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Review article

Use of fruit-derived essential oils with antioxidant properties in animal cell cultures: A review

Uso de óleos essenciais derivados de frutos com propriedades antioxidantes em cultivos de células animais: Uma revisão

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PALAVRAS-CHAVE
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ABSTRACT

The increase in the use of cell culture systems has resulted in research aimed at its optimization. The reduction of oxidative stress generated in vitro can improve culture conditions, and antioxidant natural substances have been identified. Different parts of a plant, including fruits, can be processed to obtain essential oils, which can be used as antioxidants. As the raw substrates for processing can be obtained from food industrial wastes, interest in evaluating fruit-derived substances has grown, and its possible application in cell, gamete, and embryo culture is under investigation. This review aims to present the advantages of fruits as a source of essential oils with antioxidant properties, highlight the milestones achieved, and suggest the future study prospects. In general, fruit derived essential oils have numerous beneficial properties for cells, such as anti-inflammatory, and antioxidant. The actions of these substances have been interesting strategies in in vitro cell culture systems. Additionally, studies on male gametes, such substances have been used as a contraceptive activity, while studies on female gametes and embryos are still scarce.

RESUMO

O aumento do uso de sistemas de cultivo de células tem resultado em pesquisas visando a sua otimização. A redução do estresse oxidativo gerado in vitro pode melhorar as condições de cultivo, e substâncias naturais antioxidantes têm sido identificadas. Diferentes partes de uma planta, incluindo frutos, podem ser processadas para obter óleos essenciais, os quais podem ser usados como antioxidantes. Como os substratos brutos para processamento podem ser obtidos a partir de resíduos industriais de alimentos, o interesse em avaliar substâncias derivadas de frutos tem crescido e sua possível aplicação em cultivo de células, gametas e embriões está sob investigação. Esta revisão teve como objetivo apresentar as vantagens dos frutos como fonte de óleos essenciais com propriedades antioxidantes, destacando os marcos alcançados e sugerindo as perspectivas de estudos futuros. Em geral, os óleos essenciais derivados de frutos possuem inúmeras propriedades benéficas para as células, como anti-inflamatórias e antioxidantes. As ações dessas substâncias têm sido estratégias interessantes em sistemas de cultivo celular in vitro. Adicionalmente, estudos em gametas masculinos, tais substâncias têm sido empregadas como atividade contraceptiva, enquanto pesquisas sobre gametas femininos e embriões ainda são escassas.

INTRODUCTION

Natural substances, such as essential oils from roots, trunks, sticks, leaves, flower buds, and fruits, have been widely used in therapeutic practices (BAPTISTA-SILVA et al., 2020). Bioactive molecules or phytochemicals synthesized from secondary metabolites, are often used to treat various diseases (GARCIA-SALAS et al., 2010) and assisted in the development of biotechnologies (SANTOS et al., 2018). They exhibit antioxidant potential, and they can reduce or control the level of reactive oxygen species (ROS, OZOUGWU, 2016).

ROS are free radicals synthesized by cells during respiration process; at high levels, they can modify oxidative homeostasis and cause damage to the morphology, physiology, and integrity of cells (TURRENS, 2003). In vitro studies using somatic cells (BORGES et al., 2020) and gametes (SANTOS et al., 2019a, 2019b) have shown that excess ROS are harmful and affect cell quality. Therefore, use of essential oils with antioxidant potential has been proposed as a supplement to improve the conditions of cell culture (SWALLAH et al., 2020).

Since the first extraction of natural substances with biotechnological potential in the 21st century by Paracelson, the use of essential oils from different plant parts has been proposed because of their numerous benefits (MACIEL et al., 2007; ANGIOLELLA, SACCHETTI, EFFERTH, 2018). It is known that phytochemicals with different structural properties, functions, and effects are found in fruits (GARCIA-SALAS et al., 2010). They are rich in vitamins, such as ascorbic acid, α-tocopherol, and β-carotene and contain abundant phenolic compounds, which help fruits at different stages of maturation (HE et al., 2019). All these chemical constituents have an excellent antioxidant potential (TURRENS, 2003) in addition to antibacterial, antifungal, and insecticidal activities (SAIKIA et al., 2013; ROSA et al., 2019).

Fruits are important for the production of frozen juice. During processing, approximately 50% of the raw material is discarded, including peels and seeds, which can be used to obtain essential oils (MANURANJAN et al., 2019). Additionally, the by-products generated from fruits can be used diversely in the food industry, such as in the production of sweets in syrup, jellies, and functional foods, such as flour (MATIAS, OLIVEIRA, MAGALHÃES, 2005). Alternatively, these by-products can be used as animal feed or fertilizers (SCHIEBER et al., 2001). Therefore, considering the biotechnological importance and application of essential oils derived from fruits, this review aims to present the advantages of fruits as a source of antioxidant agents in systems of in vitro culture, emphasizing the advances and the prospects.

DEVELOPMENT

This review is a literary study of expository and descriptive character, emphasizing the importance of using essential oils derived from fruits with antioxidant potential and their different applications in the culture of animal cells. For this, searches were done for scientific papers in national and international journals through research in the bases of academic websites, such as Scielo, Pubmed and Google Scholar, in the period of publications from 2000 to 2022. Therefore, scientific terms, such as techniques for obtaining essential oils, essential oils derived from fruits, oxidative stress during cell culture and antioxidant potential of essential oils from fruits, were some of the themes used for writing.

Methods of obtaining essential oils from fruits

Obtaining essential oils from fruits is a multi-step process. The preliminary step involves the development of appropriate methodologies to identify the bioactive compounds with antioxidant capacity. The concentration of such compounds depends on the technique used for the extraction. The methodologies used to obtain essential oils from fruits are similar to those used for other plant parts (SILVEIRA et al., 2012; MUTALIB, 2015) including hydrodistillation and steam distillation. These methods are considered the oldest and most versatile (KOSAR et al., 2007), as they can be used to obtain essential oils from different plant parts (e.g., seeds, pulps, and peels).

Hydrodistillation involves the distillation of plant material in sufficient water (SHAKIR, SALIH, 2015) and is widely used on a laboratory scale to obtain good yield of essential oils (KOSAR et al., 2007). KOSAR et al. (2007) used hydrodistillation technique for the recovery of bioactive compounds from whole and ground fruits of fennel (Foeniculum vulgare), which gave a chemical sample yield of 98% and 99% for the whole and ground fruits, respectively. The duration of plant immersion in water varies and it allows the progression of hydrolytic reactions with bioactive molecules resulting in the formation of alcohols, esters, and carboxylic acids (BAPTISTA-SILVA et al., 2020). This changes the essential oil composition significantly, thereby reducing the quality and biochemical value of the obtained oil, especially when the aim is to obtain biological molecules with antioxidant potential.

Additionally, steam distillation can be used to obtain essential oils. In this technique, extraction is performed by saturating the biological sample with steam (PÉRINO-ISSARTIER et al., 2013). Although this extraction method is widely used on an industrial scale because of its low execution costs (KOSAR et al., 2007), limitations similar to those in hydrodistillation are observed, which leads to changes in the quality of the biological sample.

An interesting example of the difficulty faced in steam distillation was observed by SHAKIR; SALIH (2015), who compared this method with microwave distillation. The authors showed that the yield and chemical composition of essential oils from citrus fruits (Citrus sinensis, Citrus limon, and Citrus reticulata) obtained by steam distillation were lower than those by microwave distillation, which resulted in a higher percentage of the main chemical component, D-limonene, in these essential oils. The authors proposed that the time required for the exocarp cells to interact with saturated water in steam distillation is a drawback, unlike the microwave technique. The latter utilizes electromagnetic waves, thereby allowing greater interaction between chemical components and water. Additionally, the temperature increase in microwave distillation is more when compared to steam distillation (SHAKIR, SALIH, 2015). Owing to the difficulties

associated with conventional techniques, modern techniques have been devised to obtain essential oils derived from fruits.

The modern techniques used to obtain essential oils from fruits are more efficient and provide adequate isolation of bioactive components (JAHROMI, 2019). The supercritical fluid technique is more popular than other modern techniques associated with ultrasound, microwaves, and pressurized liquid (SANTANA et al., 2009; GARCIA-SALAS et al., 2010; JAHROMI, 2019). This technique is used to develop novel pharmaceuticals (PEREIRA et al., 2016) as it results in greater purity of the natural substance. This is due to the lack of organic solvent requirement unlike in other modern techniques, and a low operating cost of supercritical fluid technique (SANTANA et al., 2009). PEREIRA et al. (2016) compared the efficiency of hydrodistillation and supercritical fluid technique in essential oil extraction from the fruits of myrtle (Myrtus communis L.) and observed more potent antioxidant activity of the essential oil obtained from the latter technique.

Another comparative study of conventional solvent, supercritical fluid, and ultrasound revealed that the essential oil of avocado (Persea americana W.) obtained using supercritical fluid had greater antioxidant activity as it enabled efficient isolation of the phenolic compounds (TAN et al., 2018). Once the essential oils are obtained, they can be used in several biochemical assays and cell cultures.

Applications of fruit-derived essential oils in cell culture

In recent years, the benefits of using plant-derived essential oils have promoted diverse research. The phytochemicals (usually 2–3) present in higher concentrations are called majorities, whereas those present in smaller quantities usually determine the biological function or activity of the natural substance (SHAABAN, EL-GHORAB, SHIBAMOTO, 2012). Table 1 highlights some studies that explored these natural substances with biotechnological potential. Knowing that fruit-derived essential oils have useful biological properties, they have wide applications in biochemical tests in vitro.

With an objective to use these essential oils in biochemical studies, various tests were carried out to determine their biological properties. Identification of these properties expands the scope of their application, as with thymol, a terpene obtained from oregano (Origanum vulgare), which is used as herbal food additive owing to its multiple beneficial properties (El-HACK et al., 2015). Similarly, FERRONATTO and ROSSI (2018) identified the antioxidant properties of essential oils derived from the peel of orange (C. sinensis) and evaluated its potential. The results demonstrated that this essential oil was capable of scavenging 81.45% and 94.64% of free radicals when evaluated using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) and 2,2'-azino-bis (3-ethylbenzothiazoline) 6-sulfonic acid (ABTS) tests, respectively.

Additionally, fruit-derived essential oils are an important source of antimicrobial agents. TEIXEIRA et al. (2012) evaluated the essential oil obtained from fresh peels versus dried peels of Citrumelo swingle (Citrus paradisi × Poncirus trifoliata). It was demonstrated that fresh peel–derived essential oil showed better effect against phytopathogens than those obtained from dry peels (380–407 mg/mL and 505–528 mg/mL respectively). The study revealed that the best antimicrobial effect was observed when the combination of fresh and dried peel–derived essential oils was used, which may be due to the number of phytochemicals present in each sample as well as the complexity and synergism of each one over the phytopathogens.

Another important application of essential oils is as supplements for in vitro culture media for cells and gametes. They play vital roles in the development of new pharmacological products, toxicity tests in tumor cells, and control of free radical levels (HERMAN et al., 2019). A study by HE et al. (2019) highlighted the antioxidant and anti-melanogenic properties of essential oils derived from grapefruit (C. maxima) peel. The antioxidant capacity was evaluated by DPPH, ABTS, and superoxide anion radical tests. Grapefruit bark essential oil reduced up to 70% of the free radicals in each trial with varying dose (0.2-1.0 mg/mL). Additionally, it showed anti-melanogenic effects at 10-50 µg/mL concentration range. It was able to alter the catalytic reactions of essential enzymes in melanoma cells, such as tyrosinase associated with melanoma-cancer and neurotoxicity.

Fruit-derived essential oils are used in culturing embryos and gametes. The essential oils have also been tested in the form of male contraceptives due to their antioxidant activity. The effect of essential oil from the fruit ajowan (Trachyspermum ammi) on the viability and integrity of the human sperm membrane was evaluated (PAUL, KANG, 2010). It was observed that a concentration of 125 µg/mL was sufficient to reduce sperm viability and mitochondrial activity, possibly due to the essential oil–mediated modifications in sperm morphology and the metabolic enzymes involved. Studies on the effect of fruit-derived essential oils on female gametes and embryos are still scarce, and studies on essential oils from other plant parts have been performed largely.

CONCLUSION

Fruit derived essential oils have numerous beneficial properties for cells, such as anti-inflammatory, and antioxidant. The actions of these substances have been interesting strategies in in vitro cell culture systems. The use of different processes for extracting essential oils from fruits can lead to the discovery of different phytochemical components.

Additionally, studies on male gametes have been used as a contraceptive activity, while studies on female gametes and embryos are still scarce. It is expected that the advances already observed with the use of essential oils derived from fruits will lead to further research in other different cell types. Despite this limitation, the use of derived essential oils is still advantageous, and it is portrayed with their use in biochemical or in vitro cellular assays. Therefore, this research requires a better approach and investigation as these natural substances can be used for numerous important purposes.

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