Influence of Candelilla Wax on Mechanical Strength, Thermal Stability, and Moisture Resistance of Cassava-based Thermoplastic Starch Composites

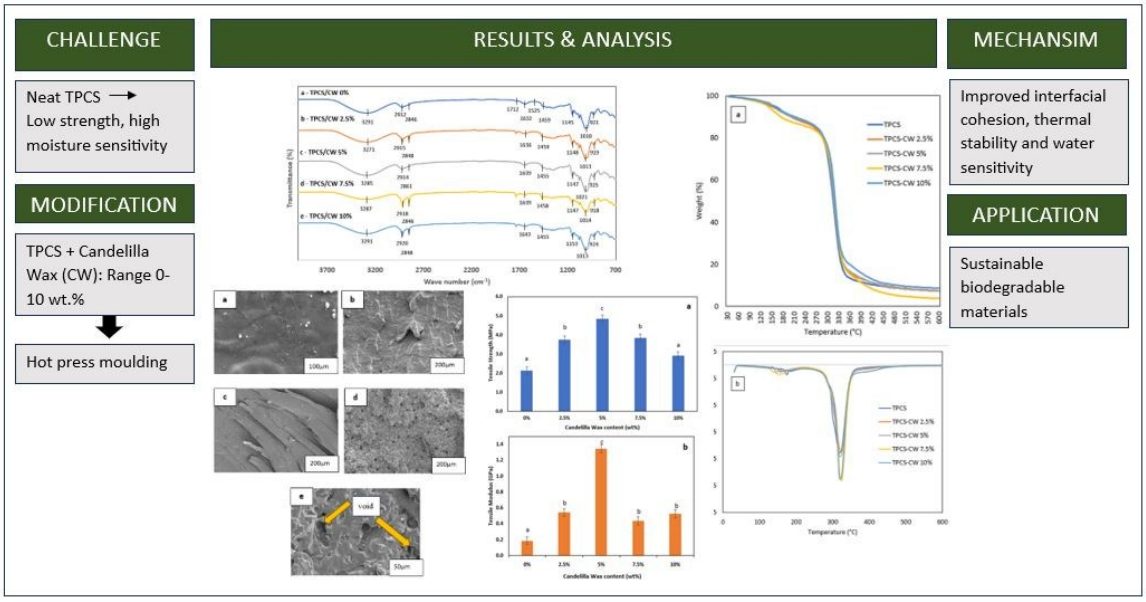
Mohd Fairus Kayat,[a] Ridhwan Jumaidin,[b,*] Mastura Mohamad Taha,[a,*] Fahmi Asyadi Md Yusof,[c] Zatil Hafila Kamaruddin,[d] Mohammad Khalid Wahid,[a] and Riza Wirawan [e]

** Corresponding authors: ridhwanj@ums.edu.my; mastura.taha@utem.edu.my*

DOI: 10.15376/biores.20.4.11074-11097

GRAPHICAL ABSTRACT

Graphical Abstract: Influence of Candelilla Wax on Mechanical Strength, Thermal Stability, and Moisture Resistance of Cassava-based Thermoplastic Starch Composites



Influence of Candelilla Wax on Mechanical Strength, Thermal Stability, and Moisture Resistance of Cassava-based Thermoplastic Starch Composites

Mohd Fairus Kayat,[a] Ridhwan Jumaidin,[b,*] Mastura Mohamad Taha,[a,*] Fahmi Asyadi Md Yusof,[c] Zatil Hafila Kamaruddin,[d] Mohammad Khalid Wahid,[a] and Riza Wirawan [e]

A bio-based reinforcement strategy was studied as a means to overcome the inherent weaknesses of thermoplastic cassava starch (TPCS), namely its low mechanical strength and high susceptibility to moisture. Candelilla wax, a natural hydrophobic additive, was incorporated into TPCS at different loadings (0, 2.5, 5, 7.5, and 10 wt %) and processed *via* hot-press compression moulding. The fabricated composites were characterised to examine the effects of wax addition on their mechanical, thermal, and moisture-resistance performance. Mechanical tests (tensile, flexural, and impact), with scanning electron microscopy (SEM), thermogravimetric analysis (TGA). The incorporation of candelilla wax notably improved the material's performance, particularly at 5 wt%, where tensile strength and modulus increased 77.4% and 615%, respectively. Flexural and impact strength also increased, indicating enhanced toughness. The SEM micrographs showed rougher fracture surfaces with increasing wax, while Fourier transform infrared spectroscopy (FT-IR) confirmed intermolecular hydrogen bonding between starch and wax. Improved thermal stability and reduced water sensitivity were also observed, with the 10 wt % wax composites exhibiting the lowest moisture absorption, solubility, and swelling. Overall, candelilla wax proved effective in strengthening TPCS both structurally and functionally, highlighting its potential for sustainable biodegradable materials in moisture-sensitive applications.

DOI: 10.15376/biores.20.4.11074-11097

Keywords: Biodegradable; Thermoplastic cassava starch (TPCS); Candelilla wax; Mechanical properties; Thermogravimetric analysis (TGA); Scanning electron microscope (SEM)

Contact information: a: Fakulti Teknologi dan Kejuruteraan Industri dan Pembuatan, Universiti Teknikal Malaysia Melaka, Hang Tuah Jaya, 76100 Durian Tunggal, Melaka, Malaysia; *b: Faculty of Engineering, Universiti Malaysia Sabah, Jalan UMS, 88400 Kota Kinabalu, Sabah, Malaysia*; *c: Malaysian Institute of Chemical & Bioengineering Technology (UniKL MICET), Taboh Naning, 78000 Alor Gajah, Melaka, Malaysia*; *d: German-Malaysian Institute, Jalan Ilmiah Taman Universiti, Kajang 43000, Malaysia*; *e: Faculty of Mechanical and Aerospace Engineering, Institut Teknologi Bandung, Jalan Ganesa 10 Bandung, Jawa Barat, Indonesia;*
** Corresponding authors: ridhwanj@ums.edu.my; mastura.taha@utem.edu.my*

INTRODUCTION

Plastics derived from petroleum have long supported industrial progress due to their durability, and adaptability, becoming deeply integrated into daily life across various sectors. However, their widespread use, especially in packaging, has come at an environmental cost. As global population growth accelerates, the demand for consumer products has increased substantially, and the generation of solid waste has increased substantially, with fossil-based plastics making up a significant portion. Although these

materials are inexpensive and widely used, their resistance to microbial activity and natural degradation makes them persist in the environment for decades. This persistence contributes to serious environmental pollution, affecting natural ecosystems and depleting essential resources. The accumulation of such non-degradable waste has raised pressing ecological concerns, including disruption to land and marine habitats. As a result, there is growing interest in developing sustainable and biodegradable alternatives that can reduce the ecological footprint of plastic use while promoting the development of cost-effective and eco-friendly polymers derived from natural resources (Azlin *et al.* 2020; Hazrati *et al.* 2021a). Bio-based polymers, derived from renewable sources, are gaining attention as viable substitutes, offering both environmental and economic advantages in tackling the global plastic waste crisis (Babaee *et al.* 2015).

Starch is a biopolymer with potential for producing eco-friendly plastics because it is abundant, inexpensive, renewable, and inherently biodegradable (Campos *et al.* 2018). It can also be converted into other chemicals, such as ethanol and acetone, through enzymatic hydrolysis and microbial fermentation processes (Öner *et al.* 2005; Bušić *et al.* 2018). Starch from sources, such as corn, potato, sago, and cassava, has many advantages but also notable drawbacks, including high moisture absorption and relatively low mechanical and thermal strength (Karimi *et al.* 2014; Hazrati *et al.* 2021b). Starch is naturally semi-crystalline and exists in granules that require plasticisation to become flexible and useful as a thermoplastic material (Rocha *et al.* 2012). The addition of plasticisers such as glycerol, citric acid, or sorbitol is a common modification technique, as these additives disrupt native starch hydrogen bonding and form new bonds with starch molecules, thereby improving thermal stability, moisture resistance, and tensile properties (Sanyang *et al.* 2016; Hazrati *et al.* 2021b). Such modifications expand the potential applications of starch-based plastics.

Cassava (*Manihot esculenta*) is an attractive starch source because of its exceptionally high starch yield (up to ~17,000 kg/ha, much higher than other starch crops) (Jumaidin *et al.* 2020). Cassava can grow in relatively poor soil and arid climates with low or inconsistent rainfall (Florencia *et al.* 2020). Cassava starch is widely used to produce biopolymer materials, such as biodegradable films, for packaging because of its availability, low cost, and biodegradability (Bergo *et al.* 2010). Such products include rigid packaging containers (Kaisangsri *et al.* 2012), and food-grade edible coatings (Chiumarelli and Hubinger 2014). However, native starch is brittle and possesses low water resistance. These limitations can be addressed by various modification techniques, including adding plasticisers or blending with other polymers, chemical derivatization, and graft copolymerisation (Hulleman *et al.* 1998; Curvelo *et al.* 2001; Chiumarelli *et al.* 2010; Prachayawarakorn *et al.* 2010; Kaisangsri *et al.* 2012; Li *et al.* 2013). Focusing on cassava starch as a feedstock for thermoplastic composites aligns with the goal of integrating renewable materials into a sustainable circular economy.

Thermoplastic starch (TPS) refers to native starch that has been plasticised so that it can be softened by heat and pressure and moulded into shapes (Ma *et al.* 2005). In TPS production, a plasticiser (such as glycerol) is blended with native starch and the mixture is heated to disrupt the starch granule structure. This process breaks the original intermolecular hydrogen bonds in starch granules and replaces them with new hydrogen bonds between starch and plasticiser molecules (Curvelo *et al.* 2001). As a result, the starch structure becomes more flexible and mouldable (Hulleman *et al.* 1998). Even so, TPS materials still have two main shortcomings: weak mechanical properties and high water affinity (susceptibility to moisture) (Prachayawarakorn *et al.* 2010). A proven approach to

overcome these issues is blending TPS with more hydrophobic materials, which is relatively simple, effective, and cost-efficient (Li *et al.* 2013). Accordingly, fillers such as waxes, chitosan, and carrageenan have been studied for improving TPS properties (Rosa and Andrade 2004; Silva *et al.* 2008; Prachayawarakorn *et al.* 2012, 2013; Rodrigues *et al.* 2014).

Candelilla wax is a natural wax with multiple functional uses. For example, it can be combined with other ingredients to create hydrophobic coatings that minimize moisture loss during fruit storage (Bucio *et al.* 2021). It is also used as an ingredient in certain vegan products as a plant-based wax alternative (Aguirre-Joya *et al.* 2019). Candelilla wax is obtained from the leaves and stems of the shrub *Euphorbia antisyphilitica* by boiling in a mildly acidic solution containing dilute sulfuric acid, which separates the wax from plant tissues (Bucio *et al.* 2021; Aranda-Ledesma *et al.* 2022). This wax is known for its hardness and brittleness, and it produces a glossy surface, making it useful in confections (such as chocolate) and fruit coatings (Bucio *et al.* 2021). Candelilla wax has been successfully applied in developing biodegradable, functional packaging materials that help preserve the physicochemical stability of food during storage. Moreover, candelilla wax exhibits some antimicrobial properties and reduces water loss and regulates gas exchange in coated products (Aranda-Ledesma *et al.* 2022). International collaboration has led to innovations in edible packaging derived from candelilla wax, mainly of two types: one is an aqueous wax-based solution that can be sprayed on food items as an edible coating, and the other is a wax-containing biopolymeric matrix that forms solid edible films applied to foods (Kowalczyk and Biendl 2016; Aguirre-Joya *et al.* 2017; Kowalczyk *et al.* 2017; De León-Zapata *et al.* 2018).

Despite extensive studies on TPS composites with various fillers, the use of candelilla wax in the thermoplastic cassava starch (TPCS) matrix remains largely unexplored. Thus, the present study addressed this gap by examining how the incorporation of candelilla wax influences the thermal performance, mechanical characteristics, dimensional stability (thickness swelling), water solubility, and moisture uptake of TPCS. Emphasising these properties is important, as improving them would broaden the applicability of TPCS as a sustainable material.

EXPERIMENTAL

Materials

Cassava starch suitable for food applications was procured from Antik Sempurna Sdn. Bhd., located in Selangor, Malaysia. Candelilla wax, primarily composed of hydrocarbons (about 50%), with hentriacontane (C31) as its major constituent, was sourced from Evergreen Engineering & Resources Sdn. Bhd., also based in Selangor. Additionally, glycerol of analytical grade and 99.5% purity was supplied by QRec Chemicals Sdn. Bhd., Selangor, to function as a plasticising agent.

Preparation of the Sample

The TPCS samples were prepared by incorporating 30 wt% glycerol as a plasticiser into the cassava starch. The cassava starch and glycerol were premixed using a Panasonic MX-GM1011 dry mixer at 1200 rpm for 5 min at ambient temperature. The mixture was then compression-moulded into sheets (approximately 3 mm thick) using a GOTECH GT7014-P30C hydraulic hot-press (Taichung City, Taiwan) at 150 °C for 60 min under a

pressure of 10 tons. This same processing method was used to produce TPCS composites containing various candelilla wax loadings (2.5, 5, 7.5, and 10 wt %). In each case, the wax was added to the starch-glycerol blend before hot pressing to obtain composites with tailored wax content. Thermoplastic Cassava Starch Samples (TPCS) were prepared in five formulations with candelilla wax contents of 0, 2.5, 5, 7.5, and 10 wt %. For each formulation, five replicate specimens were fabricated to cover all mechanical, thermal, and moisture tests as per Table 1.

Table 1. Sample Code and Candelilla Wax Loading (wt%)

Sample Code	Candelilla Wax Loading (wt %)	Description
TPCS/CW-0	0 wt %	Neat thermoplastic cassava starch (control)
TPCS/CW-2.5	2.5 wt %	TPCS blended with 2.5 wt% % candelilla wax
TPCS/CW-5	5 wt %	TPCS blended with 5 wt% % candelilla wax
TPCS/CW-7.5	7.5 wt %	TPCS blended with 7.5 wt% % candelilla wax
TPCS/CW-10	10 wt %	TPCS blended with 10 wt% % candelilla wax

Analysis of FT-IR

Fourier Transform Infrared (FT-IR) spectroscopy was employed to examine the functional groups and interactions between the thermoplastic starch matrix and candelilla wax. The FT-IR spectra for each sample were recorded on a JASCO FTIR-6100 spectrometer (Jasco, Japan) over the wavenumber range 4000 to 500 cm^{-1}. This spectral range allowed identification of key molecular bonds and any changes in the chemical structure of the composites due to wax incorporation.

Scanning Electron Microscope (SEM)

The surface morphology of tensile-fractured specimens was examined by SEM. A Zeiss EVO 18 Research SEM (Zeiss, Germany) was operated at an accelerating voltage of 10 kV to image the fracture surfaces. Prior to imaging, samples (3-mm-thick tensile pieces) were cut to the required size and coated with a thin layer of gold using a sputter coater to prevent charging. Fractured specimens were stored in sealed plastic bags until subjected to SEM analysis to prevent moisture uptake or dust contamination.

Thermogravimetric Analysis (TGA)

Thermal stability and decomposition behavior of the composites were evaluated by TGA. Approximately 5 to 10 mg of each sample was placed in an alumina crucible and loaded into a Mettler-Toledo Q-series thermogravimetric analyzer (Mettler-Toledo, Switzerland). The temperature was ramped from 25 °C to 600 °C at 10 °C/min under a nitrogen atmosphere.

Tensile Test

Tensile properties were measured according to ASTM D638 (2022). All tests were conducted in a controlled environment (23 ± 1 °C and 50 ± 5% relative humidity). Dog-bone shaped specimens (Type V) were pulled using an Instron 5969 universal testing machine (Instron, USA) equipped with a 50 kN load cell. The crosshead speed was set to 5 mm/min. Five specimens per formulation were tested, and the average values of tensile strength, tensile modulus (Young's modulus), and elongation at break were calculated.

Flexural Test

Flexural properties were evaluated following ASTM D790 (2017). Tests were conducted at 23 ± 1 °C and 50 ± 5% relative humidity. Rectangular specimens (130 mm × 13 mm × 3 mm) were tested in three-point bending on the Instron 5969 machine with a 50 kN load cell. The crosshead speed was 2 mm/min. Five replicate specimens for each composite formulation were tested to obtain average flexural strength and flexural modulus values.

Impact Test

Impact strength was measured using the Izod impact ASTM D638 (2022). Specimens (un-notched) of dimensions 60 mm × 13 mm × 3 mm were conditioned at 23 ± 1 °C and 50 ± 5% relative humidity for at least 48 h prior to testing. A digital pendulum impact tester (Victor Equipment Resources, Malaysia) was used to break five replicate specimens for each formulation. The impact strength (kJ/m²) was calculated as the absorbed energy divided by the cross-sectional area of the sample. The formula for impact strength is given in Eq. 1. All samples were conditioned for two days at ambient conditions followed by two days at 53% relative humidity before testing to ensure consistent moisture content.

$$\text{Impact strength (J/mm}^2\text{)} = \text{Impact energy (J)} / \text{Cross-sectional area (mm}^2\text{)} \quad (1)$$

Moisture Absorption

For moisture uptake evaluation, the TPCS/CW composites were placed inside a sealed Memmert GmbH climate chamber (Germany), maintained at 25 ± 2 °C and 75 ± 2% relative humidity. Five specimens of (10 × 10 × 3) mm³ were created and pre-dried at 105 °C ± 2 °C for 24 h prior to testing. Sample masses were recorded before and after exposure as M_i (initial mass, g) and M_f (final mass, g) to determine moisture uptake according to Eq. 2. The experiment was conducted until the samples achieved equilibrium moisture content.

$$Moisture\ Absorption\ (\%) = \frac{Mf - Mi}{Mi}\ x\ 100 \quad (2)$$

Water Solubility

The technique used for determining water solubility was adapted from a previous study (Reddy *et al.* 2018). Five specimens, each measuring 10 × 10 × 3 mm³, were first oven-dried at 105 ± 2 °C for 24 h to determine the initial weight (W_i). After completing this step, all specimens were subjected to agitation in a distilled water bath containing 30 mL of distilled water. Following 24 h, the specimens underwent a filtration protocol wherein the unfiltered particulate matter was retained, some water was evacuated using filter paper, and the specimen was subsequently dried at 105 ± 2 °C. The final weight acquired in this step was termed W_f, or final weight. Equation 3 was applied to calculate the percentage of water solubility:

$$Water\ Solubility\ (\%) = \frac{W_i - W_f}{W_i}\ x\ 100 \quad (3)$$

Thickness Swelling

A few approaches were based on previous studies (Jawaid *et al.* 2011; Kamaruddin *et al.* 2023) to analyse the swelling behaviour of composites. The composites were in the form of five specimens measuring (10 × 10 × 3) mm^3. The samples were scraped and placed

into an oven set at 105 ± 2 °C for a day. Each sample was taken with a baseline measure of thickness. Specimens were placed into 30 mL of distilled water maintained at room temperature (23 ± 1 °C) for 2 h. Thickness swelling (%) was calculated from the initial thickness (T_i, mm) and final thickness (T_f, mm) of each specimen after immersion using Eq. 4.

$$Thickness\ swelling\ (\%) = \frac{Ti - Tf}{Ti}\ x\ 100 \quad (4)$$

Statistical Analysis

The experimental data were statistically analysed using ANOVA to evaluate the differences among sample groups. Duncan's multiple range test was applied to determine significant differences between means at a confidence level of $p < 0.05$. All statistical analyses were performed using IBM SPSS Statistics software (Version 25.0, IBM Corp., Armonk, NY, USA).

RESULTS AND DISCUSSION

Analysis of FT-IR

The FT-IR spectra of TPCS with different candelilla wax contents are presented in Fig. 1. Changes in specific absorption bands provide evidence of both chemical and physical interactions between the components in the composites. For the control sample, Minor absorbance bands observed in the 2920 to 2850 cm^{-1} region of the neat starch spectrum correspond to C–H stretching of trace lipid or fatty acid residues commonly present in native cassava starch granules (Campos *et al.* 2018). Their presence indicates the inherent hydrophobic character of the neat TPCS matrix and supports the slight baseline peaks seen in spectral curve (a).

With the addition of candelilla wax, the intensity of the bands in this region increased markedly, corresponding to CH_2 stretching vibrations. This enhancement was associated with the contribution of alkanes and long-chain fatty alcohols that generally constitute the wax (Oliveira *et al.* 2020). Another band that grew stronger with higher wax content appeared at 1712 cm^{-1}, which is associated with C = O stretching of ester carbonyl groups; this band became more pronounced as wax was added (Aranda-Ledesma *et al.* 2022). Conversely, the absorbance peak at 1525 cm^{-1}, attributed to C–N stretching and N–H bending motions, remained unchanged across all formulations.

Furthermore, the system component interactions were corroborated by the downward shift of the broad O–H stretching band, originally located in the range of 3100 to 3700 cm^{-1} (Li *et al.* 2024). In the TPCS/CW composite, the O–H peak moved from 3291 to 3271 cm^{-1} as the candelilla wax concentration increased from 0 to 2.5 wt%, indicating the formation of new hydrogen bonds between starch molecules and wax components (Wu *et al.* 2009). Comparable interactions between starch and wax have also been identified in prior research on arrowroot starch blended with carnauba wax films (Oliveira *et al.* 2020).

Although candelilla wax contains a small fraction of hydroxyl-bearing sterols that might contribute weak polar interactions, the prevailing compatibility with starch is more plausibly mediated by non-polar van der Waals forces and dispersion effects rather than by strong chemical bonding (Muñoz-Gimena *et al.* 2023). The semicrystalline nature of thermoplastic starch allows amylose chains to adopt partial helical (V-type) conformations capable of accommodating hydrophobic molecules such as wax components within or

around the helix structure. This spatial arrangement could stabilize the starch–wax interface through van der Waals interactions, as the hydroxyl groups of amyloses are oriented inward while the hydrophobic chains interact externally with the wax phase (Zhang *et al.* 2018; Karwa *et al.* 2023; Karnwal *et al.* 2025). Such a mechanism is consistent with the moderate band shifts observed in the FT-IR spectra, which imply weak interfacial interactions rather than extensive hydrogen bonding. Comparable behaviour has also been reported for other starch–lipid or starch–wax systems, where hydrophobic additives act primarily as dispersed fillers contributing to interfacial cohesion rather than covalent bonding (Galus *et al.* 2020; Oliveira *et al.* 2020). These intermolecular associations correspond well with the observed improvements in mechanical performance, suggesting that the inclusion of candelilla wax enhances interfacial cohesion and contributes to the overall strength and stability of the TPCS/CW composites.

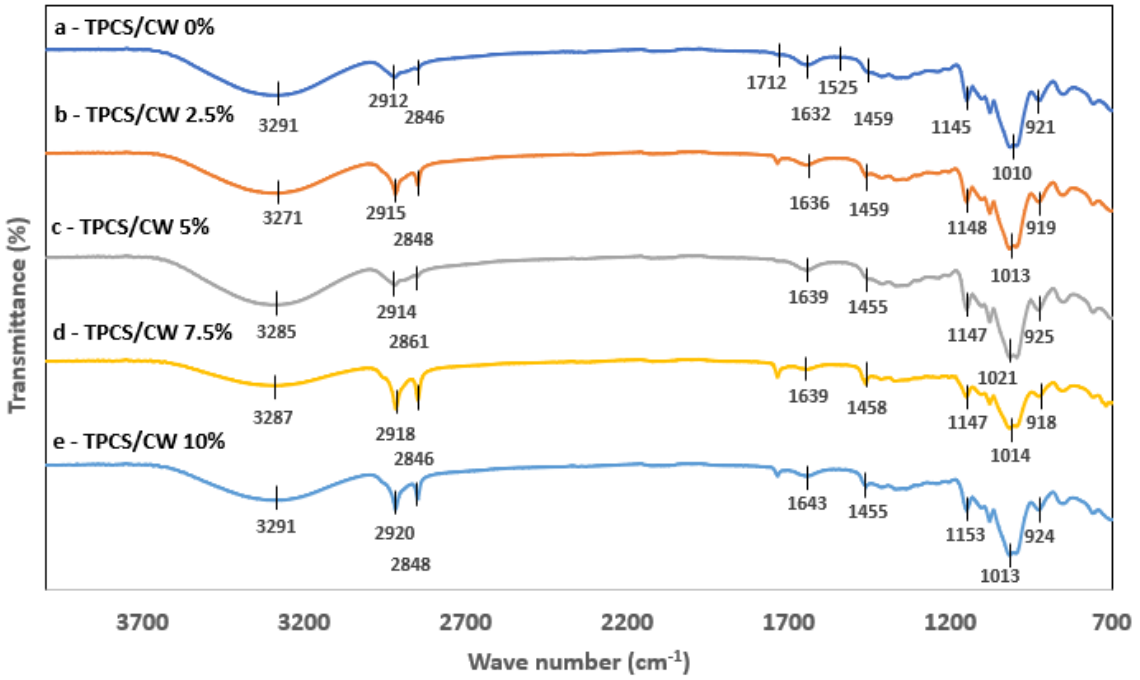


Fig. 1. The spectrum of FT-IR: a) TPCS, b) TPCS/CW wax 2.5%, c) TPCS/CW wax 5%, d) TPCS/CW wax 7.5%, and e) TPCS/CW wax 10%

SEM Analysis

Figure 2 presents the SEM micrographs of the fractured surfaces of TPCS composites modified with candelilla wax. The microstructure of the TPCS appeared tightly packed and dense, with no observable porosity, although some undissolved starch granules were noticeable. As illustrated in Fig. 2a, the reference sample exhibited a smooth and uniform surface texture with no signs of phase separation, suggesting strong interactions between the starch matrix and the glycerol plasticiser (Jumaidin *et al.* 2017).

Following the addition of candelilla wax, the surface morphology of the composites became less homogeneous, showing a coarser and stiffer appearance with surface irregularities, as depicted in Figs. 2b–2e. As the filler loading increased from 0 to 10 wt %, the fracture surfaces displayed more pronounced irregularities (Oliveira *et al.* 2020), accompanied by the emergence of pores. The presence of voids and discontinuities at higher wax concentrations, particularly in the 10 wt % sample, indicates partial phase

separation between starch and wax. These voids act as stress concentrators that reduce tensile strength and toughness, confirming incomplete interfacial compatibility at excessive wax loadings. This phenomenon may be attributed to starch retrogradation, as the incorporation of candelilla wax appeared to interfere with the starch plasticisation process, thereby enhancing phase separation between the TPCS matrix and the wax component. The effect was most evident at the highest wax concentration (10 wt %), as shown in Fig. 2e. Such discontinuities and gaps confirm that the hydrophilic starch matrix and the hydrophobic wax component could not form a fully continuous phase when the wax concentration was excessive. Moreover, the cavities observed in the 10 wt % sample suggest insufficient bonding between starch and candelilla wax, likely due to the limited miscibility and restricted mixing between the hydrophilic and hydrophobic phases (Oliveira *et al.* 2020). Consequently, discontinuities within the matrix caused by filler agglomeration at higher concentrations generated weak interstices.

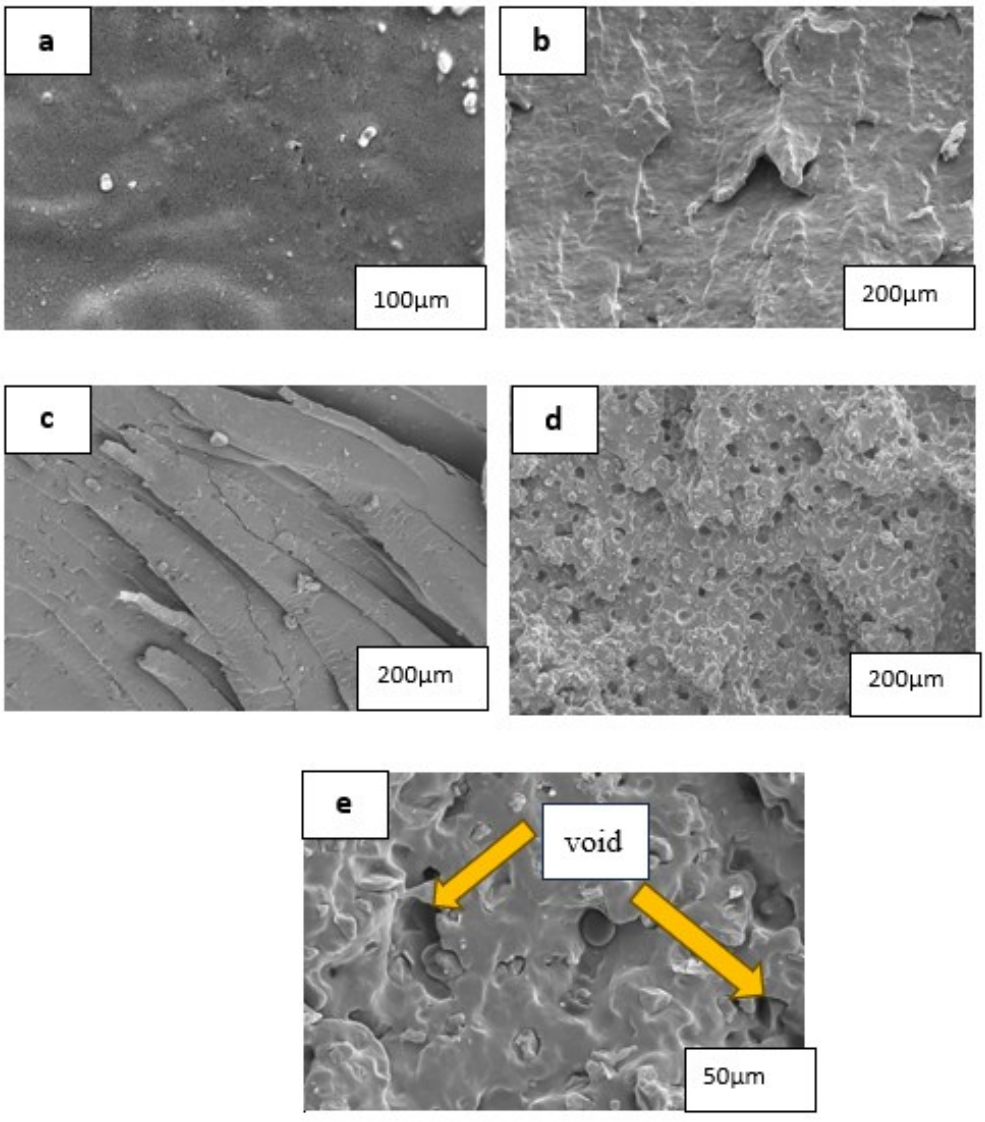


Fig. 2. Fracture surface SEM images (500X magnification) of TPCS Blended with various candelilla wax ratios: (a) 0 wt%, (b) 2.5 wt%, (c) 5 wt%, (d) 7.5 wt%, and (e) 10 wt %

These findings are consistent with previous studies on pea starch reinforced with carnauba wax (Mehyar *et al.* 2012) and edible sodium caseinate films reinforced with candelilla and carnauba waxes (Galus *et al.* 2020). The composite structure consequently became more inconsistent, forming weak interfacial regions and stress concentration points that facilitated crack initiation and propagation. This structural weakness contributed to a reduction in tensile strength, as shown in Fig. 5a (discussed later in this paper), where the composite with 10 wt % wax exhibited a marked decrease in tensile strength. Similar trends have been reported in cassava starch composites reinforced with carnauba wax (Ferreira *et al.* 2019) and palm wax (Hafila *et al.* 2022).

Thermogravimetric Analysis (TGA)

The heat resistance and decomposition patterns of TPCS/CW composites were examined through thermogravimetric (TG) profiles in Fig. 3a alongside their derivative thermogravimetric (DTG) profiles in Fig. 3b. In the case of native TPCS, the initial mass loss between 30 and 150 °C was attributed to moisture evaporation and was slightly lower for TPCS/CW composites due to the hydrophobic effect of the wax component, causing around 4.45% mass reduction. It is worth mentioning that across all TPCS/CW formulations, the mass loss at this initial phase was marginally lower, falling between 4.52 and 6.42%, which corresponded with the steeper decline seen in Fig. 3a. This reduction can be attributed to the lower water content within the TPCS/CW matrix, as candelilla wax imparts hydrophobicity to the composite. Similar behaviour was reported for beeswax-modified starch systems, where the inclusion of hydrophobic wax decreased moisture uptake and early-stage degradation (Zhang *et al.* 2018; Cheng *et al.* 2021). These findings are consistent with the present results, confirming that the incorporation of natural wax reduces water absorption and stabilises the TPCS matrix during initial thermal decomposition.

The last step is the mass reduction phase, which spans from 200 to 350 °C. This step was mainly linked to the starch's volatile constituents decomposition (Abral *et al.* 2019). Based on the current study's results, the native TPCS in this region suffered a weight loss of roughly about 74.3%, which included the loss of volatile products derived from carbohydrates and the removal of physically bound water, together with glycerol's decomposition (Cheng *et al.* 2021). However, clear peaks denoting the volatilisation of glycerol, such as the phase changes from melting or crystallisation, were not clearly seen (Travalini *et al.* 2019). Unlike the results for native TPCS, the second-stage weight loss for TPCS/CW composites was between 68.4 and 71.9%, which was slightly lower than what was obtained for native TPCS. This implies that candelilla wax could have some thermal stability and enhance the composite's strength, unlike the first stage.

Table 2 presents the results from the thermogravimetric analysis (TGA). Native TPCS started to degrade at an onset temperature of 212 °C, which indicates it had sustained thermal stability prior to undergoing mass loss. Travalini *et al.* (2019) documented a lower onset point of 124.5 °C for cassava starch films reinforced with bagasse fibres, whereas Kamaruddin *et al.* (2022) reported an onset temperature of 244 °C for TPCS blended with palm wax. Additionally, earlier research recorded maximum decomposition temperatures at 189 °C (Travalini *et al.* 2019) and 338 °C (Kamaruddin *et al.* 2022), whereas this study identified a peak decomposition temperature of 336 °C, situating it within the range of previous findings. These discrepancies were likely due to variations in experimental conditions, sample characteristics, and differences in TGA instrumentation.

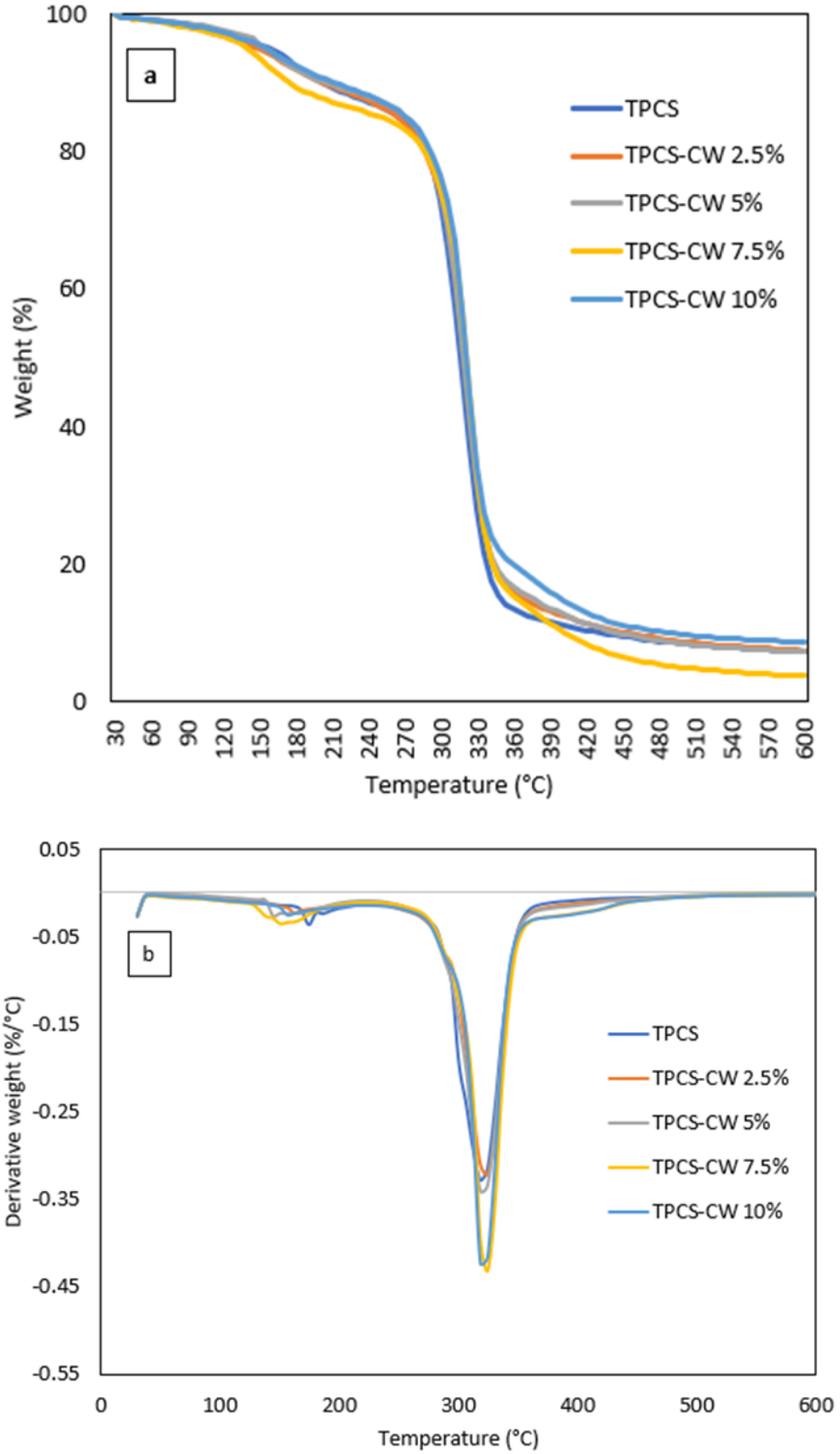


Fig. 3. Thermal degradation profiles of TPCS/CW composites, showing (a) TG and (b) DTG curves

Table 2. Thermal Decomposition Parameters (T_{on}, T_{max}, and Weight Loss) of TPCS/CW Composites

Sample Type	T_{on} (°C)	T_{max} (ºC)	Weight Loss at T_{max} (wt%)
TPCS	212.34	280.45	74.31
TPCS/CW 2.5	210.89	278.92	71.87
TPCS/CW 5	209.76	276.83	71.14
TPCS/CW 7.5	208.65	274.21	69.61
TPCS/CW 10	207.48	273.10	68.39

As depicted in Fig. 3b, the DTG analysis demonstrated progressive changes with increasing candelilla wax content. The maximum rates of decomposition were recorded as follows: 0.32%/°C for both neat TPCS and the TPCS/CW (2.5 wt%) blend, 0.34%/°C for TPCS/CW (5 wt%), and 0.43%/°C for both 7.5 and 10 wt % candelilla wax composites. This trend suggests that the addition of candelilla wax influenced the thermal degradation pathway by altering the molecular packing and phase interactions within the TPCS matrix. A comparable tendency was reported by Zhang *et al.* (2018), where the incorporation of beeswax into gelatin-based films enhanced thermal stability compared with wax-free counterparts.

The final decomposition phase was likely associated with the breakdown of inorganic oxides that form as residual ash components during thermal analysis. Among all the tested formulations, the composite containing 10 wt % candelilla wax exhibited the highest residual mass upon completion of degradation, suggesting a slightly greater amount of thermally stable residue. However, all specimens displayed similar main decomposition temperatures, indicating that the addition of candelilla wax mainly enhanced char formation rather than altering the overall thermal resistance of TPCS.

Tensile Test

The strength and flexibility characteristics of TPCS blended with different proportions of candelilla wax were assessed through tensile tests conducted at ambient temperatures. The outcomes are summarised in Fig. 4, detailing (a) tensile strength, (b) tensile modulus (stiffness), and (c) elongation at break. The experimental data were statistically analysed using one-way analysis of variance (ANOVA) to evaluate the differences among sample groups. Duncan's multiple range test was applied to determine significant differences between means at a confidence level of $p < 0.05$. All statistical analyses were performed using IBM SPSS Statistics software (Version 25.0, IBM Corp., Armonk, NY, USA). The results demonstrated that increasing the candelilla wax content from 0 to 5 wt% led to substantial improvements in both the material's strength and stiffness. Initially, the neat TPCS exhibited a tensile strength of 2.14 MPa alongside a modulus of 0.188 GPa. However, as the candelilla wax proportion was raised from 0 to 5 wt%, the tensile strength advanced from 2.74 to 4.86 MPa, while the modulus experienced a remarkable increase from 0.188 to 1.344 GPa. These enhancements represented a 127% rise in tensile strength ($p < 0.05$) and an impressive 615% boost in stiffness ($p < 0.05$) compared to the unmodified TPCS. The enhancement in tensile strength and stiffness up to 5 wt % candelilla wax indicates that the wax behaved as an effective dispersed filler, reinforcing the starch matrix through physical interaction and uniform distribution. Beyond this concentration, agglomeration and phase separation reduce stress-transfer efficiency, resulting in a decline in strength.

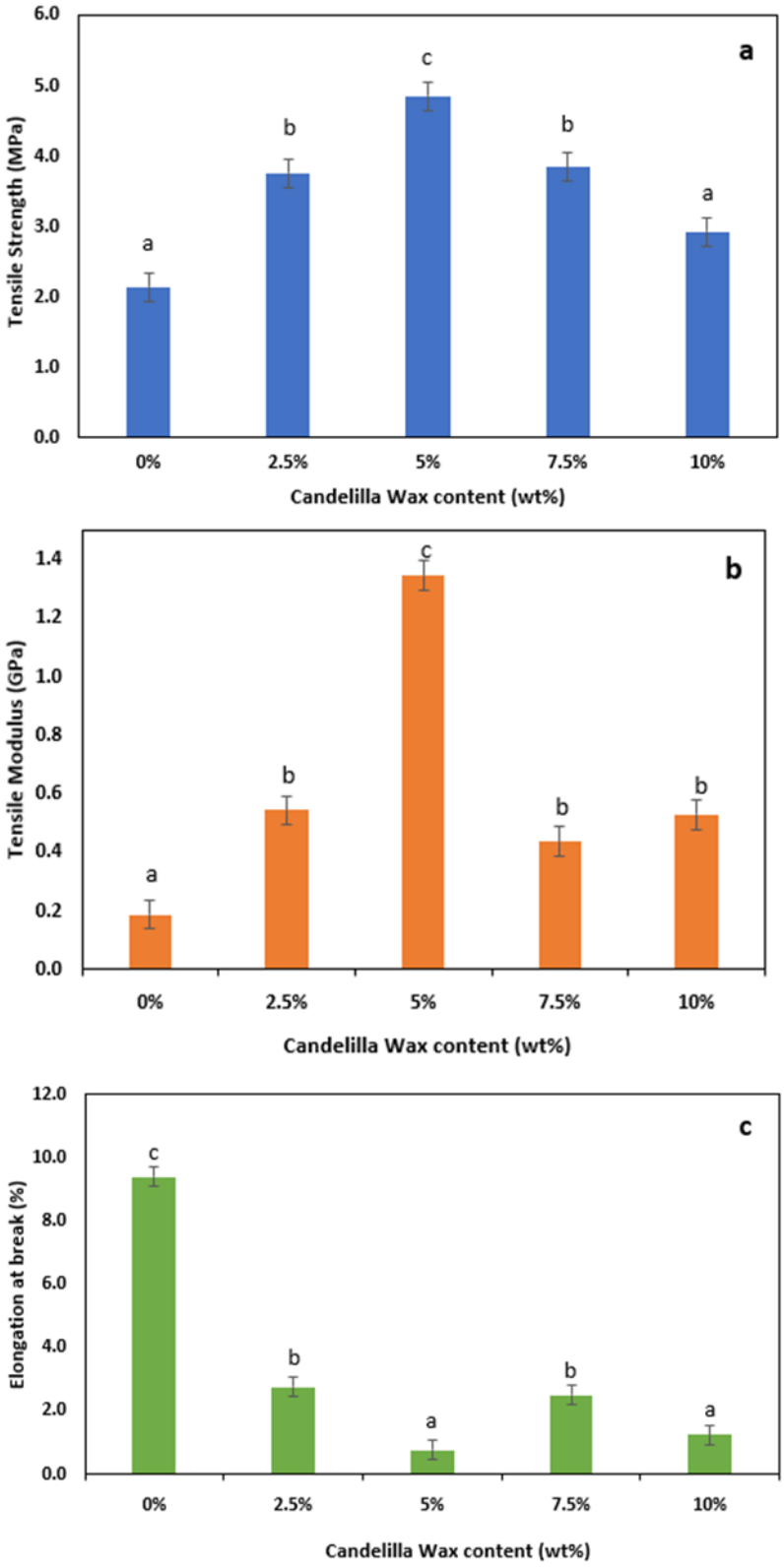


Fig. 4. TPCS/CW wax: (a) Tensile strength, (b) Tensile modulus (stiffness), and (c) Elongation at break

This notable improvement in mechanical performance was likely due to the excellent compatibility between the starch matrix and the candelilla wax, largely influenced by their similar chemical characteristics. With moderate levels of wax in the mixture, a more uniform dispersion and better phase interaction were observed, which contributed to strengthening the composite structure. A similar tendency was noted by Zhang *et al.* (2018), who found that incorporating natural waxes such as beeswax and carnauba wax improved the structural integrity and strength of biopolymer films through enhanced intermolecular interactions.

At wax concentrations above 7.5 wt %, the composites exhibited a plateau in elongation, while both tensile strength and modulus showed only marginal improvements. This behaviour could be attributed to restricted chain mobility within the matrix, as the embedded wax domains limit the movement of polymer chains and reduce their ability to deform under stress (Diyana *et al.* 2021). At higher wax loadings, the material gradually lost both strength and stretchability, likely due to phase discontinuities and interfacial defects generated by excessive filler content (Omar-Aziz *et al.* 2021; Kamaruddin *et al.* 2023). The presence of surplus candelilla wax may also hinder effective stress transfer by disrupting the continuity of the TPCS network, resulting in reduced interfacial adhesion and a more brittle composite structure.

In summary, increasing the candelilla wax content up to 5 wt% enhanced the mechanical strength of the composite. However, beyond 7.5 wt%, the material became harder, less flexible, and more brittle due to restricted molecular mobility and reduced structural cohesion.

Flexural Test

Figure 5a and 5b display the bending (flexural) strength and stiffness (modulus) of TPCS composites containing varying levels of candelilla wax, spanning from 0 up to 15 wt%. The inclusion of candelilla wax, especially within the range of 0 to 5 wt%, led to a remarkable 123% increase in flexural strength ($p < 0.05$). Likewise, the flexural modulus also showed a positive trend, increasing 110% ($p < 0.05$) with the addition of candelilla wax. Overall, the bending performance of the composites mirrored the behaviour observed in tensile tests, with clear enhancements in both flexural strength and modulus as more wax was incorporated into the TPCS matrix. The optimal bending strength and stiffness were achieved when the wax loading reached 5 wt%. However, as the wax concentration exceeded this level, a progressive decline in performance became apparent, which was consistent with the trends previously noted in tensile results. Increasing the filler content beyond 5 wt% led to a noticeable drop in flexural strength. For example, at wax loadings of 7.5 and 10 wt %, the flexural strength gradually decreased to 8.02 and 2.94 MPa, respectively. These results suggested that the greatest benefit to flexural strength for the composite occurred when the addition of candelilla wax reached 5 wt%.

Impact Test

Figure 6 illustrates the impact resistance performance of TPCS composite containing different amounts of candelilla wax. Notably, incorporating candelilla wax within the range of 0 to 5 wt% led to a substantial 79.5% improvement in impact strength ($p < 0.05$). This suggested that the TPCS and candelilla wax blended well, allowing the material to absorb impact energy more effectively, in line with the result from previous research (Kamaruddin *et al.* 2023).

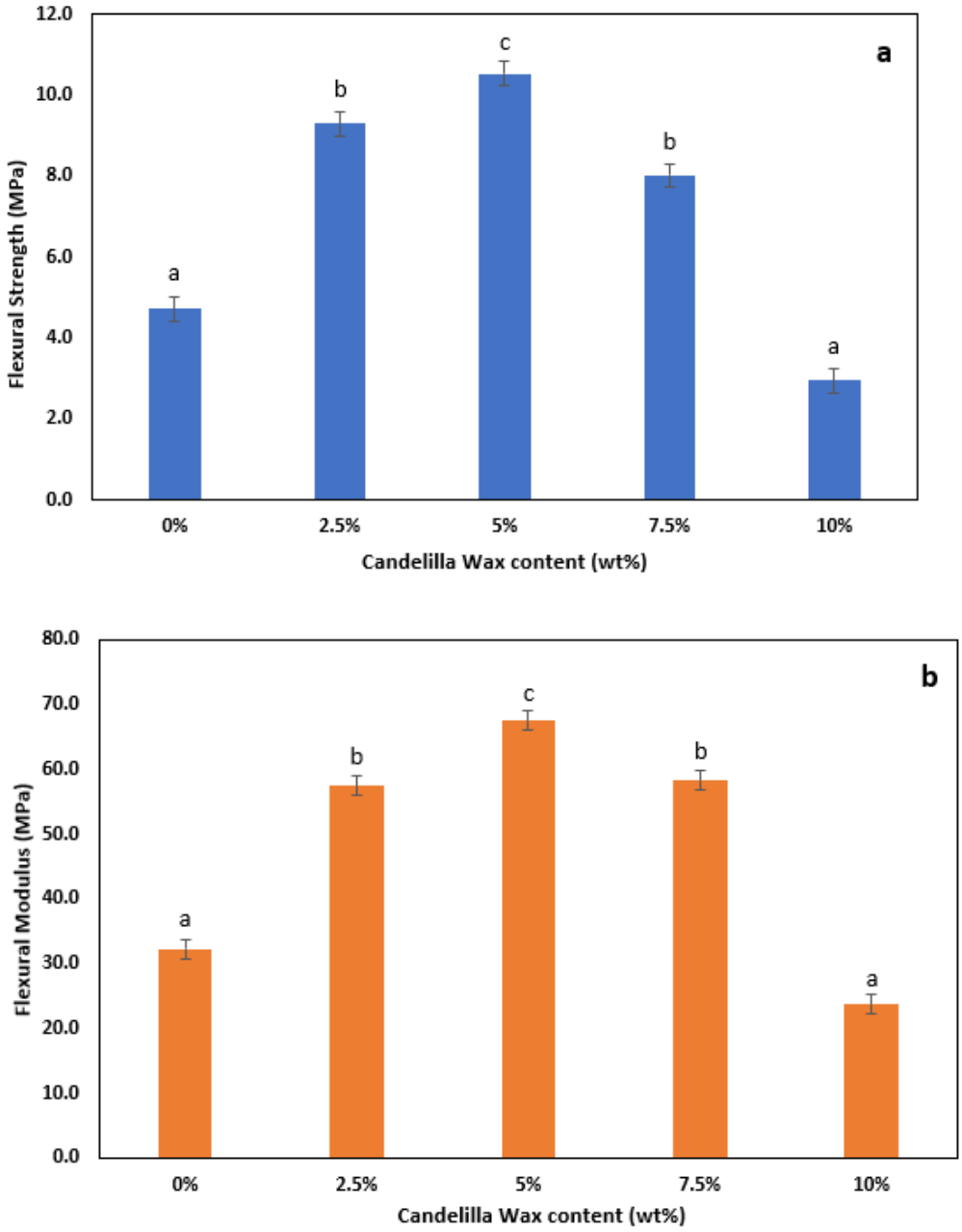


Fig. 5. TPCS/CW : (a) Flexural strength, and (b) Flexural modulus

As the concentration of candelilla wax increased past this point, the impact strength started to decline. These values illustrated that the impact strength at higher wax concentrations was lowered, which might account for the changes observed in the flexural modulus of the material. Because flexural modulus is related to stiffness and its resistance to bending, higher values are generally associated with a stronger structure, which might negatively affect impact strength (Jawaid *et al.* 2011; Hafila *et al.* 2022). However, when candelilla wax content was about 7.5 to 10 wt %, the material became more brittle, thereby reducing the ability to absorb impact energy, making it prone to fracture (Vilaseca *et al.* 2007).

The results of the candelilla wax content percentage were evaluated qualitatively through one-way analysis of variance (ANOVA) alongside the other outcomes related to tensile, flexural, and impact strength, compiled in Table 3. The analysis indicated p-values lower than the 0.05 limit, confirming that differences in tensile, flexural, and impact strength resulting from the various TPCS formulations were significantly different.

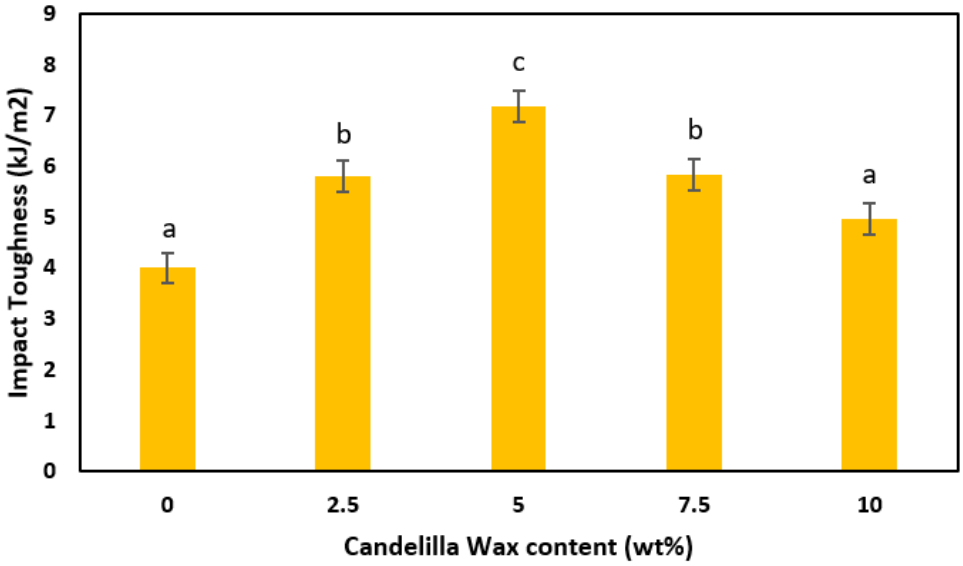


Fig. 6. Impact testing of TPCS/CW

Table 3. Statistical Analysis Summary of TPCS/Candelilla Wax Composites with ANOVA

Variables	df	Tensile strength	Tensile modulus	Flexural strength	Flexural modulus	Elongation at break	Impact strength
Mixture	4	0.001*	0.001*	0.001*	0.001*	0.001*	0.001*

*Note: Differences are statistically significant at $p < 0.05$

Water Absorption

Candelilla wax is renowned for its exceptionally strong water-repellent properties and is frequently utilised as a coating material to mitigate moisture absorption in starch-based materials due to its hydrophobic nature. Studies indicated that the incorporation of candelilla wax in starch-based biodegradable foams significantly enhanced their water resistance due to the hydrophobic barrier formed, which reduced water absorption and solubility (Alexander 2023). Furthermore, some research claimed that candelilla wax used in edible films and coatings performs remarkably well as a barrier against water vapour. It has been claimed that this efficiency is due to the hydrophobic character of the wax, which correlates with high amounts of long-chain hydrocarbons (Galus *et al.* 2020). Candelilla wax was analysed regarding potential usage as a constituent in TPCS composites based on its ability to absorb moisture. Figure 7 shows how TPCS/CW composites absorbed moisture over a 6-day duration at 75 ± 2% RH and 25 ± 2 °C. In general, all the samples exhibited a saturating trend due to extended sample storage. The remarkable observation was that the moisture absorption curve plateaued after six days, indicating that those samples reached a constant phase.

The moisture vapour barrier properties of TPCS/CW composites improved with increasing candelilla wax content, especially at higher concentrations, due to the formation of a continuous hydrophobic phase that limited water diffusion through the matrix. This demonstrated candelilla wax's contributory role as a hydrophobic barrier enhancer because the wax is assumed to have a low affinity to water. Its protective effect is due to the presence of long-chain fatty alcohols and hydrocarbons in its molecular structure (Galus *et al.* 2020).

The present results align with Zhang *et al.* (2018), who studied TPCS composites and noted that emulsions of beeswax increased the hydrophobic properties of the composites more than carnauba wax. Moreover, other studies have pointed out that carnauba wax was less effective than beeswax in reducing the hydrophilic tendencies of gelatin-based films (Diyana *et al.* 2021). This was made possible by the carnauba wax, which, along with the beeswax emulsions, enhanced the hydrophobic traits. Moreover, another study has also stated that palm wax increased the moisture barrier properties of TPCS composites (Hafila *et al.* 2022). All these findings have supported the notion that enhanced candelilla wax increases moisture resistance in the TPCS composites.

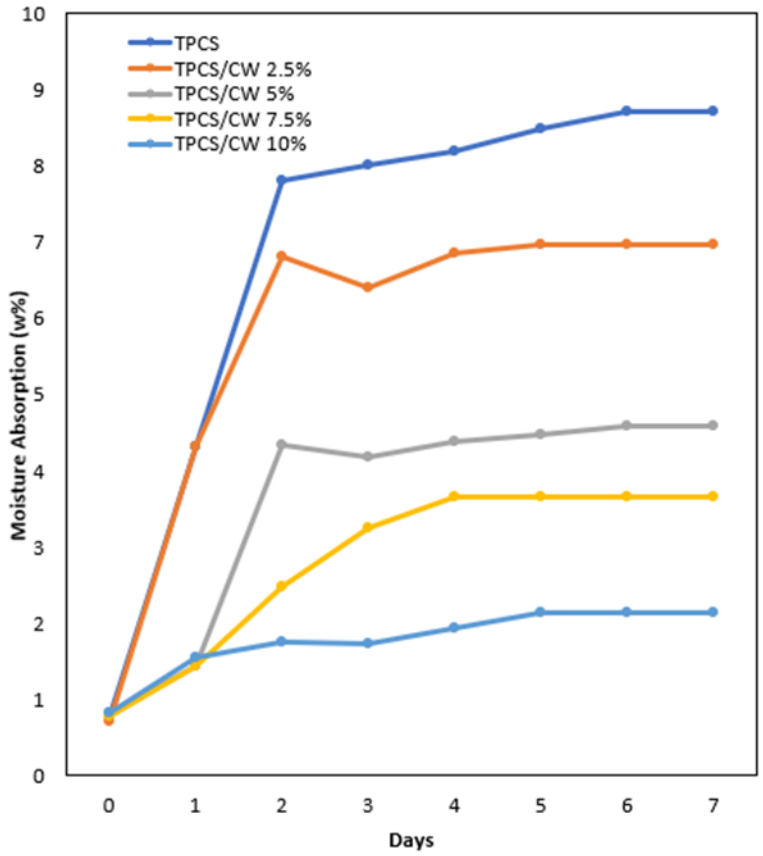


Fig. 7. Water absorption of TPCS/CW

Water Solubility

The solubility in water for candelilla wax/TPCS composites is illustrated in Fig. 8. The control sample from this study was noticeably more water soluble than the wax-infused composites, consistent with other studies. This was likely due to the very high number of hydroxyl groups within starch chains of the control sample that tend to form bonds with water molecules (Oliveira *et al.* 2020). In comparison, with increasing the TPCS/CW

composites demonstrated greater water resistance ($p < 0.05$), as the solubility decreased steadily from 30.3% for the neat TPCS sample to 24.0% for the composite containing 10 wt % wax. This trend reflects the progressive effect of wax incorporation in reducing the availability of hydroxyl sites for water interaction. This was largely due to the more hydrophobic candelilla wax, which is able to form a granular structure within the starch matrix. This, Galus *et al.* (2020) argued, could limit the rich supply of hydroxyl groups to water, thereby reducing the tendency of the material to absorb moisture and greatly increasing the starch composite's tensile strength. The same trend regarding soluble losses was also noted with TPCS composites containing palm wax, where increased palm wax content led to greater water insolubility (Hafila *et al.* 2022). Because candelilla wax is water-insoluble, its inclusion has promoted hydrophobic interactions with the starch matrix, thus decreasing the reactive sites that water molecules can utilise (Galus *et al.* 2020).

In addition, previous research has stated that increasing the hydrophobicity of biopolymer systems markedly reduces their ability to withstand water absorption. This is evident in the case of palm wax added to fish gelatin films, which decreased their water solubility from 36.06 to 15.19% (Syahida *et al.* 2020). Zhang *et al.* (2018) reported similar results with the addition of beeswax and carnauba wax to gelatin films, which decreased the films' gelatin solubility. The application of candelilla wax into the TPCS matrix not only improved water resistance but also assisted in the structural integrity of the composites in water.

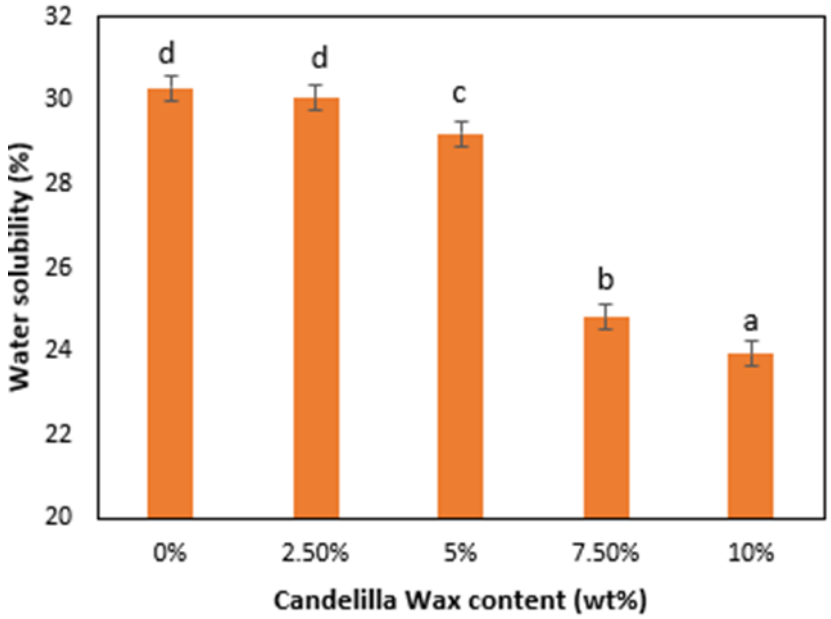


Fig. 8. Water solubility of TPCS/CW

Thickness Swelling

The composites of TPCS/CW with varying content of candelilla wax underwent immersion for 2 h, and their swelling behaviour is depicted in Fig. 9. The observed pattern in swelling behaviour was similar to that of moisture absorption. One-way ANOVA showed that the amount of candelilla wax significantly impacted the two composites' swelling properties ($p < 0.05$). Additionally, as the amount of candelilla wax changed, the amount of swelling was reduced ($p < 0.05$) from 39.5 to 25.8%. The rate of change implies that the composites became more dimensionally stable with more candelilla wax. The

stabilised composites were possibly due to the harsh hydrophobic character of candelilla, which contains long-chained waxy alkanes, fats, and fatty alcohols (Galus *et al.* 2020).

Furthermore, the lesser swelling of these samples is bound to be lipid-based due to the water repellent properties (Hromiš *et al.* 2015; Galus *et al.* 2020). This is caused due to the non-solubility of candelilla wax in the TPCS matrix, which also reduces the amount of swelling of the wax.

These findings correlated with previous works on the swelling behaviour of gelatinised fish and palm wax blended films (Syahida *et al.* 2020). In practical terms, a lower swelling index is advantageous for applications such as biodegradable packaging films, disposable food containers, and agricultural mulch sheets, where dimensional stability and moisture resistance are required.

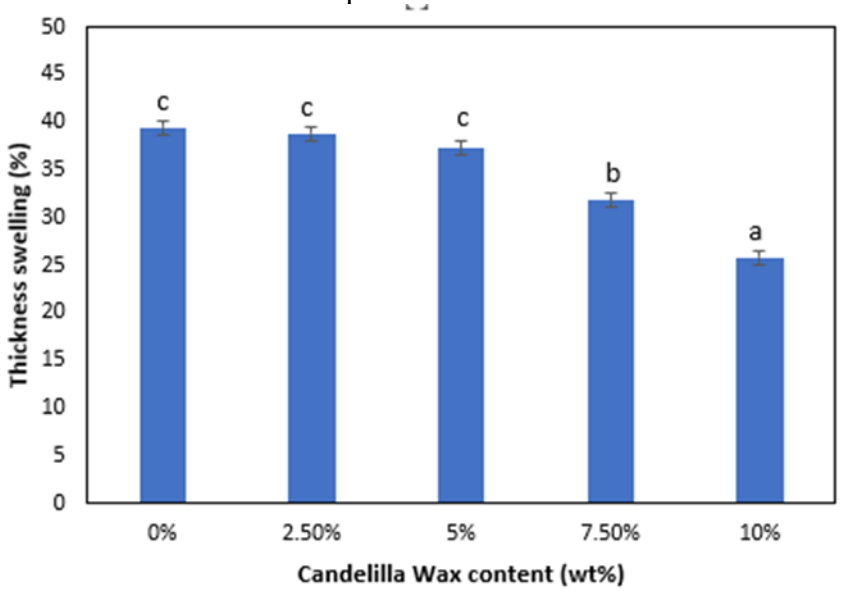


Fig. 9. Thickness swelling of TPCS/CW

CONCLUSIONS

1. The incorporation of candelilla wax into thermoplastic cassava starch (TPCS) significantly enhanced its mechanical strength, thermal stability, and water resistance, demonstrating its role as an effective bio-based modifier.
2. An optimal wax loading of 5 wt% provided the highest improvements in tensile and flexural properties, reflecting strong starch–wax interactions and enhanced stiffness.
3. The Fourier transform infrared (FT-IR) analysis confirmed hydrogen-bonding interactions between plasticized starch and candelilla wax, while SEM images showed more homogeneous structures at low-to-moderate CW loadings (2.5 to 5 wt %) but evident phase separation at higher contents (7.5 to 10 wt %).
4. The addition of candelilla wax improved thermal stability, with composites retaining more char residue at 600 °C, and drastically reduced moisture absorption, water solubility, and thickness swelling, especially at 10 wt % CW.

5. Overall, candelilla wax offers a sustainable route to overcome the inherent limitations of TPCS (low strength and high hydrophilicity), expanding its potential for biodegradable packaging, agricultural mulch films, and disposable food containers that require moderate moisture resistance. Future studies may consider incorporating mild compatibilisers such as polysorbate 20 or 80 to further enhance the interfacial adhesion between the hydrophilic starch matrix and the hydrophobic candelilla wax phase.

ACKNOWLEDGMENTS

The authors acknowledge with appreciation the support given by Universiti Teknikal Malaysia Melaka and the Ministry of Higher Education Malaysia in the form of an FRGS/1/2023/STG05/UTEM/02/2 grant as well as the technical and financial support from Universiti Malaysia Sabah.

The authors acknowledge the use of ChatGPT (OpenAI) to assist in improving the clarity, grammar, and readability of the English text. The scientific content, data analyses, results, and conclusions were generated, verified, and finalised solely by the authors.

REFERENCES CITED

Abral, H., Basri, A., Muhammad, F., Fernando, Y., Hafizulhaq, F., Mahardika, M., Sugiarti, E., Sapuan, S. M., Ilyas, R. A., and Stephane, I. (2019). "A simple method for improving the properties of the sago starch films prepared by using ultrasonication treatment," *Food Hydrocoll.* 93, 276-283. DOI: 10.1016/j.foodhyd.2019.02.012

Aguirre-Joya, J. A., Cerqueira, M. A., Ventura-Sobrevilla, J., Aguilar-Gonzalez, M. A., Carbó-Argibay, E., Castro, L. P., and Aguilar, C. N. (2019). "Candelilla wax-based coatings and films: Functional and physicochemical characterization," *Food Bioprocess Technol.* 12(10), 1787-1797. DOI: 10.1007/s11947-019-02339-2

Aguirre-Joya, J. A., Ventura-Sobrevilla, J., Martínez-Vazquez, G., Ruelas-Chacón, X., Rojas, R., Rodríguez-Herrera, R., and Aguilar, C. N. (2017). "Effects of a natural bioactive coating on the quality and shelf life prolongation at different storage conditions of avocado (*Persea americana* Mill.) cv. Hass," *Food Packag. Shelf Life* 14(Part B), 102-107. DOI: 10.1016/j.fpsl.2017.09.003

Alexander, I. (2023). "The effect of different natural waxes to hydrophobic properties of starch-based biodegradable foams," *J. Environ. Sci. Sustain. Dev.* 6(1), 86-101. DOI: 10.7454/jessd.v6i1.1199

Aranda-Ledesma, N. E., Bautista-Hernández, I., Rojas, R., Aguilar-Zárate, P., Medina-Herrera, N. P., Castro-López, C., and Martínez-Ávila, G. C. G. (2022). "Candelilla wax: Prospective suitable applications within the food field," *LWT* 159, article 113170. DOI: 10.1016/j.lwt.2022.113170

Azlin, M. N., Sapuan, S. M., Zainudin, E. S., and Zuhri, M. Y. (2020). "Natural fibre reinforced polyester composite: A review," *Postgraduate Seminar on Natural Fibre Reinforced Polymer Composites* 7(11), 3-7.

Babaee, M., Jonoobi, M., Hamzeh, Y., and Ashori, A. (2015). "Biodegradability and mechanical properties of reinforced starch nanocomposites using cellulose nanofibers," *Carbohydrate Polymers* 132, 1-8. DOI: 10.1016/j.carbpol.2015.06.043

Bergo, P., Sobral, P. J. A., and Prison, J. M. (2010). "Effect of glycerol on physical properties of cassava starch films," *Journal of Food Processing and Preservation* 34(S2), 401-410. DOI: 10.1111/j.1745-4549.2008.00282.x

Bucio, A., Moreno-Tovar, R., Bucio, L., Espinosa-Dávila, J., and Anguebes-Franceschi, F. (2021). "Characterization of beeswax, candelilla wax and paraffin wax for coating cheeses," *Coatings* 11(3), article 261. DOI: 10.3390/coatings11030261

Bušić, A., Mardetko, N., Kundas, S., Morzak, G., Belskaya, H., Šantek, M. I., Komes, D., Novak, S., & Šantek, B. (2018). "Bioethanol production from renewable raw materials and its separation and purification: A review". *Food Technology and Biotechnology*, *56*(3), 289–311. DOI:10.17113/ftb.56.03.18.5546

Campos, A., Sena Neto, A. R., Rodrigues, V. B., Luchesi, B. R., Mattoso, L. H. C., and Marconcini, J. M. (2018). "Effect of raw and chemically treated oil palm mesocarp fibers on thermoplastic cassava starch properties," *Industrial Crops and Products* 124, 149-154. DOI: 10.1016/j.indcrop.2018.07.075

Cheng, Y., Wang, W., Zhang, R., Zhai, X., and Hou, H. (2021). "Effect of gelatin bloom values on the physicochemical properties of starch/gelatin–beeswax composite films fabricated by extrusion blowing," *Food Hydrocolloids* 113, article 106466. DOI: 10.1016/j.foodhyd.2020.106466

Chiumarelli, M., and Hubinger, M. D. (2014). "Evaluation of edible films and coatings formulated with cassava starch, glycerol, carnauba wax and stearic acid," *Food Hydrocolloids* 38, 20-27. DOI: 10.1016/j.foodhyd.2013.11.013

Chiumarelli, M., Pereira, L. M., Ferrari, C. C., Sarantópoulos, C. I. G. L., and Hubinger, M. D. (2010). "Cassava starch coating and citric acid to preserve quality parameters of fresh-cut 'Tommy Atkins' mango," *Journal of Food Science* 75(5), E297-E304. DOI: 10.1111/j.1750-3841.2010.01636.x

Curvelo, A. A. S., De Carvalho, A. J. F., and Agnelli, J. A. M. (2001). "Thermoplastic starch-cellulosic fibers composites: Preliminary results," *Carbohydrate Polymers* 45(2), 183-188. DOI: 10.1016/S0144-8617(00)00314-3

Diyana, Z. N., Jumaidin, R., Selamat, M. Z., and Suan, M. S. M. (2021). "Thermoplastic starch/beeswax blend: Characterization on thermal mechanical and moisture absorption properties," *International Journal of Biological Macromolecules* 190, 224-232. DOI: 10.1016/j.ijbiomac.2021.08.201

Ferreira, E. A., De Abreu, J. G., Martinez, J. C., Amorim, R. S. S., Neto, A. B., Cabral, C. E. A., Dos Santos Braz, T. G., Silva, C. A., and Ferreira, D. P. (2019). "Productivity and nutritional value of elephant grass BRS Canará forage," *Semina: Ciências Agrárias* 40(6), 2705-2718. DOI: 10.5433/1679-0359.2019v40n6p2705

Florencia, V., López, O. V., and García, M. A. (2020). "Exploitation of by-products from cassava and ahipa starch extraction as filler of thermoplastic corn starch," *Composites Part B: Engineering* 182, article 107653. DOI: 10.1016/j.compositesb.2019.107653

Galus, S., Gaouditz, M., Kowalska, H., and Debeaufort, F. (2020). "Effects of candelilla and carnauba wax incorporation on the functional properties of edible sodium caseinate films," *International Journal of Molecular Sciences* 21(24), article 9349. DOI: 10.3390/ijms21249349

Hafila, K. Z., Jumaidin, R., Ilyas, R. A., Selamat, M. Z., and Yusof, F. A. M. (2022). "Effect of palm wax on the mechanical, thermal, and moisture absorption properties of thermoplastic cassava starch composites," *International Journal of Biological Macromolecules* 194, 851-860. DOI: 10.1016/j.ijbiomac.2021.11.139

Hazrati, K. Z., Sapuan, S. M., Zuhri, M. Y. M., and Jumaidin, R. (2021a). "Extraction and characterization of potential biodegradable materials based on *Dioscorea hispida* tubers," *Polymers* 13(4), article 584. DOI: 10.3390/polym13040584

Hazrati, K. Z., Sapuan, S. M., Zuhri, M. Y. M., and Jumaidin, R. (2021b). "Effect of plasticizers on physical, thermal, and tensile properties of thermoplastic films based on *Dioscorea hispida* starch," *International Journal of Biological Macromolecules* 185, 219-228. DOI: 10.1016/j.ijbiomac.2021.06.099

Hromiš, N. M., Lazić, V. L., Markov, S. L., Vaštag, Ž. G., Popović, S. Z., Šuput, D. Z., Džinić, N. R., Velićanski, A. S., and Popović, L. M. (2015). "Optimization of chitosan biofilm properties by addition of caraway essential oil and beeswax," *Journal of Food Engineering* 158, 86-93. DOI: 10.1016/j.jfoodeng.2015.01.001

Hulleman, S. H. D., Janssen, F. H. P., and Feil, H. (1998). "The role of water during plasticization of native starches," *Polymer* 39(10), 2043-2048. DOI: 10.1016/S0032-3861(97)00301-7

Jawaid, M., Abdul Khalil, H. P. S., Noorunnisa Khanam, P., and Abu Bakar, A. (2011). "Hybrid composites made from oil palm empty fruit bunches/jute fibres: Water absorption, thickness swelling and density behaviours," *Journal of Polymers and the Environment* 19(1), 106-109. DOI: 10.1007/s10924-010-0203-2

Jumaidin, R., Khiruddin, M. A. A., Asyul Sutan Saidi, Z., Salit, M. S., and Ilyas, R. A. (2020). "Effect of cogon grass fibre on the thermal, mechanical and biodegradation properties of thermoplastic cassava starch biocomposite," *International Journal of Biological Macromolecules* 146, 746-755. DOI: 10.1016/j.ijbiomac.2019.11.011

Jumaidin, R., Sapuan, S. M., Jawaid, M., Ishak, M. R., and Sahari, J. (2017). "Effect of seaweed on mechanical, thermal, and biodegradation properties of thermoplastic sugar palm starch/agar composites," *International Journal of Biological Macromolecules* 99, 265-273. DOI: 10.1016/j.ijbiomac.2017.02.092

Kaisangsri, N., Kerdchoechuen, O., and Laohakunjit, N. (2012). "Biodegradable foam tray from cassava starch blended with natural fiber and chitosan," *Industrial Crops and Products* 37(1), 542-546. DOI: 10.1016/j.indcrop.2011.07.034

Kamaruddin, Z. H., Jumaidin, R., Ilyas, R. A., Selamat, M. Z., Alamjuri, R. H., and Yusof, F. A. M. (2022). "Biocomposite of cassava starch-*Cymbopogan citratus* fibre: Mechanical, thermal and biodegradation properties," *Polymers* 14(3), article 514. DOI: 10.3390/polym14030514

Kamaruddin, Z. H., Jumaidin, R., Kamaruddin, Z. H., Asyraf, M. R. M., Razman, M. R., and Khan, T. (2023). "Effect of *Cymbopogan citratus* fibre on physical and impact properties of thermoplastic cassava starch/palm wax composites," *Polymers* 15(10), article 2364. DOI: 10.3390/polym15102364

Karimi, S., Dufresne, A., Md. Tahir, P., Karimi, A., and Abdulkhani, A. (2014). "Biodegradable starch-based composites: Effect of micro and nanoreinforcements on composite properties," *Journal of Materials Science* 49(13), 4513-4521. DOI: 10.1007/s10853-014-8151-1

Karnwal, A., Rauf, A., Jassim, A. Y., Selvaraj, M., Al-Tawaha, A. R. M. S., Kashyap, P., and Malik, T. (2025). "Advanced starch-based films for food packaging: Innovations

in sustainability and functional properties," *Food Chemistry X* 2025, article 102662. DOI: 10.1016/j.fochx.2025.102662

Kowalczyk, D., and Biendl, M. (2016). "Physicochemical and antioxidant properties of biopolymer/candelilla wax emulsion films containing hop extract: A comparative study," *Food Hydrocolloids* 60, 384-392. DOI: 10.1016/j.foodhyd.2016.04.010

Kowalczyk, D., Kordowska-Wiater, M., Zięba, E., and Baraniak, B. (2017). "Effect of carboxymethylcellulose/candelilla wax coating containing potassium sorbate on microbiological and physicochemical attributes of pears," *Scientia Horticulturae* 218, 326-333. DOI: 10.1016/j.scienta.2017.02.040

De León-Zapata, M. A., Ventura-Sobrevilla, J. M., Salinas-Jasso, T. A., Flores-Gallegos, A. C., Rodríguez-Herrera, R., Pastrana-Castro, L., Rua-Rodríguez, M. L., and Aguilar, C. N. (2018). "Changes of the shelf life of candelilla wax/tarbush bioactive-based nanocoated apples at industrial level conditions," *Scientia Horticulturae* 231, 43-48. DOI: 10.1016/j.scienta.2017.12.005

Li, H., Zhao, Y., Deng, J., Xie, J., Zhou, W., Liu, Y., Li, L., Xia, F., and Qi, R. (2024). "Optimizing coal wettability via anionic surfactants: An integrated experimental and molecular dynamics simulation investigation," *Geofluids* 2024(1), article 9112308. DOI: 10.1155/gfl/9112308

Li, J., Luo, X., Lin, X., and Zhou, Y. (2013). "Comparative study on the blends of PBS/thermoplastic starch prepared from waxy and normal corn starches," *Starch/Stärke* 65(9-10), 831-839. DOI: 10.1002/star.201200260

Ma, X., Yu, J., and Kennedy, J. F. (2005). "Studies on the properties of natural fiber-reinforced thermoplastic starch composites," *Carbohydrate Polymers* 62(1), 19-24. DOI: 10.1016/j.carbpol.2005.07.015

Mehyar, G. F., Al-Ismail, K., Han, J. H., and Chee, G. W. (2012). "Characterization of edible coatings consisting of pea starch, whey protein isolate, and carnauba wax and their effects on oil rancidity and sensory properties of walnuts and pine nuts," *Journal of Food Science* 77(2), E52-E59. DOI: 10.1111/j.1750-3841.2011.02559.x

Muñoz-Gimena, P. F., Oliver-Cuenca, V., Peponi, L., and López, D. (2023). "A review on reinforcements and additives in starch-based composites for food packaging," *Polymers* 15(13), article 2972. DOI: 10.3390/polym15132972

Oliveira Filho, J. G. de, Bezerra, C. C. de O. N., Albiero, B. R., Oldoni, F. C. A., Miranda, M., Egea, M. B., Azeredo, H. M. C. de, and Ferreira, M. D. (2020). "New approach in the development of edible films: The use of carnauba wax micro- or nanoemulsions in arrowroot starch-based films," *Food Packaging and Shelf Life* 26, article 100589. DOI: 10.1016/j.fpsl.2020.100589

Omar-Aziz, M., Khodaiyan, F., Yarmand, M. S., Mousavi, M., Gharaghani, M., Kennedy, J. F., and Hosseini, S. S. (2021). "Combined effects of octenylsuccination and beeswax on pullulan films: Water-resistant and mechanical properties," *Carbohydrate Polymers* 255, article 117471. DOI: 10.1016/j.carbpol.2020.117471

Öner, E. T., Oliver, S. G., & Kirdar, B. (2005). "Production of ethanol from starch by respiration-deficient recombinant Saccharomyces cerevisiae*"*. *Applied and Environmental Microbiology*, *71*(10), 6443–6445. https://doi.org/10.1128/AEM.71.10.6443-6445.2005

Prachayawarakorn, J., Chaiwatyothin, S., Mueangta, S., and Hanchana, A. (2013). "Effect of jute and kapok fibers on properties of thermoplastic cassava starch composites," *Materials & Design* 47, 309-315. DOI: 10.1016/j.matdes.2012.12.012

Prachayawarakorn, J., Limsiriwong, N., Kongjindamunee, R., and Surakit, S. (2012). "Effect of agar and cotton fiber on properties of thermoplastic waxy rice starch composites," *Journal of Polymers and the Environment* 20(1), 88-95. DOI: 10.1007/s10924-011-0371-8

Prachayawarakorn, J., Sangnitidej, P., and Boonpasith, P. (2010). "Properties of thermoplastic rice starch composites reinforced by cotton fiber or low-density polyethylene," *Carbohydrate Polymers* 81(2), 425-433. DOI: 10.1016/j.carbpol.2010.02.041

Reddy, K. O., Maheswari, C. U., Dhlamini, M. S., Mothudi, B. M., Kommula, V. P., Zhang, J., Zhang, J., and Rajulu, A. V. (2018). "Extraction and characterization of cellulose single fibers from native African napier grass," *Carbohydrate Polymers* 188, 85-91. DOI: 10.1016/j.carbpol.2018.01.110

Rocha, T. S., Felizardo, S. G., Jane, J. L., and Franco, C. M. L. (2012). "Effect of annealing on the semicrystalline structure of normal and waxy corn starches," *Food Hydrocolloids* 29(1), 93-99. DOI: 10.1016/j.foodhyd.2012.02.003

Rodrigues, D. C., Caceres, C. A., Ribeiro, H. L., de Abreu, R. F. A., Cunha, A. P., and Azeredo, H. M. C. (2014). "Influence of cassava starch and carnauba wax on physical properties of cashew tree gum-based films," *Food Hydrocolloids* 38, 147-151. DOI: 10.1016/j.foodhyd.2013.12.010

Rosa, R. C. R. S., and Andrade, C. T. (2004). "Effect of chitin addition on injection-molded thermoplastic corn starch," *Journal of Applied Polymer Science* 92(4), 2706-2713. DOI: 10.1002/app.20292

Sanyang, M. L., Sapuan, S. M., Jawaid, M., Ishak, M. R., and Sahari, J. (2016). "Development and characterization of sugar palm starch and poly(lactic acid) bilayer films," *Carbohydrate Polymers* 146, 36-45. DOI: 10.1016/j.carbpol.2016.03.051

Silva, F. de A., Chawla, N., and de Toledo Filho, R. D. (2008). "Tensile behavior of high performance natural (sisal) fibers," *Composites Science and Technology* 68(15-16), 3438-3443. DOI: 10.1016/j.compscitech.2008.10.001

Syahida, N., Fitry, I., Zuriyati, A., and Hanani, N. (2020). "Effects of palm wax on the physical, mechanical and water barrier properties of fish gelatin films for food packaging application," *Food Packaging and Shelf Life* 23, article 100437. DOI: 10.1016/j.fpsl.2019.100437

Travalini, A. P., Lamsal, B., Magalhães, W. L. E., and Demiate, I. M. (2019). "Cassava starch films reinforced with lignocellulose nanofibers from cassava bagasse," *International Journal of Biological Macromolecules* 139, 1151-1161. DOI: 10.1016/j.ijbiomac.2019.08.115

Vilaseca, F., Mendez, J. A., Pèlach, A., Llop, M., Cañigueral, N., Gironès, J., Turon, X., and Mutjé, P. (2007). "Composite materials derived from biodegradable starch polymer and jute strands," *Process Biochemistry* 42(3), 329-334. DOI: 10.1016/j.procbio.2006.09.004

Wu, Y., Geng, F., Chang, P. R., Yu, J., and Ma, X. (2009). "Effect of agar on the microstructure and performance of potato starch film," *Carbohydrate Polymers* 76(2), 299-304. DOI: 10.1016/j.carbpol.2008.10.031

Zhang, Y., Simpson, B. K., and Dumont, M. J. (2018). "Effect of beeswax and carnauba wax addition on properties of gelatin films: A comparative study," *Food Bioscience* 26, 88-95. DOI: 10.1016/j.fbio.2018.09.011

Article submitted: August 18, 2025; Peer review completed: October 5, 2025; Revised version received: October 22, 2025; Accepted: October 23, 2025; Published: November 2, 2025.
DOI: 10.15376/biores.20.4.11074-11097