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Development of fast dissolving orodispersible films loaded with cannabis extract

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ABSTRACT

Nowadays, cannabis plants are currently authorized to be used legally in Thailand for medical purposes. The current dosage form of cannabis extract is oil for sublingual drops. However, there are lots of limitations to these forms, including difficulty in administering, carrying, volume control, and the stability of the extract. Therefore, oral disintegrating films loaded with cannabis extract were developed to minimize the limitations of conventional dosage forms. The extraction process of cannabis leaves was conducted by the reflux technique, using ethanol as the solvent. The major compounds, cannabidiol (CBD) and tetrahydrocannabinoid (THC), from cannabis leaf extract were analyzed. The results showed that the crude extract contained CBD and THC concentrations of 2.45 mg and 9.93 mg per 100 g of the crude extract, respectively. The optimized formulation for oral disintegrating films was 2.5% hydroxypropyl methylcellulose, 1.5% propylene glycol, and 2.11% cannabis extract. The prepared films could disintegrate in 20 s when tested by the disintegration tester. Mechanical properties testing revealed that the film was strong and flexible. All of the experiments revealed that the oral disintegrating film containing cannabis extract had the potential to be commercialized and utilized instead of the conventional dosage forms.

Keywords: tetrahydrocannabinol; cannabidiol; oral disintegrating films; dissolution; hydroxypropyl methylcellulose

1. INTRODUCTION

Cannabis is a genus of plants belonging to the Cannabaceae family. There are three recognized cannabis species, which are *Cannabis sativa, Cannabis indica,* and *Cannabis ruderalis*, and over 700 strains (Pattnaik et al., 2022). Since ancient times, cannabis has been used medicinally across the globe. There are about 400 chemicals, including more than 60 cannabinoids, which are aryl-substituted meroterpenes exclusive to the cannabis plant genus (Ashton, 2001). Cannabis and its derivatives have the potential to treat epilepsy, anxiety, depression, tumors, cancer, Parkinson's disease, and Alzheimer's disease. Depending on the proportions and concentrations of cannabidiol (CBD) and tetrahydrocannabinol (THC), two major chemical components obtained from the *Cannabis sativa* plant, the cannabis extract has a variety of medical applications.

Typically, cannabis extract for clinical use has been prepared as oral oil drops (Chen and Rogers, 2019). The aim of an oral drop is not gastrointestinal absorption level due to its slow onset, unpredictable drug absorption, first pass metabolism by liver enzymes, and very low bioavailability (Hennig et al., 2016), but for oral mucosal and sublingual absorption. Nevertheless, the use of oral oil drops is limited in terms of per-drop dosing accuracy, usability, and stability. Consequently, cannabis extract necessitates a sophisticated and effective pharmaceutical dosage form formulation.

Orodispersible film (ODF) is a film-based oral dosage form that dissolves in the mouth within 30 s (Gijare and Deshpande, 2018). ODF has attracted a great deal of interest among oral drug delivery systems due to its many advantages, including its high stability, ease of handling, non-requirement of water for administration, suitability for dysphagia patients, and the ability to access the exact drug dose by adjusting the film's size (Huanbutta et al., 2021).

In an ODF formulation, a number of different natural and synthetic polymers, including pullulan, maltodextrin, starch, and gum, among others, have been used as a film former (Huanbutta et al., 2021; Pacheco et al., 2021). Due to its low toxicity, low cost, and ease of availability, hydroxypropyl methylcellulose (HPMC) has a high potential for application as a film forming material in ODF. In addition, this material's hydrophilic characteristic, which allows it to have mucoadhesive properties and to swell and dissolve in water, contributes to its tremendous potential for use in ODF formulation (Karavas et al., 2006; Kraisit et al., 2017; Kraisit et al., 2018). In the previous report, HPMC and polyethylene glycol 400 (PEG400) were both used in the hot-melting extrusion procedure for preparing ODF (Dahiya et al., 2009). The findings demonstrated that the film is flexible and disintegrates within a short period of time. The hot melt extrusion approach, on the other hand, makes use of an advantageous instrument; however, it is not appropriate for the processing of pharmaceuticals that are sensitive to heat. In order to prepare the HPMC film for this study, the solvent casting method was used.

The aim of this study was to fabricate ODF loaded with cannabis extract. Initially, *Cannabis sativa* L. leaves were extracted using the reflux technique. The CBD and THC levels of the extract were then determined using the HPLC technique. HPMC-based ODF was developed to obtain suitable mechanical properties and fast disintegration. The cannabis extract was then loaded onto the film, yielding a THC concentration of 3 mg/cm^2. In addition, the loaded cannabis extract was evaluated to determine its effect on the physical properties of the ODF.

2. MATERIALS AND METHODS

2.1 Materials

HPMC E4M (Lot No. D180K67012) was purchased from Dupont (Delaware, United States). Cannabidiol (Lot No. FE10071912) and (-)-delta9-THC-D3 (Lot No. FE05272003) standards were purchased from Sigma-Aldrich (Missouri, United States). All other chemicals and reagents were of analytical grade.

2.2 Cannabis extraction

The fresh leaves of *Cannabis sativa* L. were obtained from Thabunmee community enterprise, Chonburi province in April 2021. The *Cannabis sativa* L. leaves were identified by expert in phytochemistry. The voucher specimens (KM No.0415004) were deposited at the Faculty of Pharmaceutical Sciences, Burapha University, Thailand. The fresh leaves of *Cannabis sativa* L. were gradually dried in a chemical fume hood (FH120/2014 SE-PP, PRO LAB®, Thailand) for 1 week. Then, the dried leaves were ground and re-dried at 120°C for 30 min for decarboxylation purposes. After that, 10 g of ground and dried *Cannabis sativa* L. leaves were soaked with 500 mL of 95% ethanol and concentrated using a rotary evaporator (R-100, Buchi, Switzerland) at 90°C for 50 min. Finally, the percent yield of crude extract powder was calculated using Equation 1.

$$\text{Percent yield} = \frac{\text{weight of the extrct}}{\text{weight of the dried leaves}} \times 100 \qquad (1)$$

2.3 Identification and quantification of cannabis extract

According to the previous report (De Vita et al., 2020), two important active substances (THC and CBD) of cannabis extracts can be used to monitor the efficiency of the extraction process. THC and CBD were quantified using high-performance liquid chromatography coupled with an ultraviolet detector (HPLC-UV) (SPD-M20A, Shimadzu, Japan), as previously reported and validated by Piani and team (Piani et al., 2022). The analytical column used in the present study was a C18 reverse-phase Kinetex™ column (C18, 2504.6 mm, pore size 5 m, Phenomenex, California, USA). The mobile phase was a binary gradient of acetonitrile (A) and 0.1% trifluoroacetic acid (B) at a flow rate of 0.60 mL/min. The gradient started at 70% of A for 5 min, and then increased linearly to 77% of B in 7 min and to 90% of B in 0.10 min. This composition was kept constant up to 35 min before a linear decrease to the initial conditions (70% of A) in 0.10 min and held up to 60 min for the equilibration of the column. The detecting wavelength was 210 nm and the volume of the injecting sample was 10 µL. The HPLC system temperature was set at 35±1°C (Piani et al., 2022).

The referenced standard solutions of CBD and THC were cannabidiol and (-)-delta9-THC-D3, respectively. Both standards were dissolved in methanol in the concentration range of 0.3906 – 100 µg/mL. Then the prepared standard solutions were filtered through a 0.45-µm membrane before being injected into the HPLC. After that, the standard curves of cannabidiol and (-)-delta9-THC-D were generated. The cannabis liquid extract sample was prepared by dissolving 40 mL of liquid extract in 10 mL methanol. Then the sample was filtered by a 0.45-µm membrane before being injected into the HPLC under similar conditions as described above.

2.4 ODF preparation

2.4.1 Preliminary study

First, three concentrations of HPMC (2.5%, 5%, and 7.5% by weight) were prepared by dissolving the polymer in purified water. The film was then cast by pouring the solution into the mold and allowing it to dry at room temperature for 24 h. After determining the optimal HPMC concentration range, PEG400 or propylene glycol (PG) was added to the formulation as a plasticizer for the film in order to determine the suitable concentration range for each plasticizer.

2.4.2 ODF formulation development

The ODF formulation was developed by varying the concentrations of PEG400 and PG in 2.5% HPMC, as shown in Table 1. The concentrations of PEG400 and PG were varied between 1-3.5% and 0.3-3%, respectively. Then, all formulations were adjusted with purified water to achieve a final weight of 10 g. After achieving the optimal ODF formulation (rapid disintegration and durable film characteristics), liquid cannabis extract was added to obtain a similar concentration to the reference product from Thailand's Government Pharmaceutical Organization (GPO), THC forte (one drop contains THC of 3 mg or 81 mg/mL).

Table 1. ODF formulation

Formulation	Ingredients		
	HPMC (g)	PEG400 (g)	PG (g)
1a	0.25	0.1	-
1b	0.25	0.15	-
1c	0.25	0.2	-
1d	0.25	0.25	-
1e	0.25	0.3	-
1f	0.25	0.35	-
2a	0.25	-	0.03
2b	0.25	-	0.1
2c	0.25	-	0.15
2d	0.25	-	0.2
2e	0.25	-	0.25
2f	0.25	-	0.3

Note: Purified water was added to adjust all formulations to achieve the final weight of 10 g.

2.5 ODF evaluation

2.5.1 Film disintegration

Two methods were used to test the disintegration of the film: the beaker method and the disintegration tester. The beaker method was adapted from Hoffmann et al. (2011). The ODF film was cut into a square shape at a size of 1 cm^2. The film was then submerged in 25 mL of room-temperature purified water. Every 10 s, the water was gently swirled, and the film disintegration time was determined and recorded. The experiment was performed in triplicate.

For the second method, the USP disintegration tester (ZT 320 series, ERWEKA, Germany) was used to test the disintegration time of the square ODF film at a size of 1 cm^2. The disintegration medium consisted of 750 mL of purified water heated to 37±2°C (Huanbutta et al., 2021).

2.5.2 Film mechanical properties

The mechanical properties of the prepared ODF with/without the loaded cannabis extract were evaluated by measuring the tensile strength, breaking strain, and toughness using a texture analyzer (TA.XT plus, Stable Micro Systems, UK). The substrate was cut to a size of 1 × 10 cm^2. The substrate was then fixed from top to bottom to obtain a gap length of 1 cm. The substrate was then stretched at a rate of 100 mm per minute using a 0.05-N pulling force. The maximum force and pulling length before film breakage were recorded. The toughness values were then determined by calculating the entire positive area under the force versus time curve (Boateng et al., 2009). Each test was repeated 6 times.

2.6 Statistical analysis

The analysis of variance test and Levene's test for the homogeneity of variance were performed using SPSS version 10.0 for Windows (SPSS Inc., Chicago, IL, USA). Post-hoc testing ($p<0.05$) of the multiple comparisons was performed by performing either the Scheffé or Games–Howell test depending on whether Levene's test for homogeneity is insignificant or significant (Patom chaiviwat et al., 2022)

3. RESULTS AND DISCUSSION

3.1 Appearance and percent yield of cannabis crude extract

The cannabis leaf crude extract from the reflux method using 95% ethanol as menstruum was dark green and resinous. The cannabis crude extract percent yield was 11%. This extract yield result was quite similar to the previous study by Rožanc et al. (2021). They reported that the extraction yield of *Cannabis sativa* using ethanol was 13.2% by weight.

3.2 Identification and quantification of cannabis extract

As reference standards, cannabidiol and (-)-delta9-THC-D3 were used to identify and quantify the CBD and THC content of the cannabis leaf extract, respectively. In accordance with a prior finding, the retention periods of the reference standards, cannabidiol and (-)-delta9-THC-D3, at a concentration of 50 µg/mL were 12.87 and 15.97 min, respectively (Figure 1a). Then, the reference standards were diluted to achieve a concentration range of 0.39–50 µg/mL, and the entire dilution was injected into the HPLC to generate a standard curve. After that, the cannabis leaf extract at a concentration of 4000 µg/mL was analyzed by HPLC technique, as shown in Figure 1b. The results revealed that 1,110 mg of the crude extract from cannabis leaves contained 27.2 µg of CBD and 9835.2 µg of THC. This result is in agreement with the prior work, in which they discovered that the CBD level of commercial cannabis leaves was significantly lower than the THC concentration by approximately 100 times (Bernstein et al., 2019). On the other hand, the yield of CBD and THC from the cannabis leaves sample from Chonburi, Thailand, was lower than that reported by Bernstein et al. (2019). This could be because of the plating condition or the diversity of cannabis strains.

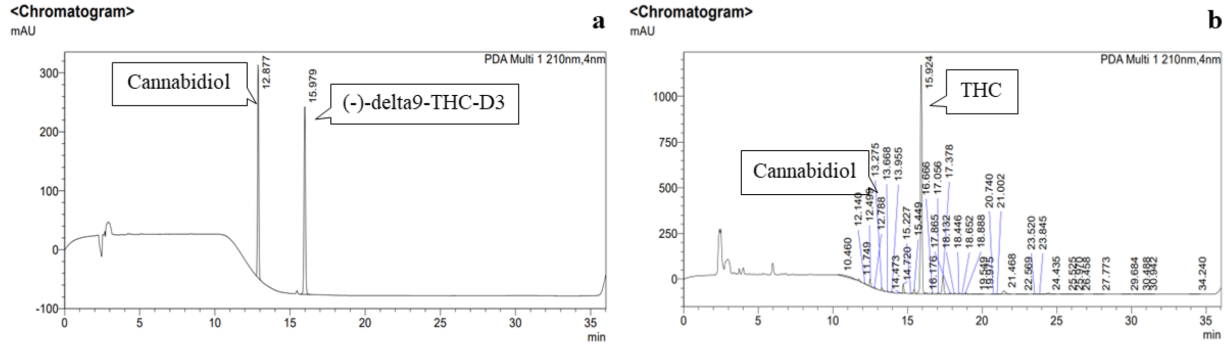

Figure 1. HPLC chromatograms of (a) the reference standards (cannabidiol and (-)-delta9-THC-D3), and (b) the cannabis leaf extract

3.4 Film disintegration

Figure 2 displays the results of the film disintegration experiments conducted using the beaker and disintegration tester methods. All of the ODF disintegration times were lower than 65 s. The disintegration tester yielded significantly shorter disintegration times than the beaker technique. This could be the result of the disintegration tester's applied force, medium volume, and temperature accelerating the process of film disintegration. The addition of greater than 2.5% PEG400 (formulations 1d–1f) could delay film degradation. This may be due to the fact that PEG400 boosts the hydrophobic characteristics of the ODF, making it difficult for water to penetrate the polymer layer (Khunteta et al., 2019). While adding PG, the disintegration time is barely altered. According to prior studies, the hydroxyl groups of PG could partially hydrate the film; hence, water molecules can penetrate the film more effectively (Rhys et al., 2016). Due to their rapid disintegration and excellent mechanical film characteristics, the ODF formulations 1b and 2c were chosen for loading cannabis crude extract. As demonstrated in Figure 3b, the addition of the extract resulted in a slight change in the disintegration times. All of the ODF loaded with cannabis extract disintegrated in less than 35 s, which is acceptable according to criteria in European Pharmacopoeia, which stipulate that the disintegration time must be less than 180 s.

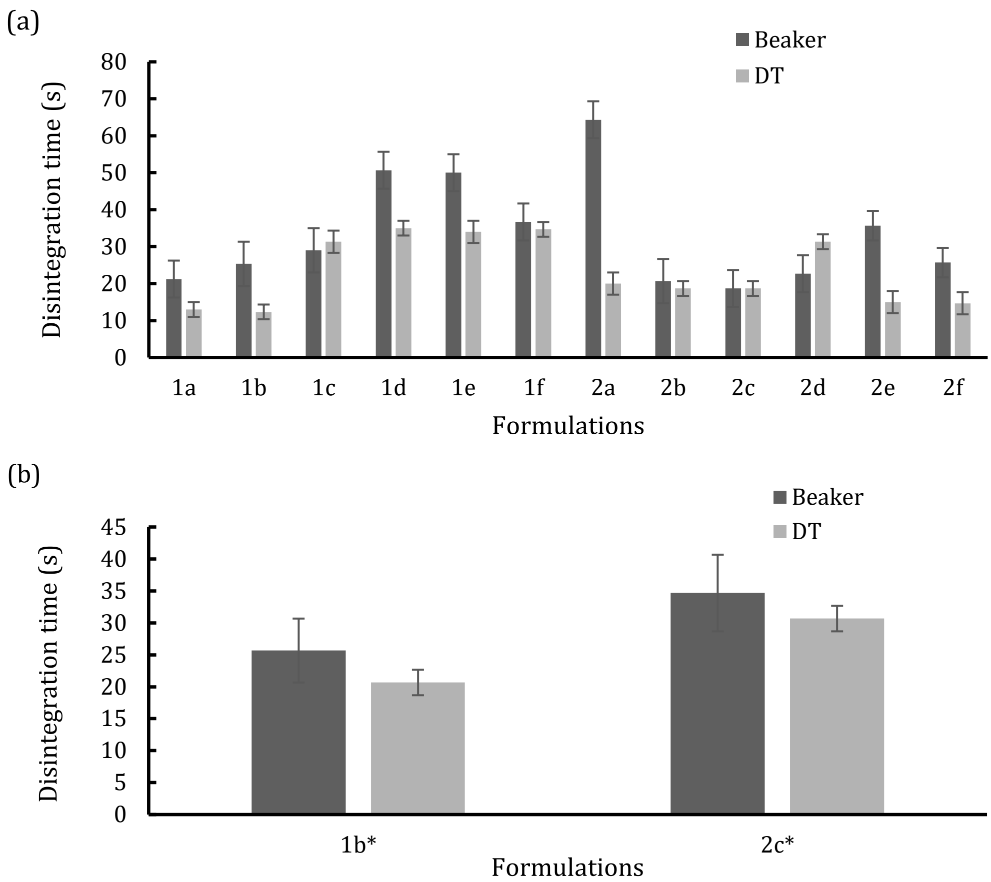

Figure 2. Disintegration time of different formulations of (a) the blank ODF, and (b) the extract loaded ODF evaluated by the beaker and disintegration tester (DT) methods

3.5 Film mechanical properties

The mechanical properties of the ODF, including mechanical strength, breaking strain, and toughness, are presented in Figure 3. It may be inferred from the results that PEG400 boosted the film strength of ODF more than PG (Figure 3a). In contrast, PG provided greater film flexibility than PEG400, as evidenced by the breaking strain (Figure 3b) and film toughness (Figure 3c). PEG400 and PG were both capable of transforming the severely brittle mechanical properties of film into more malleable and stretchable qualities. This is due to the ability of plasticizer molecules ability to insert themselves into the HPMC polymer chain. This makes the van der Waal force between the HPMC molecules weaker, which makes the film more flexible (Lim and Hoag, 2013). According to Orliac et al. (2003), the reason that PEG400 was able to improve film flexibility more than PG was likely due to the fact that PEG400 has a large molecular size. This large molecular size prevented HPMC from becoming fully integrated into the network when very large quantities were used (Orliac et al., 2003). However, an oily feel in ODF can be caused by an excessively high proportion of PEG400 in the film.

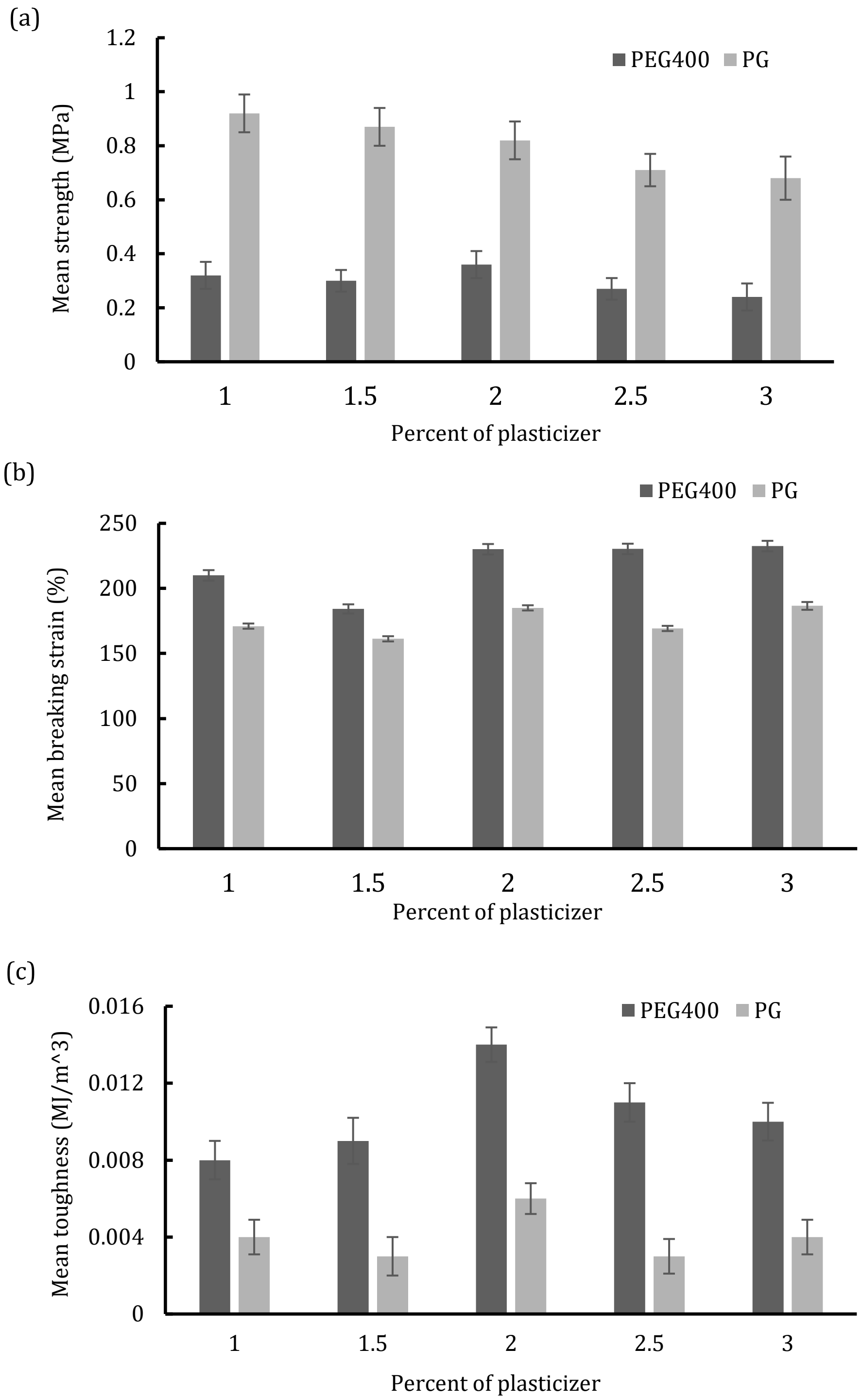

Figure 3. Effect of plasticizers on the (a) mechanical strength, (b) strain, and (c) toughness of the ODF

5. CONCLUSION

The results indicated that an ODF containing cannabis extract could be developed successfully. The film loaded with extract could disintegrate in less than 35 s, which is acceptable according to the EU pharmacopeia. The mechanical characteristics of ODF were acceptable after a sufficient amount of the plasticizers (PEG400 and PG) was added. The cannabis extract was added to the ODF formulation to achieve a THC concentration of 3 mg/cm^2, which is comparable to the THC Forte, a commercial product, which has a THC content of 3 mg/drop. The developed ODF has an advantage over the conventional oil formulation in terms of drug administration precision and adjustability. This is crucial due to the fact that THC has a narrow therapeutic index and requires a precise dose to reduce adverse effects and improve therapeutic efficacy.

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REFERENCES

Ashton, C. H. (2001). Pharmacology and effects of cannabis: a brief review. *The British Journal of Psychiatry*, 178(2), 101-106.

Bernstein, N., Gorelick, J., Zerahia, R., and Koch, S. (2019). Impact of N, P, K, and humic acid supplementation on the chemical profile of medical cannabis (*Cannabis sativa* L). *Frontiers in Plant Science*, 10, 736.

Boateng, J. S., Stevens, H. N., Eccleston, G. M., Auffret, A. D., Humphrey, M. J., and Matthews, K. H. (2009). Development and mechanical characterization of solvent-cast polymeric films as potential drug delivery systems to mucosal surfaces. *Drug Development and Industrial Pharmacy*, 35(8), 986-996.

Chen, P. X., and Rogers, M. A. (2019). Opportunities and challenges in developing orally administered cannabis edibles. *Current Opinion in Food Science*, 28, 7-13.

Dahiya, M., Saha, S., and Shahiwala, A. F. (2009). A review on mouth dissolving films. *Current Drug Delivery*, 6(5), 469-476.

De Vita, D., Madia, V. N., Tudino, V., Saccoliti, F., De Leo, A., Messore, A., Roscilli, P., Botto, A., Pindinello, I., Santilli, G., Scipione, L., Costi, R., and Di Santo, R. (2020). Comparison of different methods for the extraction of cannabinoids from cannabis. *Natural Product Research*, 34(20), 2952-2958.

Gijare, C., and Deshpande, A. (2018). Orodispersible films: A systematic patent review. *Recent Patents on Drug Delivery & Formulation*, 12(2), 110-120.

Hennig, I. M., Barrett, D. A., Constantinescu, C. S., Fischer, P. M., and Gershkovich, P. (2016). Dietary fats and pharmaceutical lipid excipients increase systemic exposure to orally administered cannabis and cannabis-based medicines. *American Journal of Translational Research*, 8(8), 3448.

Hoffmann, E. M., Breitenbach, A., and Breitkreutz, J. (2011). Advances in orodispersible films for drug delivery. *Expert Opinion on Drug Delivery*, 8(3), 299-316.

Huanbutta, K., Sriamornsak, P., Singh, I., and Sangnim, T. (2021). Manufacture of 2D-printed precision drug-loaded orodispersible film prepared from tamarind seed gum substrate. *Applied Sciences*, 11(13), 5852.

Karavas, E., Georgarakis, E., and Bikiaris, D. (2006). Application of PVP/HPMC miscible blends with enhanced mucoadhesive properties for adjusting drug release in predictable pulsatile chronotherapeutics. *European Journal of Pharmaceutics and Biopharmaceutics*, 64(1), 115-126.

Khunteta, A., Gupta, M. K., and Swarnkar, S. K. (2019). Formulation of rapid dissolving films containing granisetron hydrochloride and ondansetron hydrochloride. *Journal of Drug Delivery and Therapeutics*, 9(4A), 516-527.

Kocis, P. T., and Vrana, K. E. (2020). Delta-9-tetrahydrocannabinol and cannabidiol drug-drug interactions. *Medical Cannabis and Cannabinoids*, 3(1), 61-73.

Kraisit, P., Limmatvapirat, S., Luangtana-Anan, M., and Sriamornsak, P. (2018). Buccal administration of mucoadhesive blend films saturated with propranolol loaded nanoparticles. *Asian Journal of Pharmaceutical Sciences*, 13(1), 34-43.

Kraisit, P., Limmatvapirat, S., Nunthanid, J., Sriamornsak, P., and Luangtana-Anan, M. (2017). Preparation and characterization of hydroxypropyl methylcellulose/polycarbophil mucoadhesive blend films using a mixture design approach. *Chemical and Pharmaceutical Bulletin*, 65(3), 284-294.

Lim, H., and Hoag, S. W. (2013). Plasticizer effects on physical-mechanical properties of solvent cast Soluplus® films. *AAPS Pharmscitech*, 14(3), 903-910.

Orliac, O., Rouilly, A., Silvestre, F., and Rigal, L. (2003). Effects of various plasticizers on the mechanical properties, water resistance and aging of thermo-moulded films made from sunflower proteins. *Industrial Crops and Products*, 18(2), 91-100.

Pacheco, M. S., Barbieri, D., da Silva, C. F., and de Moraes, M. A. (2021). A review on orally disintegrating films (ODFs) made from natural polymers such as pullulan, maltodextrin, starch, and others. *International Journal of Biological Macromolecules*, 178, 504-513.

Patomchaiviwat, V., Sriamornsak, P., Chansiri, G., Limmatvapirat, S., Supawattanakul, A., Chonganon, T., Keattiteerachai, A., and Piriyaprasarth, S. (2022). Development of edible bubbles of calcium alginate for encapsulating energy drinks. *Science, Engineering and Health Studies*, 16, 22050018.

Pattnaik, F., Nanda, S., Mohanty, S., Dalai, A. K., Kumar, V., Ponnusamy, S. K., and Naik, S. (2022). Cannabis: Chemistry, extraction and therapeutic applications. *Chemosphere*, 289, 133012.

Piani, B., Ferfuia, C., Bortolomeazzi, R., Verardo, G., and Baldini, M. (2022). Development and optimization of an HPLC-PDA method for the determination of major cannabinoids in hemp (*Cannabis sativa* L.) essential oil obtained by hydrodistillation. *Food Analytical Methods*, 15(6), 1677-1686.

Rhys, N. H., Gillams, R. J., Collins, L. E., Callear, S. K., Lawrence, M. J., and McLain, S. E. (2016). On the structure of an aqueous propylene glycol solution. *The Journal of Chemical Physics*, 145(22), 224504.

Rožanc, J., Kotnik, P., Milojević, M., Gradišnik, L., Knez Hrnčič, M., Knez, Ž., and Maver, U. (2021). Different *Cannabis sativa* extraction methods result in different biological activities against a colon cancer cell line and healthy colon cells. *Plants*, 10(3), 566.