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Phenolic compounds-containing fruit peel extracts of *Garcinia humilis* exhibit anti-melanoma activity

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ABSTRACT

There has been considerable interest in commercializing fruits of *Garcinia humilis* due to its pleasant taste and nutritional quality. However, the industrial processing of the fruit generates the peel as waste and this study focussed on upcycling of the peel for its anti-melanoma activity at different extraction conditions (24 °C and 37 °C for 24 h, and 70 °C for 1 h). Although the extraction yield was highest at 70 °C, the phenolics and flavonoids extracted, and anti-cancer activity was highest when extracted at 37 °C. Polyphenolic compounds including epicatechin, procyanidins, mangostanol, and gamma-mangostin were tentatively identified based on LC-Orbitrap MS/MS analysis. These extracts showed dose-dependent anti-melanoma activity on MM418C1 (BRAF mutated oncogene) and D24 (wild type BRAF oncogene) cells with IC₅₀ in the range of 130–200 µg/mL and 122–176 µg/mL. Furthermore, the extracts had little effect on control fibroblast cells, indicating selective activity on cancer cells, and displayed morphological changes indicative of apoptosis. Although these extracts were 10 fold less effective compared to anti-cancer drug controls, cisplatin and vemurafenib, the moderate anti-cancer activity of *Garcinia* peel provides future opportunities for upcycling of this discarded waste as a functional food ingredient.

CRedit author statement

Mahesha M. Poojary: Conceptualization; Methodology; Validation; Visualization; Formal analysis; Investigation; Writing - Original Draft; Writing - Review & Editing; Funding acquisition. **Thi Dao Nguyen:** Methodology; Formal analysis. **Chaitali Dekiwadia:** Methodology; Formal analysis. **Daniel A. Dias:** Conceptualization; Writing - Review & Editing; Resources; Visualization; Supervision. **Tien Huynh:** Conceptualization; Writing - Review & Editing; Visualization; Resources; Supervision.

1. Introduction

Garcinia humilis (Vahl) C. D. Adams (Clusiaceae) is a tree, commonly

known as “achachairú” or “achacha”, produces small edible fruits, similar to mangosteen (*Garcinia mangostana*), having a whitish, slightly acidic to sweet pulp and orange-yellow peel. The fruit comprised 40% pulp, 47% peel and 12% seed. It predominately contains water (83.9%) and other major constituents including carbohydrates (14.2%), fats (0.5%), proteins (0.4%) and fibres (0.6%) (Lim, 2012). The plant was introduced into Australia in 2012 and fruits were commercially available in 2018 (John et al., 2018). The fruits are sold whole or processed as pulp to produce juice, jams and ice creams. The peel can be used to prepare tea infusions in households, however, in most cases, it is discarded as waste. This provides an opportunity to upcycle the waste as an alternative, sustainable and low-cost source for obtaining bioactive compounds but the scientific justification is limited.

Fruits from the *Garcinia* genus, mainly *G. mangostana* and *G. indica*,

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are known to contain several bioactive classes including phenolic acids, xanthenes, tannins, anthocyanins (Nayak et al., 2010; Panthong et al., 2006; Zadernowski et al., 2009). Previous studies have shown that peel extracts of mangosteen fruits exhibited antioxidant, antimicrobial, antiallergic, anti-inflammatory, anti-hypertensive, and antiviral activities (Aukkanimart et al., 2015). The bioactive phenolic compounds and xanthenes present in the peel possess anticancer activity (Yang et al., 2017). In the case of *G. humilis*, the juice of the peel assisted in the suppression of appetite and showed skin nourishment properties (Lim, 2012). The peel of *G. humilis* improved cardiovascular function in rats (John et al., 2018) and possess antioxidant activity (Tome et al., 2019). The seed and stem demonstrated gastroprotective and antiproliferative activities (Mariano et al., 2016; 2016; Niero et al., 2012). These studies suggest that xanthenes are the most common class of bioactive compounds present in *G. humilis*.

Cancer is a major chronic disease responsible for the high mortality rate worldwide. Melanoma is one of the most invasive forms of skin cancer (Kanavy & Gerstenblith, 2011), and ~325,000 cases were estimated worldwide in 2020 (Arnold et al., 2022). DNA damage due to UV light exposure is the major reason for melanoma (Sun et al., 2020). Many studies have demonstrated that plant extracts with bioactive molecules could reduce the melanoma risk and act as chemotherapy agents by modulating carcinogenic pathways (Albuquerque et al., 2018). BRAF mutations occur in ~50% of melanomas via the MAPK signalling pathway and targeting intermediates of this pathway have been the inspiration of anti-melanoma drugs.

Although there is an increasing interest amongst industries and consumers for *G. humilis* fruits, the phytochemical and biological activity of this exotic fruit is limited (John et al., 2018; Tome et al., 2019) and anticancer activities of the peel are unknown. The industry is still underdeveloped as a niche specialty product that has limited seasonal availability. Since the peel constitutes more than half the biomass of the fruit, its usage would reduce landfill waste and create additional resources for farmers as part of our global sustainable goals for a circular agricultural economy. This investigation aimed to (1) extract and characterize phytochemicals (phenolics, flavonoids and anthocyanins) from the peel of *G. humilis* using conventional solvent extraction, (2) demonstrate its potential *in vitro* anticancer activities on human melanoma cells, and (3) evaluate the morphological changes induced by the extracts on the cellular ultrastructure.

2. Materials and methods

2.1. Plant samples and chemicals

$$\text{Anthocyanin (cyanidin - 3 - glucoside equivalents, mg/L)} = \frac{A \times MW \times DF \times 1000}{\epsilon \times l} \quad \text{Eq. 1}$$

Whole fruits of *G. humilis* were purchased from Achacha farms, cultivated in Palm Creek Plantation, Giru area (Australia), harvested late autumn and air posted to RMIT Laboratory in Melbourne. The fresh peel was separated from the fruit, rinsed in Milli-Q water and lyophilized for 48 h using a freeze dryer (Martin Christ, John Morris, Australia). The dried samples were finely powdered and stored in closed containers at -20 °C. Gallic acid (≥99%), quercetin (≥98%), dimethyl sulfoxide (DMSO, ≥99.7%), methanol (≥99.9%), acetonitrile (≥99.9%) and formic acid (≥98%) were purchased from Sigma Aldrich, Australia.

2.2. Extraction

Dried samples (5 g each) were extracted using acidified methanol (0.01% v/v HCl in methanol, 100 mL) in a round bottom flask under constant agitation (500 rpm) at three different levels of temperature and time: (i) 24 °C for 24 h (ii) 37 °C for 24 h and (iii) 70 °C for 1 h. Each extract was filtered through Whatman No. 1 filter under vacuum and the filtrate was concentrated under reduced pressure at 37 °C using a rotary evaporator. The resultant extracts were stored at -20 °C.

2.3. Total phenolics

The total phenolic content of the extracts was determined using the Folin–Ciocalteu assay, on 96-well microplates, according to (Sánchez-Rangel et al., 2013) with modifications. Briefly, 15 µL of the extract was diluted with 240 µL of distilled water and mixed with 15 µL of 1:10 diluted Folin–Ciocalteu reagent. The mixture was incubated at room temperature for 3 min and 30 µL of 10% sodium carbonate was added. The resultant solution was incubated in darkness for 1.5 h at room temperature. The absorbance was measured at 765 nm using a multi-well plate reader (CLARIOstar, BMG Labtech, Germany). The analysis was performed in triplicates and the results were expressed as mg Gallic Acid Equivalent/g Dry Matter (mg GAE/g DM).

2.4. Total flavonoids

The total flavonoid content was determined based on the aluminium complexation reaction (Pekal & Pyrzynska, 2014) on 96-well microplates. Briefly, diluted extracts (40 µL) were incubated with water (100 µL) and 2% w/v aluminium chloride solution (20 µL) for 10 min at room temperature (n = 3). Absorbencies of the resultant solutions were measured at 415 nm against the blank composed of Milli Q water instead of aluminium chloride. Quercetin was used as a standard (10–100 µg/mL) and the results were expressed as mg Quercetin Equivalent/g of Dry Matter (mg QE/g DM).

2.5. Total anthocyanins

The presence of total monomeric anthocyanins in the extract was determined based on the pH differential method by (Giusti et al., 2001). Briefly, the test extract was diluted with 25 mM potassium chloride buffer (pH 1.0) and 0.4 M sodium acetate buffer (pH 4.5). The absorbance was measured at 520 and 700 nm considering distilled water as blank and the concentration was measured according to Eq. (1).

where A(absorbance)=(A_{520nm}–A_{700nm}) in pH1.0–(A_{520nm}–A_{700nm}) in pH 4.5; MW = molecular weight for cyanidin-3-glucoside, which is 449.2 g/mol; DF = dilution factor; l = path length in cm; ε = 26900 molar extinction coefficient in L mol⁻¹ cm⁻¹ for cyanidin-3-glucoside; and 1000 = conversion factor from g to mg.

2.6. HPLC-ESI-MS characterization

HPLC-MS analysis was performed to characterize the phenolic compounds present in the peel. Analytes were separated using an

Agilent 1290 Infinity (Agilent, CA, USA) UHPLC fitted with a Phenomenex C18 column (100 mm × 2.1 mm × 1.8 µm). The mobile phase consisted of Milli-Q Water with 0.1%v/v formic acid (mobile phase A) and acetonitrile (mobile phase B). The gradient condition was: 0–0.7 min, 2% B; 0.7–14.0 min, 40% B, 14.0–22 min 80% B. 22.0–28.0 min, 2% B. The column was maintained at ambient temperature and the injection volume was 10 µL. Compounds were identified using an Orbitrap MS instrument (Q Exactive, Thermo Scientific, USA) with an ESI source operated on negative and positive modes. The instrument was operated at full scan mode covering a mass range from 50 to 2000 *m/z* with a resolution power set at 70,000 FWHM. The MS operational parameters were: 300 °C source temperature; 300 °C capillary temperature; −4 and 3.62 kV source voltage for negative and positive modes, respectively; 28 arb units sheath gas flow rate; 15 arb units auxiliary gas flow rate; 4 arb units sweep gas flow rate. The instrument was controlled using Xcalibur (Thermo Scientific, MA, USA) data handling system. Compounds were tentatively identified using Mass Frontier Spectral Interpretation software (Thermo Scientific, MA, USA) and compared with available databases (METLIN and mzCloud).

2.7. Cell lines and culture conditions

Two human melanoma cell lines were used; MM418C1 (BRAF mutated oncogene abbreviated as C1 henceforth) and D24 (wild type BRAF oncogene). Human fibroblasts served as the control (non-melanoma skin) cell line. The cell lines were cultured in RPMI 1640 medium (Sigma, USA) supplemented with 10% FBS at 37 °C temperature in a humidified atmosphere with 5% CO₂.

2.8. Cytotoxicity assay

The cytotoxicity of the extracts against melanoma cell lines was measured using a Cell Counting Kit-8 (CCK-8, Sigma-Aldrich, USA) according to (Fong et al., 2016). Briefly, the extracts were dissolved in dimethyl sulfoxide (DMSO, 0.09% v/v) at a concentration of 11 mg/mL. The cells were seeded in a 96-well microplate (Greiner, Austria) at a concentration of 5 × 10³ cells/well and incubated at 37 °C for 24 h. Thereafter, cell cultures were added with extracts at a final concentration of 15–500 µg/mL and incubated for 48 h. Cells treated with 0.09% DMSO served as the negative control. The absorbance in each well added with CCK-8 solution was measured at 450 nm using a microplate reader (CLARIOstar, BMG Labtech, Germany). The cell viability was calculated by determining the percentage of viable cells according to the following equation (Eq. (2)).

$$\text{Viability (\%)} = \frac{\text{Absorbance of treated}}{\text{Absorbance of control}} \times 100 \quad \text{Eq. 2}$$

The half inhibition concentration resulting in 50% cell viability (IC₅₀ value) was calculated from the dose response values. Two known anti-melanoma drugs; cisplatin and vemurafenib (BRAF V600E inhibitor), were used as positive controls.

2.9. Phase contrast microscopy

The morphology of treated and untreated cells was observed after 72 h of treatment using a Nikon Eclipse TS100 (Japan) inverted microscope under 20 × objective. The optical images produced by the microscope were captured using a DS-Fi 1 camera with a DS-L2 control unit.

2.10. Transmission electron microscopy (TEM) for cell ultrastructure

The morphological changes induced by the extracts on cell ultrastructure was investigated through TEM. The sample preparation for TEM was performed as before (Fong et al., 2016) and observed using a EOL JEM 1010 (Japan) transmission electron microscope operated at 80 eV (see Supplementary datasheet for a detailed protocol).

Table 1

Extraction yields, phenolics, flavonoids and anthocyanins from *G. humilis* peel.

Extraction condition	Yield of crude extract g/100 g sample)	Total phenolics (mg GAE/g)	Total flavonoids (mg QE/g)	Total monomeric anthocyanins
25 °C, 24 h	27.59	25.22 ± 1.42 ^a	1.04 ± 0.03 ^a	ND
37 °C, 24 h	32.00	24.35 ± 1.30 ^a	0.81 ± 0.05 ^b	ND
70 °C, 1 h	39.54	23.17 ± 1.74 ^a	0.83 ± 0.04 ^b	ND

ND-not detected. Means with different letters within a row are significantly different (Tukey's HSD, *p* < 0.05).

2.11. Statistical analysis

All analytical results were expressed as mean ± standard error (*n* = 3). One-way ANOVA was used to determine significant differences between means using Tukey's post-hoc test (*p* values < 0.05). Statistical analyses were performed using EXCELSTAT software (Addinsoft, USA).

3. Results and discussion

3.1. Total phenolics, flavonoids and anthocyanins

In this study, acidic methanol was chosen as the extraction solvent as it provides optimal recovery of phenolic compounds and other antioxidants compounds from plant biomass (Boeing et al., 2014; Magwaza et al., 2016). The highest crude yield was obtained when extraction was carried out at 70 °C for 1 h (Table 1) compared to the lower temperatures (24 and 37 °C) that required longer times for extraction. However, high yields did not equate to better extraction, but only marginally more flavonoids were extracted from the lower temperatures, whilst phenolics were similar (Table 1). The extraction was performed at three different combinations of temperatures and time (24 °C for 24 h, 37 °C for 24 h and 70 °C for 1 h) that are known determinants of stability, diversity and concentration of phytochemicals extracted (Poojary et al., 2016). Given the modest differences in extractables, the higher yield at 70 °C was likely due to the short incubation time of 1 h that had minimal effects on biomass.

The extracts contained substantial amounts of phenolic compounds (Table 1). The total phenolics ranged between 23 and 25 mg GAE/g DM, however, no significant differences in the yield were observed based on the extraction condition, suggesting that temperature did not affect the phenolics. Previous studies aimed at extraction of phenolic compounds from different plants have shown that the highest total phenolic content could be obtained at extraction temperatures of 60–80 °C (Antony & Farid, 2022). The total phenolics from *G. humilis* was more than a third lower than that detected in the peel of mangosteen, which contained about 74 mg GAE/g (Cheok et al., 2012). It is well known that the content and concentration of polyphenols vary depending on the type of fruit, season, and geographical conditions (Urbonaviciene et al., 2022). If the season and geographical conditions are disregarded, this study shows that *G. humilis* peel is an inferior source of phenolics compared to that of mangosteen peels. Therefore, *G. humilis* peel is not recommended as a potential source of phenolics when other plants have higher native concentrations.

The highest level of total flavonoids was detected in extracts obtained at 24 °C (1.04 mg QE/g) compared to those obtained at elevated temperatures (37 °C and 70 °C). This observation was attributed to the degradation of flavonoids at high temperatures (Biesaga, 2011).

Anthocyanins were not detected in any *G. humilis* peels. In contrast, the allied species with darker pigmented peels such as mangosteen *G. mangostana* (purple peel) and *G. indica* (red peel) contained considerable amounts of anthocyanins in their peels (2.92 mg cyanidin-3-

Table 2LC-Orbitrap MS based tentative identification of bioactive compounds in *G. humilis* peel extracts.

Retention time (min)	Monoisotopic mass	ESI mode	Exact mass m/z	Accurate mass m/z	Molecular formula	Tentative identification
4.45	578.1424	[M+H] ⁺	579.1497	579.1494	C30H26O12	Procyanidin B2 ^a
4.58	290.0790	[M+H] ⁺	291.0863	291.0862	C15H14O6	Epicatechin ^a
4.60	866.2058	[M+H] ⁺	867.2130	867.2126	C45H38O18	Procyanidin C1 ^a
4.93	578.1636	[M+H] ⁺	579.1708	579.1705	C30H26O12	Procyanidin B1 ^a
4.85	432.1057	[M+H] ⁺	433.1129	433.1126	C21H20O10	Apigenin 6-C-glucoside ^a
5.15	576.1268	[M+H] ⁺	577.1340	577.1337	C30H24O12	Procyanidin A2 ^a
6.36	558.1162	[M+H] ⁺	559.1234	559.1230	C30H22O11	Garcinia biflavonoid 1 ^a
8.36	426.1678	[M-H] ⁻	425.1605	425.1606	C24H26O7	Mangostanol ^b
9.56	396.1573	[M-H] ⁻	395.1500	395.1501	C23H24O6	Gamma-mangostin ^b

^a Flavonoids.^b Xanthenes.

glucoside/g hull powder) (Cheok et al., 2013; Nayak et al., 2010).

3.2. HPLC-ESI-MS putative identification of phenolic compounds

The extracts were subjected to LC-Orbitrap MS/MS analysis to putatively identify bioactive molecules. The chromatograms (see Supplementary datasheet Fig. S1 for a typical HPLC-MS chromatogram) showed the presence of epicatechin and procyanidin isomers (Table 2), which is in agreement with a previous study that also detected these molecules in *G. humilis* fruit peels (John et al., 2018). Quantifying identified compounds could not be carried out either due to matrix effects from overlapping peaks or lack of standards. Extracts with epicatechin and procyanidin isomers are known to exhibit antioxidant and antimelanoma activities (Bae et al., 2020; Ombra et al., 2019). Xanthenes specifically mangostanol and gamma-mangostin, were also tentatively identified in our extracts, typically found in mangosteen fruits (Ibrahim et al., 2016). The putative compounds were present in all the extracts irrespective of the extraction condition suggesting that these

phenolics were heat stable and mirrored the biochemical assays. Numerous studies have reported that flavonoids exhibit a wide range of biological benefits (Ullah et al., 2020). Furthermore, prenylated xanthenes possess ideal chemical structures for anticancer activities; and numerous studies have shown that xanthenes from *Garcinia* sp. exhibited anticancer activity against several cancer types [reviewed in (Kurniawan et al., 2021)]. A previous study reported that xanthenes isolated from *G. humilis* stems exhibited antioxidative capacity in mice tissues by increasing glutathione levels and prevented tumour necrosis factor levels (Mariano et al., 2016). The HPLC-MS chromatogram also showed the presence of other phytochemicals, but they could not be identified based on the MS spectral patterns and the database used on the present study (Fig. S1).

3.3. Cytotoxicity assay

A significant growth inhibition for C1 and D24 melanoma cells was observed after treatment with all extracts, and the viability of cancer

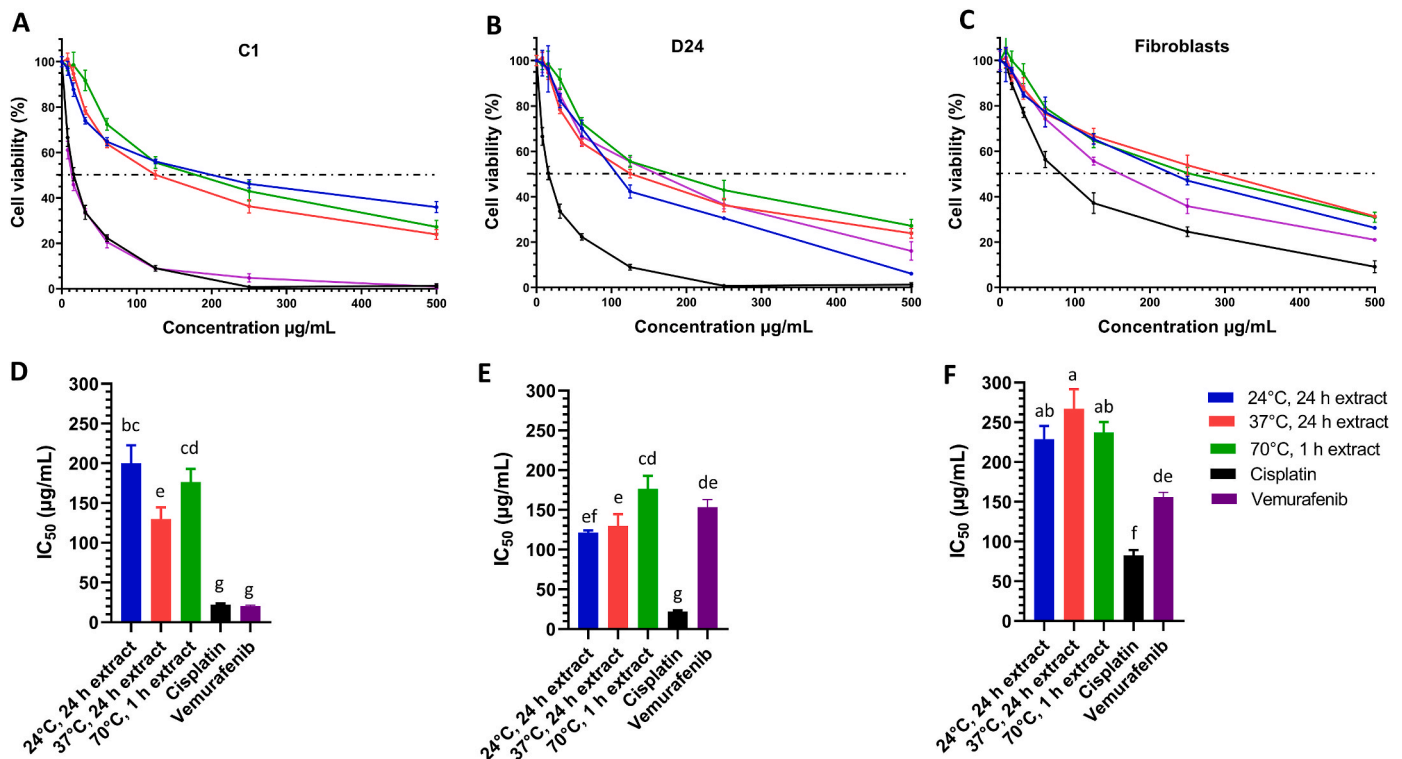


Fig. 1. Cell viability of *G. humilis* peel extract compared to anticancer drugs cisplatin and vemurafenib on C1 melanoma (A), D24 melanoma (B), and fibroblasts (C) cells. The dotted lines represented 50% cell viability corresponding to half inhibition concentration (IC₅₀) for C1 melanoma cells (D), D24 melanoma cells (E), and fibroblasts cells (F). The results were presented as the mean ± SEM, different letters indicated statistical significance at $p \leq 0.05$ between different treatments (Analysis by One-way ANOVA with Turkey's LSD test).

cells decreased as a function of extract concentration (Fig. 1A and B). In the case of C1 melanoma cells with BRAF mutated oncogenes, the extract obtained at 37 °C showed the highest cytotoxicity (IC₅₀ of 130 µg/mL) compared to that of extracts obtained at 24 °C (IC₅₀ = 200 µg/mL) and 70 °C (IC₅₀ = 176 µg/mL) (Fig. 1A). However, the extracts had significantly ~10x higher IC₅₀ than the control anticancer drugs used in the study, cisplatin and vemurafenib (22 and 20 µg/mL, respectively) (Fig. 1D). The modest cytotoxicity on C1 melanoma cells suggests inferior activity of the peel extracts to existing anti-cancer drugs but given that these plant extracts are crude and unrefined mixtures, the comparison with purified synthetic commercial drugs justifies further development as a herbal medicine or food ingredients.

In the case of D24 cells that contain wild type BRAF oncogenes, extracts obtained at low temperatures (i.e. 24 °C and 37 °C) induced higher cytotoxicity compared to 70 °C (Fig. 1B., with lower IC₅₀ (122–130 vs. 176 µg/mL) (Fig. 1E). These extracts had IC₅₀ comparable to the control drug vemurafenib that has specific activity on BRAF mutations not found in D24 cell types (153 µg/mL) but substantially higher than the generic melanoma drug, cisplatin (22 µg/mL) (Fig. 1E). The variation in cytotoxicity was reflective of the cell type and its susceptibility to different stimulants or drugs. Cisplatin is a generic anti-cancer drug and both C1 and D24 cells were therefore susceptible to cytotoxicity. Vemurafenib is a highly selective inhibitor of mutated BRAF V600E which is found in C1 cells. Again, this modest cytotoxicity on D24 melanoma cells would suggest its inferiority to current generic anti-cancer drugs like cisplatin, but given the crude and unrefined nature of the peel extract, especially extracted at 24 °C that surpassed the vemurafenib drug, this could justify further development as an alternative herbal medicine or food ingredients.

More importantly, the peel extracts exhibited lower cytotoxicity against the fibroblast cells than melanoma cells and the control drugs (Fig. 1C and F), suggesting that these extracts contain biomolecules that selectively target cancer cell lines, particularly D24 melanoma cells. This translates to similar inhibitory activity on melanoma cells as current anti-cancer drugs if the peel extracts were purified or refined, with less damage on normal cells, therefore a superior alternative that is natural and sustainable from a discarded waste plant product. The observed anti-melanoma activity of extracts could be attributed to the presence of flavonoids and xanthones (Table 2). Previous studies demonstrated that purified seed and stem extracts of *G. humilis* containing xanthones possess antiproliferative activities (Mariano et al., 2016; 2016; Niero

et al., 2012). However, reports on the anti-melanoma activity of peel extracts containing xanthones are unknown. Using crude plant extracts is more convenient than purified molecules in food products as they are less expensive and can be easily prepared. Moreover, natural product mixtures often show other beneficial activities than purified compounds due to synergistic interactions (Caesar et al., 2019).

3.4. Cellular morphology

G. humilis extracts induced ultramorphological changes on C1 and D24 melanoma cells (Fig. 2). The control untreated cancer cells were normal, healthy and firmly attached to the substrate, growing adjacent to each other and demonstrated typical cell morphologies characterised by distinct nucleolus and nuclear membranes (Fig. 2A and E). In comparison, the cells treated with the most effective extractions at 24 °C were abnormal, shrunken by more than half, detached from the substrate and floating in the media, fragmented nucleus and increased number of large vacuoles (Fig. 2B and F), characterizing the complex morphological changes that coincide with apoptotic cell death (Shalom & Cock, 2018). Extractions at 37 °C and 72 °C were not as morphologically different from untreated cells (Fig. 2C and D and 2G-H) suggesting less bioactivity than 24 °C extractions. Bioactive compounds, including flavonoids and xanthones, induce apoptosis by activating signal transduction cascades responsible for cell death (Li et al., 2016) and based on the viability and morphological results reported here, the most suitable extraction for these bioactive compounds were 24 °C. Previous investigations showed that flavonoids such as epicatechin and apigenin (identified in the present study) trigger apoptosis via elevation of reactive oxygen species, attenuating mitochondrial function, or by inducing DNA damage (Abotaleb et al., 2019). The phytochemicals present in *G. humilis* peels may independently or synergistically be involved in triggering apoptosis of C1 and D24 melanoma cells. However, in depth molecular studies is required to explore the underlying mechanisms.

4. Conclusions

The study showed that extracts obtained at lower temperatures (24 °C and 37 °C) exhibits anti-melanoma activity against human melanoma cells in a dose dependent manner and could be superior to current anti-cancer drugs with more refinement and purification, particularly against D24 melanomas. The best extract was obtained from

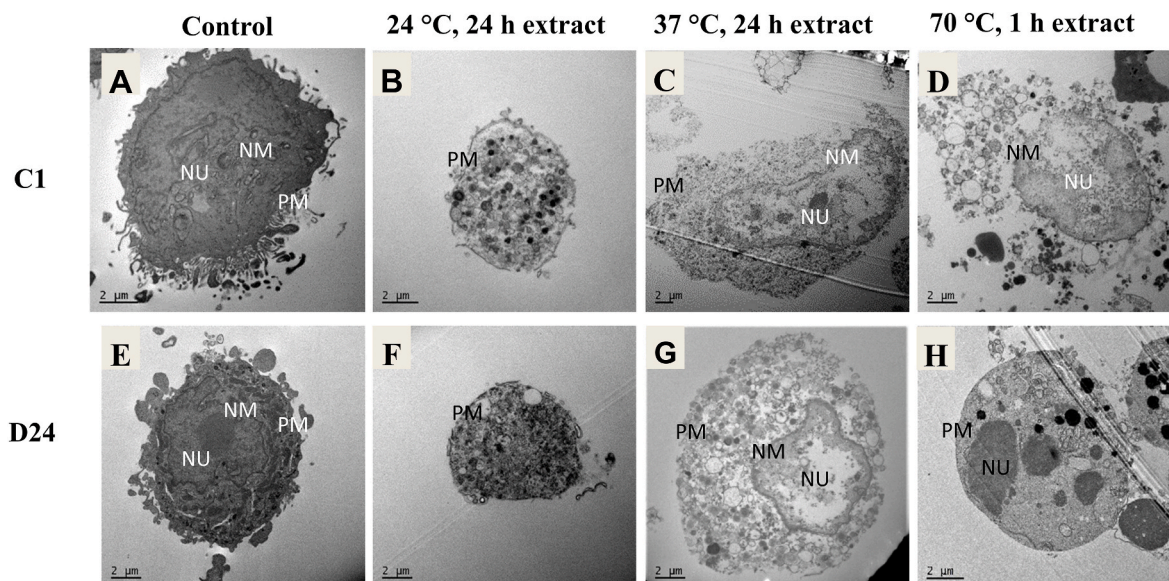


Fig. 2. Ultramorphological changes of C1 (A–D) and D24 (E–H) melanoma cells after 72 h treatment with *G. humilis* peel extracts. Cells were incubated with the vehicle only as the control (untreated) or extracts (treated) obtained at 24 °C, 37 °C and 70 °C. PM: plasma membrane, NM: nuclear membrane, NU: nucleolus.

24 °C against D24 melanoma and 37 °C against C1 melanoma and induced morphological changes consistent with apoptosis. Extracts contained bioactive flavonoids and xanthenes (epicatechin, procyanidin isomers, and mangostenol) which could be attributed to the anticancer activity. The extracts also contained several unidentified phytochemicals, which could potentially contribute to the observed bioactivity. Overall, this study shows that *G. humilis* peel could be developed as an anti-skin cancer ingredient as a sustainable and natural alternative.

Declaration of competing interest

The authors declare that there is no conflict of interest.

Data availability

Data will be made available on request.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.fbio.2023.102428>.

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