Research article

Effect of microwave heating on the phenolic and carotenoid composition and antioxidant properties of *Momordica charantia*

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A B S T R A C T

Momordica charantia L. (MC) is a widely consumed vegetable known for its nutritional benefits, as it is a rich source of carotenoids and phenolic compounds. Various cooking methods are use in domestic settings, including microwave cooking. Therefore, it is crucial to investigate the impact of microwave cooking on the bioactive composition of MC. MC fruits were subjected to microwave heat for 5-, 10-, and 15-min. High performance liquid chromatography was used to identify of carotenoids and phenolic compounds, and total bioactive composition and antioxidant assays were conducted using spectroscopic techniques. There were 17 carotenoids and chlorophylls identified in MC fruit, including lutein, violaxanthin, antheraxanthin, pheophytin *a*, and all-*E*-β-carotene. The levels of these compounds significantly increased upon exposure to microwave heating. Similarly, 16 phenolic compounds were identified, and their amounts increased during the treatments, except for 3-hydroxyphloretin-6′-hexoside, quercetin-3-(6″-acetyl)-glucoside, petunidin-3-(6″-acetyl)-glucoside and petunidin-3-(6″-acetyl)-glucoside. The sample subjected to microwave treatment for 15 min exhibited the highest concentration of total phenolic compounds (TPC) at 754.5 mg/100g. The total flavonoid content (TFC) reached 94.6 mg/100g after 10 min of treatment. Additionally, the maximum total anthocyanin content, reported as 54.8 mg/L, was observed in the sample exposed to microwave heating for 15 min.

1. Introduction

Momordica charantia L. (MC) belongs to the Cucurbitaceae family and is widely recognized as bitter gourd in English, and "*Karela*" in Urdu. This slender vine, characterized by tendrils, commonly grows on walls and shrubs [1]. In the fruits and leaves of '*Momordica*' different alkaloids were discovered, so they were called Momordicine II and Momordicine IV [2]. It is extensively cultivated in the tropical regions of Asia and Africa and grown in Pakistan. MC has gained significant importance in the field of vegetables due to its wide range of bioactive compounds, including phenolic compounds and carotenoids [3]. Importantly, MC has a strong antioxidant potential, which can be attributed to its flavonoids and other phenolic constituents [4]. Research has shown variations in antioxidant activity across different parts and stages of MC [5]. These bioactive compounds demonstrate antioxidant, antidiabetic, anticancer, antimicrobial, and antihypertensive properties [6].

The culinary versatility of MC extends to its consumption as a standalone cooked vegetable, occasionally integrated with meat or other vegetables, or prepared in a fried form like chips. Boiling, frying, and microwaving are common domestic techniques used to

prepare MC, with microwave cooking often used for dehydration purposes [7]. This method stands out as the most popular and efficient, reducing drying time and enhancing consumer acceptability [8]. Microwaving has become increasingly popular as a cooking technology due to its ability to rapidly heat food, thus shorten cooking times, ensure safe handling, offer convenient operation, and require minimal maintenance [9,10]. Furthermore, when compared to conventional cooking techniques, microwaving is less likely effect on the flavor and nutritional characteristics of food [11].

The effects of microwave power and air velocity was shown to affect total phenolic contents, vitamin C and antioxidant activities of MC [12]. Similarly, microwave cooking also produce deleterious effects on total phenolic contents and antioxidant activities [13]. The degradation of bioactive components has been reported to be influenced by temperature and drying time. The ideal treatment conditions are 13.52 m/s air velocity, 55.87 °C temperature, and 550.89 W microwave power [14]. While previous investigations have primarily examined the presence of bioactive compounds in MC in its raw form or when combined with other cooking methods, the impact of microwaving on the bioactive composition (phenolic and carotenoid profile) and antioxidant potential of MC has yet to be thoroughly explored. To the best of our knowledge there is also lack of carotenoid and phenolic profiling of MC of Pakistani origin. Therefore, this study holds importance in precisely assessing both profile and the effects of microwave heating on the bioactive composition and antioxidant properties of MC.

2. Materials and Methods

2.1. Sample collection and preparation

Fresh MC fruits were procured from the consumer market in Chakdara, District, Dir (L), with geographical coordinates at latitude 34.666° N, and longitude 72.029° E, at an altitude of 705 m. The fruits were carefully washed, and then divided into four portions, each consists of 5 fruits were cut into 1 × 1 cm sections and then subjected to microwave heating (Dawlance, Pakistan; 850W) heating for 5-, 10-, and 15-min. One portion of untreated fresh MC fruits were used as a control.

2.2. Analysis of carotenoids

2.2.1. Extraction

The extraction of carotenoids from each processed and control group followed a reported procedure [15]. Briefly, MC fruits (1 g) were ground into a homogenize paste, and 5.0 mL of ice-cold acetone with 0.1 % BHT was added. The mixture underwent 60 min of agitation using a Wised vortex mixer (Daihan Scientific, Korea) and was subsequently filtered. The residual material underwent a comparable procedure involving 5.0 mL of acetone, agitation for 30 min, and subsequent filtration. This extraction process was repeated until the leaves showed a loss of color. After vacuum expulsion of the filtrate at 35 °C, the residual material was dissolved in 2.0 mL of methanol, filtered using Agilent PTFE syringe filters (0.45 μm), and then transferred into a high-performance liquid chromatography (HPLC) vial.

2.2.2. Chromatography

A high-performance liquid chromatography with diode array detector (HPLC-DAD) system operating in reverse phase was used to separate the carotenoid pigments. The Agilent Zorbax C18 reversed-phase column, which measures 4.6 × 100 mm and has a particle size of 3.5 μm, was utilized in conjunction with an auto-sampler, quaternary pump, and degasser of HPLC system (1260 Infinite Better) for this purpose. A tertiary gradient system as mentioned in the original method [15] was employed. Solvent A consisted of methanol-deionized water (92:8 v/v), solvent B consisted of ammonium acetate (0.1 mM), and deionized water, and solvent C consisted of 100 % methyl tertiary butyl ether (MTBE). A 50 μL injection volume was used, and the flow rate was set at 1 mL/min. The spectra were determined at 190–750 nm, following the established methodology [16].

Chromatograms at 450 and 650 nm were obtained using Agilent Technologies' Open Lab Chemstation software. Compounds were identified by comparing retention times and absorption spectra with those of the standards. If standards were not available, compound identification was done by comparing the absorption spectra of unknown compounds with those reported in the literature. Quantification was achieved through peak area measurement, calibration curve analysis, and expressed as μg/g of fresh weight basis (FW).

2.3. Analysis of phenolic compounds

2.3.1. Extraction

To extract phenolic compounds (PCs), 1 g of the homogenized sample paste was mixed with 10 mL of methanol-water (9:1) and shaken for 2 h. After shaking, the mixture was filtered twice using Whatman filter paper, followed by ultra-filtration using a PTFE filter syringe (0.45 μm, Agilent Technologies, Germany). The filtrate was collected in HPLC vials for analysis.

2.3.2. Chromatography

The reverse-phase HPLC-DAD methodology was used to separate phenolic compounds. This involved the use of an HPLC system (1260 Infinity Better) consisting of an auto-sampler, quaternary pump, degasser, diode array detector (DAD), and a reversed-phase column (Agilent Zorbax C18, dimensions 4.6 × 100 mm, particle size 3.5 μm, maintained at 25 °C). The solvent system used was a binary solvent system, consisting of solvents A (methanol-acetic acid-water, 10: 2: 88, v/v/v), and B (methanol-acetic acid-water, 90: 2: 8, v/v/v). The flow rate was 0.6 mL/min, and the injection volume was 8 μL. The chromatographic elution followed the HPLC

method reported previously [17]. The diode array detector (DAD) chromatogram underwent calibration at 280, 320, and 360 nm across a spectral range of 200–600 nm. The identification of phenolic compounds (PCs) in the sample extract was based on the examination of retention time and absorption spectra, with results quantified and expressed as mg/100 g F.W.

2.4. Determination of total bioactive contents

2.4.1. Total phenolic contents

The quantification of total phenolic compounds (TPC) in the extract was carried out using a procedure described in the previous study [18]. In brief, 1 mL of 7.5 % Na_2CO_3 and 2.5 mL of Folin-Ciocalteu's reagent (0.2 N) were combined with 0.5 mL of the sample extract. After incubating for 1 h in the absence of light, the absorbance at 765 nm was measured using a spectrophotometer (Shimadzu-1700, Tokyo, Japan) and compared to blank. TPC for each sample, expressed as mg/100g of gallic acid equivalents (GAE) F.W, was determined in triplicate by utilizing a calibration curve for gallic acid as a reference.

2.4.2. Total flavonoid contents

The determination of the total flavonoid concentration (TFC) in the MC extract was conducted using quercetin as a standard. To do this, 0.5 mL of a 2.0 % $AlCl_3$ aliquot was added to 0.5 mL of the extract. The resulting mixture was then incubated for 1 h. The
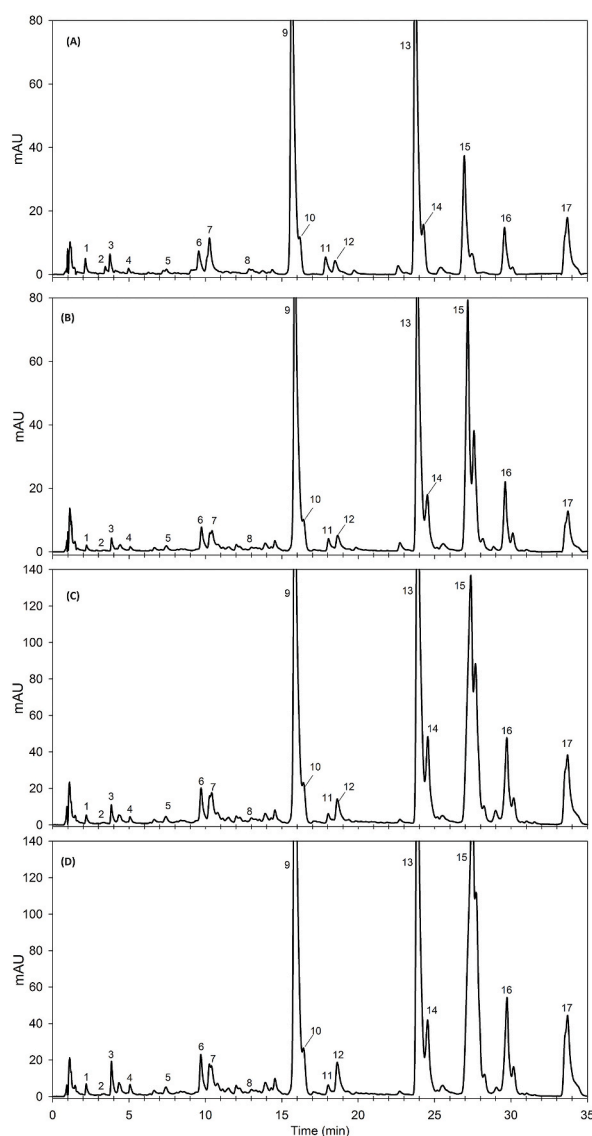
Fig. 1. HPLC-DAD chromatogram of carotenoids in MC. (A) Control samples, (B) microwaved for 5 min, (C) microwaved for 10 min, (D) microwaved for 15 min. Details are given in Table 1.

absorbance of the sample mixture at 420 nm was measured using a spectrophotometer. Triplicate measurements were taken, and the TFC for each sample was determined by referencing a quercetin calibration curve. The TFC was expressed as mg of quercetin equivalents (QE)/g FW.

2.4.3. Total anthocyanin contents

The determination of total anthocyanin content (TAC) in the extract was conducted using a pH differential method as outlined in the original method [19]. Briefly, solutions of potassium chloride (0.025 M) and sodium acetate (0.4 M) were adjusted to pH 1.0 and 4.5, respectively. Subsequently, 4.0 mL of sodium acetate solution was mixed with 1.0 mL of the sample extract, and absorbance measurements were taken at 520 and 700 nm. Similarly, 4.0 mL of potassium chloride and 1 mL of the sample extract were mixed, and absorbance at 520 nm and 700 nm was measured. TAC, reported as cyanidin-3-glucoside equivalents (mg/kg) FW, was calculated in triplicate for each sample.

2.5. DPPH radical scavenging activity

The evaluation of the sample extract's ability to neutralize free radicals was performed using a DPPH assay [20]. In summary, 1900 µL of freshly prepared DPPH reagent (0.1 mM) was mixed with 100 µL of the sample extract. After allowing the reaction mixture to sit in the dark for 30 min, the absorbance at 517 nm was measured for each sample. The assay was performed three times for each test, and the percentage radical scavenging activity was computed the absorbance of the sample and blank.

2.6. Statistical analysis

Data are presented as mean values of replicates (n = 5) or otherwise mentioned. Statistical analysis involved an ordinary one-way ANOVA, followed by multiple comparisons of variables using Dunnett's multiple comparison test. The significance level was set at $P < 0.05$.

3. Results

3.1. Carotenoid profile as affected by microwave

There were 17 carotenoid compounds identified and quantified in the MC fruits, as shown in Fig. 1. The chromatographic features and the impact of microwaving on the detected compound quantities are presented in Table 1. The first compound, 8-apocarotenal, had an elution time of 2.1 min and absorption maxima ($\lambda_{\max}$) at 466 nm. In the control group, it had a determined amount of 7.4 µg/g F W. After 15 min of microwave heating, this amount increased to 9.6 µg/g F W. The next compound, 13-Z-Zeaxanthin, eluted at 3.4 min with $\lambda_{\max}$ at 470 and 320 nm. It initially had a content of 4.1 µg/g F.W, which significantly ($P < 0.05$) increased to 12.0 µg/g F. W during a 15 min microwaving period. Pheophytin b and pheophorbide b, eluting at 3.8 and 4.9 min, respectively, showed substantial increases to 31.1 and 12.0 µg/g F.W after 15 min of heat exposure. The compounds (8'R)-neochrome and (8'S)-neochrome, eluting at 7.5 and 9.6 min, respectively, exhibited significant increases to 10.6 and 57.4 µg/g F.W, respectively, following 15 min of micro-waving. Microwaving also notably enhanced the concentrations of 13'-Z-violaxanthin and 13-Z-antheraxanthin, especially after a longer duration (15 min), reaching 353.1and 10.5 µg/g, respectively. Peak 9 represents lutein, eluted at 15.6 min with a spectral range of 472, 448 nm. Its initial content in the control was 361.9 µg/g F.W, which significantly escalated to 930.9 µg/g F.W after 15 min of

Table 1
Effect of microwave heating on carotenoid composition (µg/g) of MC fruit.

| Peak	Rt.(min)	Identity	$\lambda_{\max}$ (nm)	Control	05 min	10 min	15 min
1	2.1	8-Apocarotenal	466	7.4 ± 0.3a	4.3 ± 0.1b	7.8 ± 0.2a	9.6 ± 0.1a
2	3.4	13-Z-Zeaxanthin	470, 320	4.1 ± 0.2a	4.7 ± 0.1a	7.5 ± 0.2b	12.0 ± 0.1c
3	3.8	Pheophytin b	654, 438	10.5 ± 0.3a	7.2 ± 0.2b	18.3 ± 0.7c	31.1 ± 0.5d
4	4.9	Pheophorbide b	654, 435	3.6 ± 0.1a	3.0 ± 0.1a	8.6 ± 0.4b	12.0 ± 0.3c
5	7.5	(8'R)-Neochrome	450, 422, 400	5.5 ± 0.2a	3.6 ± 0.1a	9.3 ± 0.3b	10.6 ± 0.4c
6	9.6	(8'S)-Neochrome	450, 422, 400	20.2 ± 0.5a	16.1 ± 0.4b	53.1 ± 0.9c	57.4 ± 0.5d
7	10.3	13'-Z-violaxanthin	466, 436, 414	36.2 ± 0.5a	7.0 ± 0.3b	42.4 ± 0.4c	353.1 ± 0.0d
8	12.9	13-Z-antheraxanthin	466, 436, 415	7.1 ± 0.2a	6.0 ± 0.2a	9.3 ± 0.3a	10.5 ± 0.3b
9	15.6	Lutein	472, 448, 472	361.9 ± 4.4a	332.3 ± 2.4b	728.6 ± 3.0c	930.9 ± 4.1d
10	16.1	Violaxanthin	474, 446, 422	28.1 ± 0.3a	23.0 ± 0.4b	55.1 ± 0.7c	61.5 ± 0.3d
11	17.8	9-Z-Lutein	468, 442, 418, 325	15.4 ± 0.2a	10.9 ± 0.3b	14.0 ± 0.3a	11.9 ± 0.1c
12	18.5	Antheraxanthin	474, 446, 422	16.2 ± 0.3a	16.1 ± 0.4a	42.7 ± 0.3b	61.0 ± 0.6c
13	23.7	13'-Hydroxy-lactone chlorophyll b	650, 600, 464	338.9 ± 2.8a	316.9 ± 1.5b	750.6 ± 3.4c	750.0 ± 3.6d
14	24.3	15'-Hydroxy-lactone chlorophyll a	650, 600, 465	41.3 ± 0.4a	60.4 ± 0.3b	165.8 ± 2.5c	136.3 ± 0.7d
15	26.9	Pheophytin b	652, 528, 436	115.9 ± 0.6a	239.4 ± 2.4b	544.6 ± 1.2c	784.1 ± 5.0d
16	29.6	Pheophytin a	666, 608, 408	48.0 ± 0.8a	62.1 ± 1.1b	160.3 ± 1.5c	177.1 ± 1.5d
17	33.7	All-E-β-carotene	478, 452, 423	91.2 ± 0.2a	60.9 ± 0.6b	205.6 ± 3.2c	230.2 ± 0.5d

Different letters (a-d) represent significant difference with respect to control using Dunnett's multiple comparisons test at $p < 0.05$.

heating. Violaxanthin, the next identified compound at λ_{max} 474, 446, 422 nm, was determined to be 28.1 µg/g F.W, in control sample and significantly increased to 61.5 µg/g F.W during a 15 min microwaving period. Microwaving negatively impacted the quantity of 9-*Z*-lutein, eluted at 17.8 min, which decreased from 15.4 µg/g F.W in the control sample to 11.9 µg/g F.W after 15 min of microwaving. Antheraxanthin, identified at λ_{max} 474, 446, 422 nm and eluted at 18.5 min, exhibited a significant increase during microwaving. The compounds 13'-hydroxy-lactone chlorophyll *b* and 15'-hydroxy-lactone chlorophyll *a*, eluted at 23.7 and 24.3 min, respectively, with λ_{max} 650, 600, 464 and 650, 600, 465 nm, demonstrated significant improvements in quantity during microwave heating. Pheophytin *b* and pheophytin *a*, eluted at 26.9 and 29.6 min with λ_{max} 652, 528, 436 and 666, 608, 408 nm, respectively, significantly increased to 784.1and 177.1 µg/g F.W during microwave heating. Peak 17 represented the final compound, all-*E*-β-carotene, eluted at 33.7 min with λ_{max} 478, 452, 423 nm. The initial amount in the control sample was 91.2 µg/g F.W, and it increased significantly to 230.2 µg/g F. W during a 15 min microwaving period.

3.2. Phenolic profile as affected by microwave heating

A total of 16 phenolic compounds (PCs) were identified and quantified in the MC fruits, as illustrated in Fig. 2. The detailed chromatographic features and the effect of microwaving on the detected compound quantities are shown in Table 2. Quinic acid,
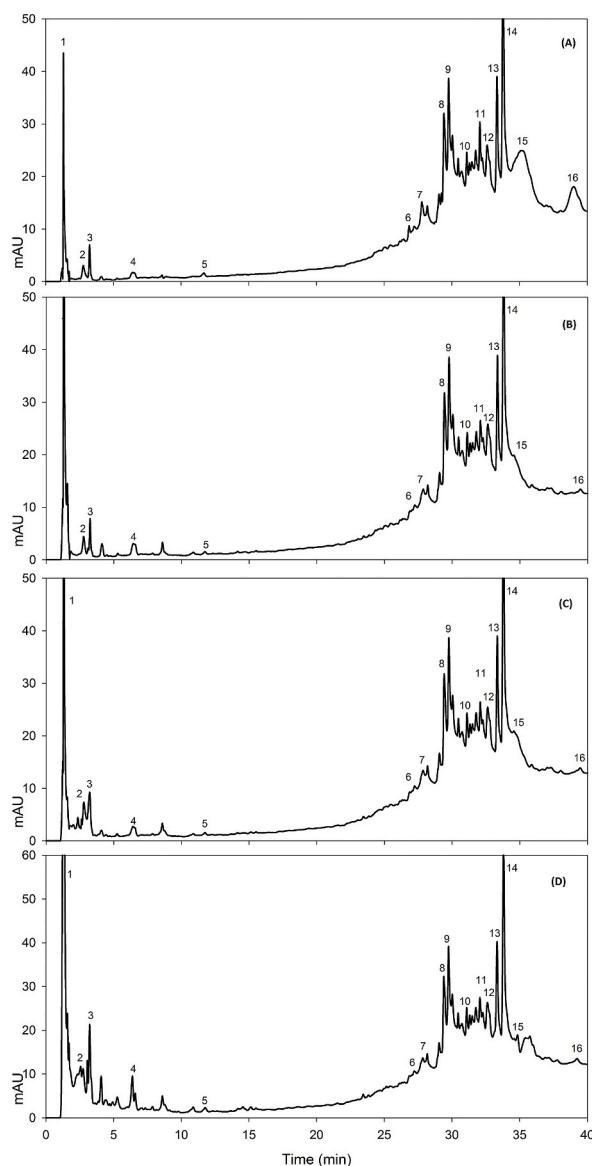
Fig. 2. HPLC-DAD chromatogram of phenolic compounds in MC. (A) control, (B) microwaved for 5 min, (C) microwaved for 10 min, (D) microwaved for 15 min. Details are given in Table 2.

initially present in minimal quantities in MC fresh fruit, exhibited an increased concentration of 0.6 mg/100 g F.W after 15 min of microwaving. Similarly, cinnamic acid and ferulic acid, initially measured at 0.19 and 1.2 mg/100 g F.W, in the control sample respectively, elevated to 0.29 and 4.5 mg/100 g F.W, after 15 min of heating. Peak 4, which denotes 5-caffeoylquinic acid with an elution time of 6.4 min and identification at λ_{max} 332 nm, showed an increase from 0.9 to 1.4 mg/100 g F.W, during 5 min of microwaving, followed by a subsequent decline. Quercetin-3,7-di-glucoside, which eluted at 11.7 min and was identified by a characteristic wavelength of 333 nm, exhibited no significant alteration in concentration during microwaving. Epicatechin-3-(4-methyl) gallate, which eluted at 26.8 min and was identified at λ_{max} 276 nm, increased from 0.8 to 1.5 mg/100 g F.W, during 5 min of microwaving with a subsequent decrease upon further heating. Peak 7, which represents 3-hydroxyphloretin 6′-hexoside, eluted at 27.8 min and was identified at λ_{max} 274 nm. It experienced a substantial decrease in concentration during microwaving. 5,7,3′-Trihydroxy-6,4′,5′-trimethoxyflavone, which eluted at 29.4 min with λ_{max} 275 nm, increased from 7.8 to 8.7 mg/100 g F.W, during 15 min of microwaving. Delphinidin-3,5-diglucoside, identified at λ_{max} 519, and 277 nm, and was eluted at 29.8 min, exhibited no notable change in concentration during microwaving. Petunidin-3-glucoside and malvidin-3-glucoside were identified by the characteristic wavelengths 523, 276 and 525, 276 nm, respectively, showed no notable change in concentration after microwaving. Petunidin-3-(6″-acetyl)-glucoside was identified at λ_{max} 523, 277 nm, and eluted at 32.6 min, displayed a slight decrease in concentration during microwaving. Isorhamnetin-3-(caffeoyldiglucoside)-7-rhamnoside and kaemferol-3-p-coumaroyl sinapoyldiglucoside, obtained at λ_{max} 333 nm and eluted at 33.3 and 33.8 min, respectively, exhibited a notable rise in their amounts during microwave heating. Quercetin-3-(6″-acetyl)-glucoside and petunidin-3-(6″-acetyl)-glucoside, the last two compounds eluted at 35.1 and 39.1 min and identified at λ_{max} 350, 246 and 523, 277 nm, respectively, experienced a significant decrease in concentration upon microwaving.

3.3. Determination of total bioactive content

3.3.1. Total phenolic content
The control sample exhibited a total phenolic content (TPC) of 410 mg/100 g F.W. However, in the sample subjected to 5 min of microwaving there was a significant increase to 496.8 mg/100 g F.W. Similarly, the TPC of the sample heated for 10 min was determined to be 578.9 mg/100 g F.W. Furthermore, when microwaved for 15 min, the TPC increased even more to 754.5 mg/100 g F. W, as depicted in Fig. 3.

3.3.2. Total flavonoid contents
The total flavonoid contents (TFC) of the raw MC fruits were initially measured at 91.5 mg/100 g F.W. After microwaving for 5 min, the TFC rose to 93.1 mg/100 g F.W. Similarly, the TFC further increased to 94.6 mg/100 g F.W after 10 min of microwaving. However, it was observed that prolonged heating (15 min) resulted in a decrease in the TFC of MC fruit, reaching 77.8 mg/100 g F.W as shown in Fig. 3.

3.3.3. Total anthocyanin contents
The total anthocyanin content (TAC) of MC fruit was found to be 11.3 mg/kg F.W in the control group. However, in the sample heated for 5 min, the TAC increased to 12.49 mg/kg F W. This increase was even more pronounced, reaching 48.7 mg/kg F.W in the sample microwaved for 10 min and peaking at 54.8 mg/kg F.W in the sample heated for 15 min. Fig. 3 illustrates the impact of microwave heating on the TAC of MC fruits, leading to the conclusion that microwaving induces significant changes in the TAC of MC.

Table 2
Effect of microwave heating on phenolic composition (mg/100g) of MC fruit.

Peak	Rt.(min)	Identity	λ_{max} (nm)	Control	05 min	10 min	15 min
1	1.3	Quinic acid	270	0.1 ± 0.01a	0.2 ± 0.01a	0.2 ± 0.010a	0.6 ± 0.01b
2	2.8	Cinnamic acid	280	0.19 ± 0.01a	0.20 ± 0.01a	0.28 ± 0.01a	0.29 ± 0.01a
3	3.2	Ferulic acid	323, 293	1.2 ± 0.08a	1.4 ± 0.08a	3.1 ± 0.1b	4.5 ± 0.1c
4	6.4	5-Caffeoylquinic acid	332	0.9 ± 0.04a	1.4 ± 0.2b	1.0 ± 0.1a	0.9 ± 0.1a
5	11.7	Quercetin −3,7-di-glucoside	333	0.5 ± 0.04a	0.6 ± 0.2a	0.4 ± 0.1a	0.6 ± 0.04a
6	26.8	Epicatechin-3-(4-methyl) gallate	276	0.8 ± 0.1a	1.5 ± 0.1b	1.1 ± 0.1a	0.9 ± 0.05a
7	27.8	3-Hydroxyphloretin 6′-hexoside	274	3.7 ± 0.1a	2.6 ± 0.2b	2.3 ± 0.2c	3.1 ± 0.1d
8	29.4	5, 7, 3′-Trihydroxy-6, 4′, 5′-trimethoxyflavone	275	7.8 ± 0.1a	7.3 ± 0.6a	7.2 ± 0.4c	8.7 ± 0.2d
9	29.8	Delphinidin-3,5-diglucoside	519, 277	10.0 ± 0.3a	8.5 ± 0.2b	8.4 ± 0.3c	10.0 ± 0.3a
10	31.1	Petunidin-3-glucoside	523, 276	4.4 ± 0.1a	3.0 ± 0.3b	2.7 ± 0.1c	4.6 ± 0.2a
11	32.0	Malvidin-3-glucoside	525, 276	5.4 ± 0.2a	3.2 ± 0.2b	3.0 ± 0.1c	5.4 ± 0.3a
12	32.6	Petunidin-3-(6″-acetyl)-glucoside	523, 277	10.8 ± 0.2a	4.8 ± 0.1b	4.6 ± 0.2c	9.7 ± 0.3d
13	33.3	Isorhamnetin-3-(caffeoyldiglucoside)-7-rhamnoside	333	9.4 ± 0.2a	5.0 ± 0.2b	4.7 ± 0.2c	12.3 ± 0.1d
14	33.8	Kaemferol-3-p-coumaroyl sinapoyldiglucoside	333	19.6 ± 0.4a	12.9 ± 0.3b	13.3 ± 0.3c	21.1 ± 0.1d
15	35.1	Quercetin-3-(6″-acetyl)-glucoside	350, 246	31.9 ± 0.8a	3.7 ± 0.1b	8.2 ± 0.3c	3.5 ± 0.1d
16	39.1	Petunidin-3-(6″-acetyl)-glucoside	523, 277	5.4 ± 0.1a	1.1 ± 0.1b	1.5 ± 0.1c	1.0 ± 0.1d

Different letters (a-d) represent significant difference with respect to control using Dunnett's multiple comparisons test at $p < 0.05$.
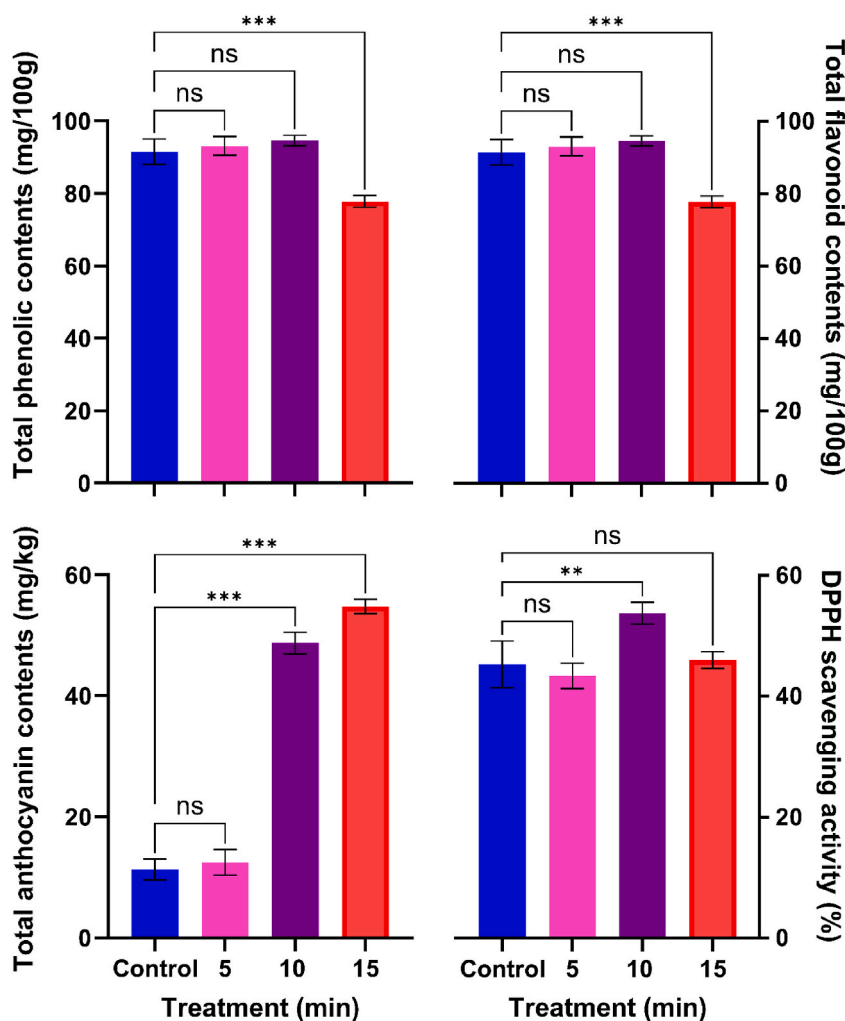
Fig. 3. Effects of microwaving on total bioactive contents and antioxidant activity of MC (F.W). Data are the mean of replicates (n = 5) with standard deviation. p * = 0.03, ** = 0.002, *** <0.001, ns = non-significant.

3.4. DPPH radical scavenging activity

The DPPH radical scavenging activity (% RSA) of MC fruit was found to be 45.3 % in the control sample, and no significant changes in the scavenging ability were observed after microwaving. For example, the % RSA of the sample heated for 5 min was reported as 43.4 %, and it increased to 53.8 % in the sample microwaved for 10 min. However, further heating resulted in a decrease in % RSA to 46 %, as illustrated in Fig. 3.

4. Discussion

This study identifies and quantifies essential bioactive compounds in MC fruit, such as 8-apocarotenal, lutein, antheraxanthin, violaxanthin, and all-*E*-β-carotene. While 8-apocarotenal was previously reported by Ahrazem et al. [21], lutein, violaxanthin, antheraxanthin, and all-*E*-β-carotene were reported in different studies [22,23]. The current investigation reaffirms the presence of these crucial carotenoids in MC fruit. Moreover, the study explores the impact of microwaving on the carotenoid composition of MC fruit, identifying a total of 17 carotenoid compounds. Microwave heating appears to positively influence the concentration of most of the identified compounds in MC fruit, except for 9-*Z*-lutein, which decreased with prolonged heat exposure. Mehmood and Zeb [24] suggested that the extractability of carotenoids may be stimulated by heat treatment, disrupting carotene-protein complexes in the food matrix. This disruption could be a key factor contributing to the observed significant increase in carotenoid contents in MC fruit.

The impact of heat treatment on the nutritional value and phytochemicals in green vegetables can lead to significant alterations or remain unchanged. This outcome depends on the structural characteristics, location within the food matrix, and heat stability of specific compounds. In the case of MC fruit, 16 phenolic compounds were identified, with the majority experiencing a notable increase

in concentration. This increase could be attributed to the release of bound phenolics from the fruit matrix induced by microwave heating. According to Turkmen et al. [25] the phenolic content in vegetables increases with heat processing, as thermal treatment can release linked and bound phenolic compounds. Our findings corroborates with Subramaniam et al. [26], Aminah and Permatasari [27] and Turkmen et al. [25] as most phenolic compounds in our study exhibited increased concentrations with longer heat duration. Nevertheless, specific compounds such as quercetin-3-(6″-acetyl)-glucoside, petunidin-3-(6″-acetyl)-glucoside, and petunidin-3-(6″-acetyl)-glucoside exhibited a decline under extended heat exposure. PCs have been recognized as chemo-preventive agents against oxidative damage, efficiently mitigating oxidative stress by impeding the oxidation of macromolecules [28]. Additionally, MC fruit serves as a source of antioxidants, playing a role in inhibiting or suppressing lipid oxidation and aiding in the repair of damaged cells [29]. The total anthocyanin content of MC fruit demonstrated a significant increase during microwaving, due to the heat effect that concentrated these compounds. It is important to highlight that there is no analogous data available in the literature regarding the influence of heat on anthocyanin content in MC fruits, emphasizing the unique contribution of the current study.

In conclusion, *Momordica charantia* is a vegetable that is rich in nutrients and contains important bioactive compounds, such as carotenoids and phenolic compounds. When cooked, the bioactive composition of this vegetable undergoes significant changes, with most of these compounds being enhanced extractability during microwave heat treatment. However, prolonged exposure to microwave heating of MC may result in a decrease in the levels of many bioactive compounds.

Data availability

Data will be made available on request.

CRediT authorship contribution statement

Alam Zeb: Writing – review & editing, Supervision, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **Ubaid Ullah:** Investigation, Formal analysis, Data curation. **Arif Mehmood:** Writing – original draft, Visualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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