

QUANTITATIVE ANALYSIS OF NUTRITIONAL AND BIOACTIVE  
COMPOUNDS IN FRUITS OF KIWANO (*CUCUMIS  
METULIFERUS* E. MEY) TEMPUS CULTIVAR

Micu Simona-Mariana, Popoviciu Dan Răzvan\*

„Ovidius” University of Constanța, Faculty of Natural Sciences and Agricultural Sciences, Constanța,  
Romania

\* Correspondence author. E-mail: dr\_popoviciu@yahoo.com

**Keywords:** *Cucumis metuliferus*, carotenoids, phenolic compounds, ascorbic acid, sugars, lipids

**ABSTRACT**

Locally-grown kiwano fruits, *Tempus cultivar*, were evaluated for their content in some main classes of nutritional organic compounds. With 448/98 mg/kg DW (pulp/peel) total carotenoid content was average when compared to other Cucurbitaceae. Among identifiable carotenoids, lycopene was the only one in notable amounts, in peel (12% of total carotenoids). With 51,751/37,624 mg/kg, total phenolic inventory was extremely rich. Flavonoids were clearly dominant (78/49%). Hydroxycinnamic acids were found in significant proportions (11/27%). Fruit pulp also showed some amounts of tannins (5.9%) and stilbenes (2%), while coumarins were found in low amounts. Ascorbic acid was abundant, 20,140 mg/kg in pulp and 7,689 mg/kg in peel. Soluble sugars were 15,915 /6,634 mg/kg while lipids were 5,556 /2,721 mg/kg.

**INTRODUCTION**

*Cucumis metuliferus* E. Mey (kiwano, horny cucumber, horny melon, jelly fruit, melano, pikano) belongs to the *Cucurbitaceae* family. A native of Southern Africa (Kalahari Desert area), it became a popular crop during the '80s in many parts of the world, including European countries. In Romania, the first noteworthy results were obtained at the SCDL Buzău in 1996 and since then some cultivars were successfully acclimatized in our country. *Tempus cultivar* is one of the most commonly grown here, omologated in 2008 at the same research station.

An annual, climbing plant, propagated through seeds, kiwano has deep root system, 2-3 m long stems, single tendrils, pentagonal leaves, monoecious yellow flowers and a characteristically spiky, fruit, with a thick, yellow-orange epicarp and a green fleshy mesocarp. Kiwano is appreciated outside Africa as a unique, exotic fruit, but also for its nutritional and potentially nutraceutical properties. The fruit is known to contain high amounts of vitamins A, C and E, iron, potassium, valuable fatty acids and terpenoids (ursolic acid). Known nutraceutical properties include free radical scavenging, blood cholesterol lowering, hypoglycemic etc. (Vînătoru et al. 2012; Busuioc et al. 2023). The species has been studied from a biochemical point of view, and there are studies especially on African cultivars, that found notable amounts of 31-64,000 mg/kg phenolic compounds, of which 2,100-8,500 mg/kg flavonoids, 150-260 mg/kg ascorbic acid, 15-16 mg/kg  $\beta$ -carotene etc. (Maluleke et al. 2021). *Tempus cultivar* was also studied, but mostly focusing on fruit juice and

using chromatographic means, pointing to several individual compounds (Busuioc 2023; Busuioc et al. 2023). The objective of this paper was to determine the amount of several classes of nutritional compounds and compound classes in both peel and pulp mature of Tempus kiwano fruit.

## MATERIAL AND METHODS

Ripe kiwano fruits (Tempus cultivar) were collected from the experimental farm of the “Ovidius” University of Constanța. Plants were grown under greenhouse conditions, at 70-80 cm distance. The seedlings were produced in the laboratory within „Ovidius” University, using seeds acquired from Vegetable Research and Development Station, Buzău. The date of sowing in Laboratory was March 15<sup>th</sup> and the planting date of seedlings in the greenhouse was 3<sup>rd</sup> of May, 2023. Plants were grown biologically. Fruit epicarp (peel) respectively mesocarp and endocarp (pulp) were separated. Peel was oven dried and extracted in 70% ethanol, while pulp was ground, filtered and used as juice. Amounts of both were oven dried at 105°C for determining dry weight.

Total carotenoids were determined by 10-fold dilution in 80% acetone and spectrophotometric absorption reading (ONDA UV-21 spectrophotometer) at 470 nm (Popoviciu et al. 2023). Among individual carotenoids,  $\beta$ -carotene, lycopene, lutein (and lutein esters), cryptoxanthin and zeaxanthin were determined by dilution in acetone:hexane:petroleum ether, petroleum ether, ether, acetone, respectively ethanol and determining absorbance at 445, 446, 453, 505, 645, 663 nm. Concentrations were calculated according to Braniša et al. 2014, Sujith et al. 2010, Biehler et al. 2009, Butnariu et al. 2014 and expressed as mg/kg DW.

The total amounts of phenolic compounds were determined by 10-fold dilution in methanol and Folin-Ciocalteu reaction. Absorbance was read at 765 nm against a gallic acid calibration curve (Popoviciu et al. 2023). Total flavonoids were determined by 10-fold dilution in water:methanol (4:8) and absorbance reading at 340 nm (Szabo et al. 2012). Flavanols (catechins), by dilution in 70% ethanol and reading at 280 nm (Yaneva et al. 2020). Anthocyanins, by dilution in 70% ethanol and reading at 520 and 700 nm (Braniša et al. 2014). Phenolic acids (hydroxybenzoic and hydroxycinnamic) were analyzed by dilution in ethanol-hydrochloric acid mixture and spectrophotometry at 220, 275, 325, 345 and 380 nm (Paula et al. 2017). Tannins were determined after thermal hydrolysis in water-hydrochloric acid mixture and reading at 550 nm (Moutari et al. 2018). Stilbenes, by 10-fold dilution in 70% ethanol and reading at 304 nm (Bancuta et al. 2015). Coumarins, by 10-fold dilution in 80% methanol and reading at 275 nm (Soares e Silva et al., 2012).

Total ascorbic and dehydroascorbic acid were determined by ethanol dilution, reaction with ammonium molybdate and sulfuric acid, and spectrophotometric reading at 494 nm (Riscahyani et al. 2019). Total soluble carbohydrates were determined by reacting extracts with sulfuric acid and phenol (5%) and reading at 490 nm (Agrawal et al. 2015). Lipids were determined by petroleum ether dilution and gravimetry (Orphanides et al. 2011). All concentrations were expressed as mg/kg DW.

## RESULTS AND DISCUSSIONS

The concentrations of various classes of compounds with bioactive potential are shown in Figures 1-3.

Average total carotenoid concentration was 448 mg/kg DW in fruit pulp and 98 mg/kg in peel. The only major identifiable compound was lycopene, with 12 mg/kg in peel (12% of the total amount). Some noteworthy amounts of lutein and cryptoxanthin (above 1 mg/kg) were also found in peel.  $\beta$ -carotene was found in concentrations below detection limit in all samples, different from the 15-17 mg/kg found in a South African cultivar by Maluleke et al. (2021).

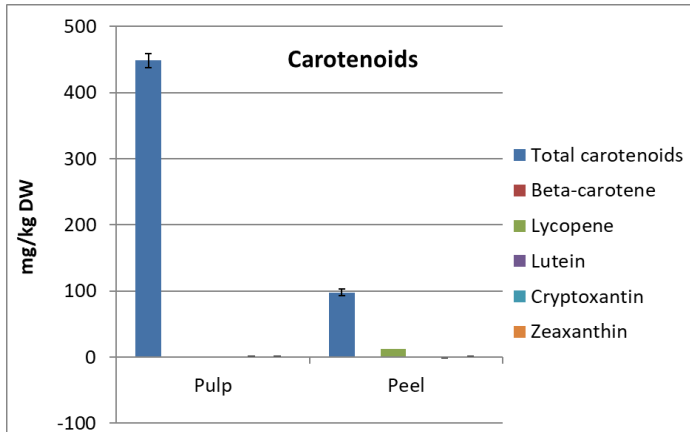


Figure 1. Concentrations of total and some individual carotenoids in ripe kiwano fruits – Tempus cultivar (mg/kg DW).

For comparison, in a study on three *Cucurbita* species (*C. maxima*, *C. moschata*, *C. pepo*), wide array of 41-3,797 mg/kg total carotenoids were found (Grassino et al. 2023). In local Romanian cultivars of *C. maxima*, *C. moschata* and *C. pepo* in Romania 42-65 mg/kg FW carotenoids were found (maximum in *C. moschata*, Dinu et al. 2016). 231 mg/kg (peel) and 251 mg/kg (pulp) were found in *Momordica charantia* (Popoviciu et al. 2023).

Average total phenolic content was 51,751 mg/kg in pulp and 37,624 mg/kg in peel. mg/kg. Of these, the flavonoid fraction was dominant, with 41,070 mg/kg in pulp (79%) and 18,271 mg/kg in peel (49%). Among flavonoids, flavonols and anthocyanins formed minute fractions in fruit pulp (1,513, respectively 50 mg/kg), while in peel were below detection limit. Hydroxycinnamic acids were also abundant, with 5,739 mg/kg in pulp (11%) and 10,331 mg/kg in peel (27%). Tannins formed 4-6% of the phenolic inventory, being richer in pulp tissue (2,417 mg/kg). Stilbenes (resveratrol and related compounds) were only found in pulp – 1,024 mg/kg – along with a small amount of coumarins (143 mg/kg).

*C. metuliferus* is known to be one of the richest Cucurbitaceae in terms of phenolic compounds, although the amounts seem to be highly variable or dependent on extraction method, from below 60,000 mg/kg (of which flavonoids formed a minority; Maluleke et al. 2021) to over 75,000 mg/kg (with some notable amounts of catechin, epicatechin and procianidins; Busuioc 2023; Busuioc et al. 2023).

For comparison, other Cucurbitaceae rich in phenolics are squash (*Cucurbita pepo*) with 3,867-5,306 mg/kg FW, pumpkin (*Cucurbita maxima*) – 549-921 mg/kg, cucumber (*Cucumis sativus*) – 238-323 mg/kg, bottle gourd (*Lagenaria vulgaris*) – 949-3,000 mg/kg and colocynth (*Citrullus colocynthis*) – 1,386-1,655 mg/kg (Al-Bakheit & Abu Zahra 2019). *Cucumis melo* peel was found to contain

3,320 mg/kg FW phenolics of which 924 mg/kg FW flavonoids (Mallek-Ayadi et al. 2017). *Coccinia grandis* had 18,960 mg/kg phenolics (3,000 mg/kg flavonoids) in peel and 9,500 (2,870) mg/kg in pulp; *Trichosanthes cucumerina*, 13,920 (3,240) in peel, 9,140 (2,510) in pulp and *Cucurbita moschata*, 15,310 (3,120) in peel and 14,380 (2,950) mg/kg in pulp (Akhter et al. 2022). In *Momordica charantia*, in peel and 18,598 mg/kg (13,527 mg/kg in peel), with dominant flavonoids (64-78%) and around 10% tannin fraction (Popoviciu et al. 2023).

Ascorbic acid concentration reached 20,140 mg/kg in pulp and 7,689 mg/kg in peel, higher than the amounts below 1,000 mg/kg found by Busuioc 2023; Maluleke et al., 2021, similar to those in *Momordica charantia* (12,847-14,689 mg/kg; Popoviciu et al., 2023). Lipid content was also higher in pulp (5,556 mg/kg) than in peel (2,721 mg/kg), the same being valid for sugars (15,915 mg/kg in pulp, 6,634 mg/kg in peel), both higher than, for example, in *Momordica charantia* (9,035-9,579 mg.kg sugars, 3,200-3,800/mg/kg lipids; Popoviciu et al., 2023).

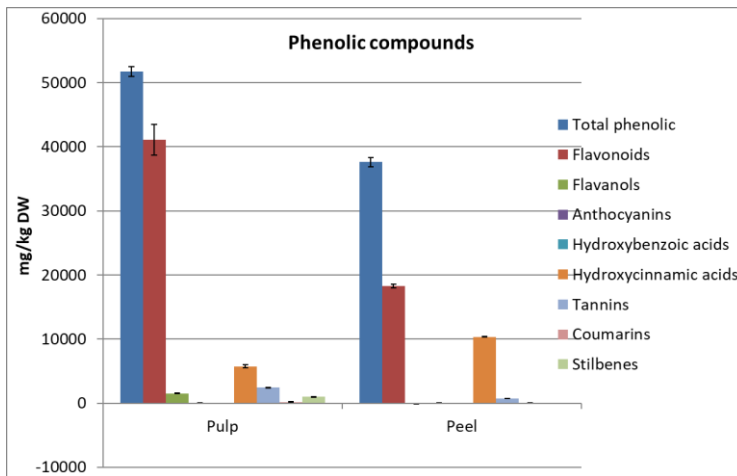


Figure 2. Concentrations of phenolic compound classes in ripe kiwano fruits – Tempus cultivar (mg/kg DW).

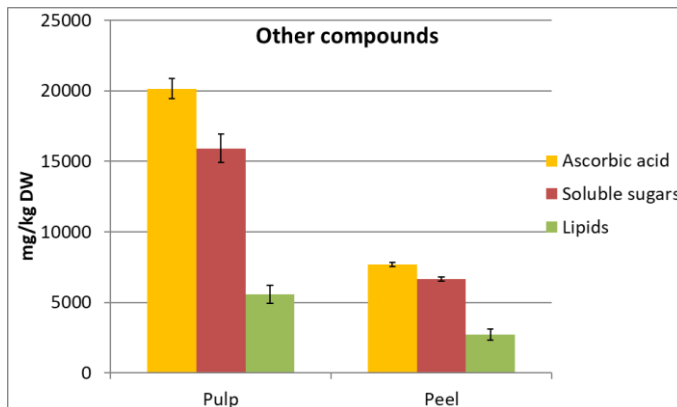


Figure 3. Concentrations of other nutritional compound classes in ripe kiwano fruits – Tempus cultivar (mg/kg DW).

## CONCLUSIONS

The carotenoid content in analyzed fruits was 448/98 mg/kg DW (pulp/peel). Among identifiable carotenoids, lycopene was the only one in notable amounts, in peel (12%), while lutein and cryptoxanthin were present in values above 1 mg/kg.

With 51,751/37,624 mg/kg, total phenolic inventory was extremely rich. Flavonoids were clearly dominant (78/49%); among them flavanols and catechins constituted minute fractions. Phenolic acids, namely hydroxycinnamic ones were also present (11/27%). Fruit pulp also showed some amounts of tannins (5.9%) and stilbenes (2%), while coumarins were found in low amounts.

Ascorbic acid was abundant, 20,140 mg/kg in pulp and 7,689 mg/kg in peel. Soluble sugars were 15,915 /6,634 mg/kg while lipids were 5,556 /2,721 mg/kg.

## REFERENCES

- Akhter K., Bibi A., Rasheed A., ur Rehman S., Shafique U., Habib T. 2022. Indigenous vegetables of family Cucurbitaceae of Azad Kashmir: A key emphasis on their pharmacological potential. *PloS One*, 17(6), doi: 10.1371/journal.pone.0269444.
- Al-Bakheit A.A.M., Abu-Zahra T.R. 2019. Determination of total polyphenolic compounds in organically and conventionally produced fruits of Cucurbitaceae family. *Acad. J. Nutr.*, 8 (3): 29-33.
- Agrawal N., Minj D.K., Rani K. 2015. Estimation of total carbohydrate present in dry fruits. *IOSR J. Environ. Sci. Toxicol. Food Technol.*, 1(6): 24-27.
- Bancuta O.R., Chilian A., Bancuta I., Ion R.M., Setnescu R., Setnescu T., Gheboianu A., Lungulescu M. 2015. FT-IR and UV-VIS characterization of grape extracts used as antioxidants in polymers. *Rev. Roum. Chim.*, 60(5-6): 571-577.
- Biehler E., Mayer F., Hoffmann L., Krause E., Bohn T. 2009. Comparison of 3 spectrophotometric methods for carotenoid determination in frequently consumed fruits and vegetables. *J. Food Sci.*, 75(1), doi: 10.1111/j.1750-3841.2009.01417.x.
- Braniša J., Jenišová Z., Porubská M., Jomová K., Valko M. 2014. Spectrophotometric determination of chlorophylls and carotenoids. An effect of sonication and sample processing. *J. Microbiol. Biotech. Food Sci.*, 3(2): 61-64.
- Busuioc A.C. 2023. Studiul compușilor chimici din specii de plante de interes terapeutic. Teză de doctorat, Universitatea „Dunărea de Jos” din Galați.
- Busuioc A.C., Costea G.V., Botezatu A.V.D., Furdui B., Dinica R.M. 2023. *Cucumis metuliferus* L. fruits extract with antioxidant, anti-inflammatory, and antidiabetic properties as source of ursolic acid. *Separations*. 10(5), doi: 10.3390/separations10050274.
- Butnariu M., Rodino S., Petrache P., Negoescu C., Butu M. 2014. Determination and quantification of maize zeaxanthin stability. *Dig. J. Nanomater. Biostructures*, 9(2): 745-755
- De Lima D.B., Agustini B.C., da Silva E.G., Gaensly F., Cordeiro R.B., Fávero M.L.D., Brand D., Maraschin M., Bonfim T.M.B. 2011. Evaluation of phenolic compounds content and in vitro antioxidant activity of red wines produced from *Vitis labrusca* grapes. *Food Sci. Technol. (Campinas)*, 31(3): 783-800.
- Dinu M., Soare R., Hoza G., Becherescu A.D. 2016. Biochemical composition of some local pumpkin population. *Agriculture and Agricultural Science Procedia*, 10: 185-191.
- Grassino A.N., Brnčić S.R., Sabolović M.B., Žlabur J.Š., Marović R., Brnčić M. 2023. Carotenoid content and profiles of pumpkin products and by-products. *Molecules*, 28(2), doi: 10.3390/molecules28020858.

Mallek-Ayadi S., Bahloul N., Kechaou N. 2017. Characterization, phenolic compounds and functional properties of *Cucumis melo* L. peels. Food Chem., 221: 1691-1697.

Maluleke M.K., Moja S.J., Nyathi M., Modise D.M. 2021. Nutrient concentration of African horned cucumber (*Cucumis metuliferus* L) fruit under different soil types, environments, and varying irrigation water levels. Horticulturae, 7(4), doi: 10.3390/horticulturae7040076.

Moutari S.K., Abdoukadi A.M., Boulhassane A.S., Rabani A., Khalid I. 2018. Determination contents of total phenolic pigments and spectrophotometric characterization of crude extracts of ten tinctorial plants of Niger which is usable in solar energy. Eur. Sci. J., 14(33): 389-407.

Orphanides A., Goulas V., Chrysostomou M., Gekas V. 2011. Recovery of essential oils from carobs through various extraction methods. In Mastorakis N., Mladenov V., Lepădătescu B., Karimi H.R., Helmis C.G. (eds.), Recent Advances in Environment, Energy Systems and Naval Science, WSEAS Press, Athens, 219-224.

Paula V.B., Estevinho L.M., Dias L.G. 2017. Quantification of three phenolic classes and total phenolic content of propolis extracts using a single UV-vis spectrum. J. Apic. Res., 56(5): 569-580.

Popoviciu D.R., Pricop S.M., Radu M.D. 2023. Quantitative analysis of nutritional and bioactive compounds in bitter melon (*Momordica charantia* L.) fruits. Ann. Univ. Craiova, Ser. Biol. Hort. Food Prod. Process. Environ. Eng., 28(64): 21-26.

Riscahyani N.M., Ekawati E.R., Ngibad K. 2019. Identification of ascorbic acid content in *Carica papaya* L. using iodimetry and UV-Vis spectrophotometry. Indones. J. Med. Lab. Sci. Technol., 1(2): 58-64.

Soares e Silva L., Santos da Silva L., Larissa Brumano L., Stringheta P.C., de Oliveira Pinto M.A., Dias L.O.D., de Sá Martins Muller C., Scio E., Fabri R.L., Castro H.C., da Penha Henriques do Amaral M., 2012. Preparation of dry extract of *Mikania glomerata* Sprengel (guaco) and determination of its coumarin levels by spectrophotometry and HPLC-UV. Molecules, 17(9), doi: 10.3390/molecules170910344.

Sujith A.M.A., Hymavathi T.V., Devi P.Y. 2010. Supercritical fluid extraction of lutein esters from marigold flowers and their hydrolysis by improved saponification and enzyme biocatalysis. Int. J. Nutr. Food Eng., 4(1): 74-83.

Szabo I., Vonhaz G., Fodor A., Bungău S., Țiț D.M. 2012. The quantitative analysis through spectrophotometry of flavonoids and polyphenols from vegetable products Hibiscus trioni herba, radix and fructus. Analele Univ. Oradea, Fasc. Protecția Mediului, 18, 73-80.

Vinătoru C., Teodorescu E., Zamfir B. 2012. *Cucumis metuliferus*, a new acclimatized and bred species at V.R.D.S. Buzău. Lucr. Științ. USAMV Iași, Ser. Hort., 55(2): 185-190.

Yaneva Z., Ivanova D., Beev G., Besheva K. 2020. Quantification of catechin in *Acacia catechu* extract by non-derivative, first derivative UV/Vis spectrophotometry and FT-IR spectroscopy. Bulg. Chem. Commun., 52: 41-47.