Figure 2. (a) Closed type microwave system and; (b) Open type microwave system (Modified from Mandal et al., 2007).

In MAE, the acceleration of chemical reactions through heating with microwaves results from the interaction between waves and materials. Waves with a frequency of 2.5 GHz can be taken up by water, sugar, and fat but not by materials such as glasses, ceramics, and some plastics. The metallic material even reflects these waves; thus, the material only absorbs the microwaves by. MAE can increase extraction yield due to the raw material's capillary absorption properties and higher water absorption capacity (Quoc, 2014).

The MAE can be influenced by several factors, including microwave ability, length of extraction, extraction heat, type of solvent, and comparison of materials with solvents (Kurniasari, 2008). The solubility of the target compound to be extracted depends on the solvent selectionused in the extraction. This is where the interaction occurs between solvent and matrix material, as well as the energy absorption properties of solvent microbubbles (Mandal et al., 2007). The solvents used are suggested to have high selectivity towards the target compound and must absorb microwaves; typically, polar compounds can absorb the energy from microwaves efficiently (Santos-Buelga et al., 2012).

Besides the type of solvent used, the volume of the solvent is also a critical factor in this process. The main principle in determining the solvent volume is that it must be capable of immersing the substance during extraction. However, the extraction using microwaves with more solvent volumes can result in a lower yield. This is because the solvent absorbs more microwave energy before it reaches the material matrix (Mandal et al., 2007). Therefore, the comparison between ingredients and solvents must be chosen appropriately to maximize the extraction yield. The previous study reported that the ratio of ingredients and solvents used in the MAE method ranged between milligrams and milliliters (on a laboratory scale) with a minimum application ratio of 1:10 (mg/mL) to 1:20 (mg/mL) (Xiao et al., 2012).

The temperature determination in the MAE method must be adjusted to the solvent's boiling point (Jun et al., 2003). Excessively large power consumption can cause degradation in the structure and quality of the extracts produced. High power and high temperature will quickly tear down cell walls, allowing undesired chemicals to be transported into the solvent along with the target product. Whereas at low power, the cell wall breaks down gradually, allowing the solvent to be selectively reactive with the desired molecule. Power must be selected appropriately to prevent over-heat, which can reduce the target compound and overpressure in this procedure (Mandal et al., 2007). Based on the research of Bhadoriya et al. (2011), the power optimal for the process of the MAE method ranges from 80-420 Watts. In the MAE method, the usage of microwave power and irradiation duration are also mutually influencing aspects. The optimal strategy for this method is the merger's low power and extended exposure time. High power with a long exposure time will raise the temperature causing thermal degradation of the target compound. In contrast, cell wall fragmentation will occur gradually, allowing the solvent to be more selective to the target compound (Mandal et al., 2007).

The extraction hour affects the amount of heat produced, which longer the extraction time, the stronger the cell wall degradation process will also be. In addition, the longer extraction time can also cause evaporation of the solvent, reducing the volume. Generally, the amount of target compound and yield increases with increasing extraction time. However, there is also a chance of reduction of the object compound itself. The length of extraction hour of 15-20 minutes is often considered sufficient for extraction using MAE (Mandal et al., 2007).

f. Ultrasound-assisted extraction (UAE)

Ultrasound is a type of sound wave that is beyond the range of human hearing and has a frequency between 20 kHz to 100 MHz.. Like other waves, this wave can pass through matter with the power of compression and expansion. In addition, this procedure results in a phenomenon called cavitation, which results in the bubbles' production, growth, and collapse. This energy conversion produces a large amount of energy which then spurs the heating of the bubble contents (Herrera and Luque-de-Castro, 2004). According to Suslick and Doktycz (1990), bubbles have a heat of about 5000 K, a tension of 1000 atmospheres, a temperature rate, and a cooling rate above 1010 K/s. Deriving from this concept, the UAE has been expanded. Substances with a cavitation effect are only liquids and liquids containing solid materials.

Cavitation is closely related to the extraction intensification process using ultrasound. These ultrasonic waves cause compression and expansion cycles during passage through the liquid. Bubbles and cavities in the liquid are made by an expansion. When negative pressure is applied, the local tensile strength of the liquid is greater and varies greatly depending on its nature and purity. Cavitation also occurs if the formed vapor bubbles grow and experience a drastic decrease. The enormous shear energy waves and turbulence in the cavitation zone result from the bubble bursting at a tremendous temperature of 4500 °C and pressures of up to 100 MPa. Various factors should be considered to speed up mass transfer in the extraction process, including pressure, heat, and turbulence (Patist and Bates, 2008). The extraction process using ultrasound includes two steps: 1. Spread covering the cell wall and 2. Clean the cell's content after destroying the wall (Mason et al., 1996).

Essential factors in obtaining efficient and effective extraction are the moisture content of the sample, degree of grinding, particle size, and type of solvent. In addition, the factors that govern the action of ultrasound are affected by temperature, pressure, frequency, and sonication. Various classical techniques have been combined with UAE techniques to increase the efficiency of conventional systems. To increase extraction efficiency, place the ultrasound device in the correct position in the extraction unit (Vinatoru et al., 1998). The advantages of the UAE include efficiency of extraction time, energy absorption, and solvent utilization. More effective blending, faster energy transfer, reduced thermal gradient, extraction temperature, selective extraction. reduced equipment size, faster response to process extraction control, fast start-up, increased production, and reduced process steps are also facilitated by ultrasonic energy (Chemat et al., 2008).

g. Supercritical fluid extraction (SFE)

Many scientists are interested in this SFE technique, which has been successfully applied to pharmaceutical, polymer, and food fields (Zougagh et al., 2004). This technique has been used by several industries over the years, especially in the preparation of the decaffeinated coffee industry

(Ndiomu and Simpson, 1988). The supercritical state is when a temperature and pressure beyond the critical point of a given substance are reached. The absence of typical characteristics of temperature (Tc) and pressure (Pc) in the gas and liquid phases is defined as the critical point. (Inczedy et al., 1998). In the supercritical state, a change in the specific properties of the gas and/or liquid occurs, meaning that changes in temperature and pressure cannot liquefy the supercritical fluid. The characteristics of gas diffusion, viscosity, surface tension, and density, such as liquids and solvation power, have been possessed by supercritical fluids. These characteristics can increase the yield obtained by rapid extraction of the compound (Sihvonen et al., 1999).

The fundamental SFE technique consists of parts: a moving stage tank, generally CO₂, a pump to pressurize the gas, a co-solvent receptacle and pump, an oven containing an extraction receptacle, a controller to keep High Pressure inside the system, and a trapping receptacle. Commonly, different kinds of gauges, stream gauges, and dry/wet gas gauges can be set into the system. CO_2 is deemed a perfect solvent for SFE. the possibility of operation at moderate pressures, generally between 100 and 450 bar, has been offered at room temperature and low critical pressure (74 bar) at a critical CO_2 temperature condition (31 °C) (Temelli and Guclu-Ustundag, 2005). Its low polarity is a disadvantage of CO₂, so it is only suitable for lipids, fats, and non-polar substances and is not suitable for polar Using chemical modifiers substances. has successfully overcome the low polarity limitations of CO₂ (Lang and Wai, 2001; Ghafoor et al., 2010). Commonly, a small amount of change is required to increase the polarity of CO_2 significantly. For instance, 0.5 mL of dichloromethane (CH₂Cl₂) can increase extraction, equal to 4 hours of HD (Hawthorne et al., 1994). A symmetric diagram of the SFE apparatus is shown in Figure 3.

On several parameters, SFE becomes the main factor in the extraction of bioactive compounds from plant materials, and most importantly, these parameters can be controlled (Raverchon and Marco, 2006; Raynie, 2006, 2010). Temperature, pressure, particle size, moisture content of input material, extraction time, CO_2 flow rate, and solvent-to-input ratio are the main factors affecting extraction efficiency (Temelli and Guclu-Ustundag, 2005; Ibanez et al., 2012). The benefit of using supercritical fluids in extracting bioactive compounds are as follows: (Lang and Wai, 2001):

1. SFE has higher spread coefficients and reduced viscosities and surface tension than liquid

solvents, producing more infiltration into the substance matrix and favorable mob displacement. The extraction hour could be substantially decreased with SFE compared to the conventional methods.

- 2. Reflux is repeated on the supercritical fluid sample to complete the extraction.
- 3. Liquid solvents have lower supercritical fluid selectivity because various temperatures or pressures can regulate their solvation power.
- 4. Supercritical fluid depressurization can facilitate the separation of the solute from the solvent in the extraction process and reduce the time.
- 5. SFE is an ideal method for extracting thermolabile compounds because it can be operated at room temperature.
- 6. In SFE, when compared to solvent extraction methods, little quantity of sample can be extracted
- 7. SFE does not utilize organic solvents, so it is environmentally friendly.
- 8. Supercritical fluids may be recycled and reused, which helps to reduce the amount of waste produced.
- 9. SFE can be scaled up for industrial purposes, from very small sample sizes for laboratory scale to commercial scale in the industry.
- 10. The SFE provides detailed information on the extraction procedure to optimize the extraction process. Modification supercritical of carbon dioxide (SC-CO₂) with ethanol (15% wt) can show better naringin (flavonoid) extraction results from *Citrus paradisi* than pure SC-CO₂ at 96 MPa and 58.6 °C (Giannuzzo et al., 2003). Extraction of polyphenols and procyanidins from grape seeds using SFE, where methanol was used as a modifier, and CO₂-modified methanol (40%) yielded more than 79% catechins and epicatechins from grape seeds (Khorassani and Taylor, 2004). Pascual-Marti et al. (2001) assessed and optimized SFE conditions [pressure (80-110 bar), temperature (40 °C), ethanol concentration (5-15%), and extraction time (5-25 minutes)] to get well resveratrol from Vitis vinifera grape skins. The researchers suggested that optimal SFE extraction conditions were achieved at 110 bar, 40 °C, 7.5% ethanol, and an extraction time of 15 minutes. Resveratrol content is fully recovered (100%) under these conditions. In another research, the effect of SC-CO₂ extraction (100-400 bar/35-55 °C) and the addition of a modifier [5% (v/v) ethanol] on the recovery of resveratrol from grape seeds, stems,

skins, and pomace of the Palomino Fino grape variety was studied (Casas et al., 2010). It was found that the upper limit recuperation of resveratrol was gotten from the skin (49.1 mg/100 g dry sample) when SC-CO₂ was used at 400 bar/35 °C and 5% (v/v) ethanol as co-solvent.

Loui et al. (2004) studied the impact of an integrated liquid and supercritical solvent extraction process to get better antioxidant compounds from winery by-products. They found that the extract's antioxidant activity was influenced by the solvent type, the medium composition (i.e., skin, seed, and stems), and the pre-destruction treatment. This study explained that ethyl acetate as a solvent showed the highest antioxidant activity in the extract.

Super critical-carbon dioxide (SC-CO₂) fluid technology

The process of extracting in $SC-CO_2$ liquid is influenced by four main factors: extraction, expansion, separation, and solvent conditioning. The steps have four main generic components: an extractor (High-Pressure strong container), a pressure and temperature control system, a separator, and a pressure intensifier. Raw materials are generally ground and introduced into a temperature-controlled extractor to form a fixed room, which generally happens in batch and onestage modes. (Shi et al., 2007a, 2007c; Kassama et al., 2008). The process represented above is a semibatch continuous process in which SC-CO₂ streams in continuous modes while the extractable solid input is supercharged into the extraction container in batch.

Several extraction vessels are used in series in commercial-scale processing plants to improve operation and yield (Del Valle, 2015; Temelli, 2009). During extraction, there is an increase in efficiency when the process is shifted to another vessel prepared for extraction, unloading and/or loading of the previous vessel even though the system is stopped at the end of the extraction period. Multistage extraction is used on a commercial scale with a semi-continuous approach process involving the system using many extraction vessels simultaneously. In this method, since the process is switched to the next vessel by the extraction control valve, the process is not interrupted at the end of the extraction period. So, SC-CO₂ technology is obtained on a one-stage clutch form that can be upgraded to a multistage semi-continuous batch operation joined with a multi-separation process. The need for design upgrades is required, enabling the SC-CO₂ technology to produce higher yields.



Figure 3. A symmetric diagram of the SFE apparatus (Modification from Yi et al., 2009).

The extraction process is influenced by several factors that determine the results of an extraction. These factors are as follows:

a. Selection of solvent type

The type of solvent influences the type of bioactive compound to be extracted. This is because each type of solvent has a different efficiency and selectivity in dissolving bioactive components in the material. The concentration of different types of solvents will also affect the bioactive compounds extracted. Increasing solvent concentration may reduce the time required for extraction. Solvents should be non-hazardous to workers, non-toxic, non-flammable, and noncorrosive to extraction equipment. Solvents that widely used in the extraction process are acetone, ethanol, methanol, ethyl acetate, chloroform, hexane, and ethylene dichloride (Amiarsi et al., 2006). Selective solvents greatly affect the extraction rate of conventional methods (Cowan, 1999). Table 1 represents some examples of bioactive compounds extracted with different solvents. The compound targeted in extraction is strongly influenced by the polarity of the solvent. When choosing a solvent, the molecular affinity n, environmental safety, toxicity, profit, and loss must be considered for its antioxidant extraction efficiency. For example, the antioxidant in pandan can be taken by solvent extraction method using alcohol solvent. Alcohol is used as a solvent because it is relatively cheap, easy to obtain, and relatively safer to use for foodstuffs than other organic solvents. Pandan leaves extract obtained can be applied as a native antioxidant. Therefore, non-natural antioxidants can be reduced or eliminated and replaced by natural antioxidants. (Margaretta et al., 2013).

b. Preliminary treatment of materials

Preparing raw materials includes drying and reducing the material's size to the preferred size for extraction. The material's particle size greatly influences the success of the extraction process is much influenced by of the sample. The mass transfer rate is higher due to the short diffusion path. Reducing the particle size is parallel to increasing the surface area, thus improving the contact between the solid and the solvent (Nugroho et al., 2008). In addition, the time required for the components to be released from the material becomes shorter, thus fastening the extraction process. After shrinkage, the material's particles should be homogeneous in size to assist in the solvent's diffusion into the material. Substances with a fine particle size can also clump, making

them difficult to be penetrated by solvents (Nagarwal et al., 2009). Therefore, a powder with a size of 0.5 nm is a suitable for the extraction process. Drying to specific moisture content is an alternative preliminary treatment to reduce the water content of the material prior to the extraction. High water content can cause the extract to contain water-soluble components such as starch and sugar. Generally, plants are dried at room temperature at less than 30 °C and avoid direct sunlight. Ultraviolet radiation due to drying with direct sunlight can cause alterations in the configuration of the constituent compounds (Serratosa et al., 2008)

The number of components extracted from the material and the contact length between the material and solvent are determined by the length extraction time (Durling of the et al.. 2007; Kusuma et al., 2018). The greater possibility of contact between the material and the solvent, the longer the extraction time, increasing the solubility of the bioactive components in the solution. The stirring process is needed to accelerate the dissolution of solids and increase the diffusion rate of solutes in the extraction process. The movement of solvent around the material due to stirring can accelerate the contact of the material with the solvent. Thus, it moves the component from the material's surface into the solution by forming a suspension and dissolving the component into the solvent medium. Stirring can be carried out by mechanical means, air spraying, or combining the two.

c. Comparison of amounts of solvents and additives

The greater the volume of solvent used compared to the amount of material extracted, the greater the yield produced. The solvent's ability to dissolve the material depends on the amount of solvent added to maximally extract the components. The yield of extraction may continue to increase until the solution becomes saturated. After the saturation point of the solution is reached, solvent addition will not increase yield (Amiarsi et al., 2006).

Conclusion

Many researches have studied the extraction of bioactive compounds from pandanus plants with more convenient methods. Changing ordinary technology with modern one to extract precious compounds represents many advantages, including reduced energy consumed, non-toxic organic solvents, and increased extraction yields, in accordance with the concept of eco-friendly. In addition, many modern technologies are capable of selectively producing intracellular molecular extracts that do not destroy the preserved tissue, which is highly sought after to minimize further refinement steps. The integration and expansion of crossbred methods should also be explored, in view of the characteristics of plant material, and the option of compounds. There will be more smart extraction methods in the time to come for *Pandanus amiriliofolius Robx*, such as modern extraction method with the efficiency solvent in use and no time consume which has increased economic significance and is rich in bioactive compounds.

Declarations

Conflict of interests The authors declare no competing interests.

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