Determination of Sugar Contents of Various Postharvest Climacteric Fruits Using HPLC-ELSD; Evaluation at Different Ripening Stages and Assessment of the Greenness Profile of the Method with Analytical Eco-scale, NEMI, GAPI and AGREE

Burcu Sezgin[1*], Murat Soyseven[2], Göksel Arli[3]

[1] Eskişehir Osmangazi University, Eskişehir Vocational School, Department of Environmental Protection Technologies, Eskişehir, Türkiye

[2] Anadolu University, Yunus Emre Vocational School of Health Services, Department of Medical Services and Techniques, Eskişehir, Türkiye

[3] Anadolu University, Faculty of Pharmacy, Department of Analytical Chemistry, Eskişehir, Türkiye

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HIGHLIGHTS

- Sugar content of tomato and banana was determined at different ripening stage.
- Applied HPLC-ELSD method was evaluated via Analytical Eco-scale, NEMI, GAPI and AGREE.
- Proposed method requires easy sample preparation steps.
- Total sugar content of tomatoes and bananas increases by ripening.

Abstract

The major sugar content in fruits is crucial in terms of diet. In this study, changes in fructose, glucose and sucrose amounts and total sugar content were investigated at different ripening stages (green, ripening, ripe and over-ripe) in tomatoes and bananas using an HPLC-ELSD method. Chromatographic separations were conducted using an HPLC column with a Phenomenex Luna NH_2 stationary phase (5 μm particle size, 250 mm x 4.6 mm internal diameter, 100 Å), and eluted with a mobile phase consisting of acetonitrile:water (78:22, *v/v*). Quantitative results demonstrated a significant increase (p < 0.0001) in fructose and glucose contents of bananas from 0.87 g/100 g and 1.27 g/100 g at the green stage to 5.76 g/100 g and 11.13 g/100 g at the over-ripe stage, respectively, while sucrose peaked at 4.41 g/100 g during the ripening stage and declined thereafter. In tomatoes, fructose and glucose contents showed a moderate but statistically significant rise (p < 0.01) from 4.21 g/100 g and 6.48 g/100 g to 4.86 g/100 g and 6.55 g/100 g, whereas sucrose became undetectable after the ripening stage. Total sugar content increased by approximately sixfold in bananas and slightly in tomatoes throughout ripening. These results indicate that during fruit ripening, sucrose is converted into fructose and glucose. The applied HPLC-ELSD method has been shown to be easily applicable for analysing climacteric fruits for sugar content. In addition, the greenness profile of the applied HPLC-ELSD method was evaluated using AES, NEMI, GAPI, and AGREE tools,

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confirming that the method is environmentally acceptable and consistent with the principles of Green Analytical Chemistry.

Keywords: Climacteric Fruits; Sugars; HPLC-ELSD; Tomato; Banana; Greenness Assessment

1. Introduction

The development and maturation of fruits involves complex molecular, biochemical, and physiological processes influenced by internal and external factors. Fruit development is divided into three stages: slow growth due to cell division (stage 1), rapid growth due to cell expansion (stage 2), and fruit ripening characterized by biochemical changes resulting in organoleptic attributes (stage 3). Fruits are classified into two physiological categories, climacteric and non-climacteric, based on their respiration pattern and ethylene biosynthesis during stage 3. Climacteric fruits, such as tomato, apple, and banana, have an increase in respiration rate and ethylene production during ripening, allowing for harvest prior to complete fruit maturation (Alferez et al., 2021; Klee et al., 2011; Kou & Wu, 2018). Climacteric fruits are a type of fruit that undergoes a ripening process that is characterized by a significant increase in respiration and ethylene production, which leads to changes in colour, flavour, and aroma. This process is triggered by the release of ethylene gas, which signals the fruit to begin the ripening process. Some examples of climacteric fruits include apples, bananas, tomatoes, pears, mangos, jujube fruits, avocados (Chen et al., 2018; Kou et al., 2021; Payasi et al., 2010). The ripening process in climacteric fruits is a complex biochemical process that involves the activation of various enzymes which break down the cell walls and starches in the fruit, making it softer and sweeter. This process also leads to a change in the colour of the fruit (Kim et al., 2015; Yumbya et al., 2021).

Climacteric fruits have many health benefits as well. For instance, climacteric fruits are rich in fiber and help maintain digestive health. They also reduce the risk of cancer with their antioxidant content and are rich in vitamins and minerals (Jaglan et al., 2022; Liu, 2003; Singh et al., 2016). Climacteric fruits undergo a reaction chain that increases during the ripening process. During this process, starch in the fruits is converted into fructose and glucose when dissolved. Additionally, sucrose is another sugar found in fruits and is the largest source of sugar (Maduwanthi & Marapana, 2017; Subedi & Walsh, 2011).

Fructose (Fr), glucose (Gl), and sucrose (Sc) are the most prevalent mono- and disaccharides found in fruits and are considered the primary sugars present (Georgelis et al., 2018; Soyseven et al., 2022). Fr, Gl, and Sc molecular structures are shown in Figure 1. Various analytical methods have been studied for the detection of Fr, Gl, and Sc in fruits, such as high-performance liquid chromatography (HPLC) combined with charged aerosol detector (CAD) (Grembecka et al., 2014), refractive index detection (RID) (Filip et al., 2016; Ivancic et al., 2022), liquid chromatography/mass spectrometry (LC-MS) (Tsai et al., 2022), gas chromatography (GC) (Ruiz-jimé Nez et al., 2010).
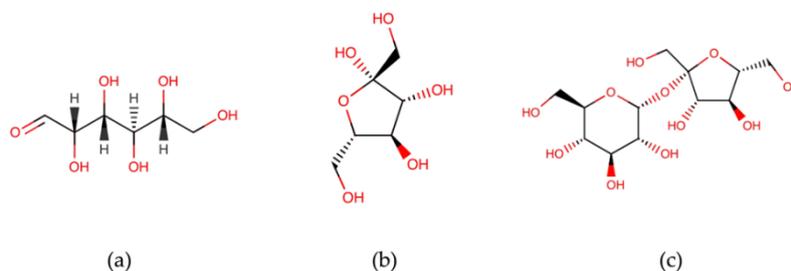

Figure 1. Molecular structure of glucose (a), fructose (b) and sucrose (c)

Evaporative light scattering detection (ELSD) is considered to be a nearly universal detector that is suitable for a wide range of analytes, including non-absorbing and end-absorbing analytes. ELSD's capability to provide a more consistent level of detection sensitivity for most analytes, regardless of their physical or chemical properties. This is because ELSD relies on the measurement of scattered light intensity rather than absorption, making it less dependent on the specific optical properties of the analyte. This can be particularly

useful when analysing complex mixtures or when using a variety of solvents (Ma et al., 2014; Mojsiewicz-Pieńkowska, 2009; Nogueira et al., 2005; Soyseven et al., 2022, 2023).

In recent years, Green Analytical Chemistry (GAC) has received growing attention for its significant contribution to minimizing the environmental footprint of analytical processes and fostering sustainable laboratory practices. The main goals of GAC are to protect human health, reduce the use of hazardous substances, and minimize waste generation throughout analytical workflows (Hussain et al., 2025; Sezgin et al., 2025). In this study, the greenness profile of the developed HPLC-ELSD method was evaluated using the most widely applied green assessment tools in order to assess the overall environmental impact of the analytical procedure. These tools include the National Environmental Methods Index (NEMI) (Keith et al., 2007), which provides a simple visual representation of method greenness by evaluating parameters such as reagent toxicity, waste generation, and pH of the waste; the Analytical Eco-Scale (AES) (Gałuszka et al., 2012), a semi-quantitative tool that assigns penalty points to non-green aspects of a method, with higher scores indicating a greener analytical procedure; the Green Analytical Procedure Index (GAPI) (Płotka-Wasylka, 2018), which assesses the environmental impact of an analytical method throughout all procedural steps using a color-coded pictogram; and the Analytical GREEnness metric (AGREE) (Pena-Pereira et al., 2020), a holistic approach that quantifies the overall greenness of analytical methods based on the twelve principles of Green Analytical Chemistry, providing both a numerical score and visual output.

This research aims to analyse the changes in the sugar content of tomato and banana samples due to ripening using an easy and efficient way. For this purpose, the amounts of Fr, Gl, and Sc present in tomatoes and bananas were determined at different stages of ripening using the HPLC-ELSD method. It could play an important role in determining harvest times and optimizing storage conditions of the fruits.

2. Materials and Methods

2.1. Chemicals, materials and apparatus

Standards of Fr, Gl, and Sc with a purity of over 99% and analytical grade were purchased from Sigma-Aldrich (Germany). HPLC-grade water and acetonitrile (MeCN) with a purity of over 99% were obtained from Carlo-Erba (France). Nitrogen gas (N_2) with analytical purity was used. All chemicals and reagents used were of analytical grade. Prior to injection into the HPLC system, the solutions of both the working standards and the samples were subjected to filtration through non-sterile syringe filters composed of polyvinylidene fluoride (PVDF) material, featuring a pore size of 0.22 μm and a diameter of 13 mm.

The HPLC-ELSD analysis was performed using an HPLC (Shimadzu Nexera-i 2040C 3D HPLC system) that included an ELSD LT-II Model (Tokyo, Japan). The system was also equipped with a binary pump, a gas degasser, an autosampler, and a temperature-controlled column compartment. To prepare the reference standards and plant extract samples, sonication was carried out using a LabCompanion CS-10 ultrasonic bath (Seoul, Korea). Mixing of the prepared samples was done using a Heidolph Vortex (Schwabach, Germany). All weighing procedures were performed using an analytical weighing system from Ohaus (USA). Data processing of HPLC-ELSD system results was carried out using LabSolution Shimadzu Corporation software (Tokyo, Japan), and MS Excel from Microsoft Corporation (USA).

2.2. Sample collection and sample preparation

Samples of unripe climacteric fruits including banana and tomato were obtained from local producers and were evaluated for changes in Fr, Gl, and Sc content at four different ripening stages (green stage, ripening stage, ripe stage, over-ripe stage) using HPLC-ELSD. Bananas and tomatoes were ripened under carefully controlled laboratory conditions to assess sugar transformations at defined ripening stages. Temperature was consistently maintained at 23 ± 1 °C and relative humidity at $65 \pm 5\%$, and no external ripening agents were applied to ensure natural ripening processes. Ripening was categorized into four stages based on visual colour changes, reflecting the inherent ripening dynamics of each fruit. For bananas, green stage (fully green) lasted from day 0 to 2, ripening stage (onset of yellowing) from day 3 to 5, ripe stage (fully yellow) from day 6 to 8, and over-ripe (mostly brown, ripe) from day 9 to 11. For tomatoes, green stage (fully green) spanned day 0 to

2, ripening stage (green-red transition) day 3 to 4, ripe stage (fully red) day 5 to 7, and over-ripe (mostly deep red with brown spots) day 8 to 10. Corresponding to the collected fruits at each predefined ripening stage, about 2.0 g of banana and tomato samples were extracted with 100 mL of water in an ultrasonic bath for 20 min. All extracts were then centrifuged for 10 min, and the supernatant was filtered through a 0.22 μm membrane prior to HPLC-ELSD analysis (Soyseven et.al., 2022).

2.3. Chromatographic conditions

The method employed in our previous study was used for chromatographic separation with minor modifications (Soyseven et al., 2022). For separation of major sugars, a Phenomenex Luna NH_2 HPLC column (5 μm particle size, 250 mm x 4.6 μm diameter, 100 Å) was used. Isocratic elution was performed with a mobile phase comprising MeCN:water (78:22, *v/v*). The mobile phase was delivered to the HPLC-ELSD system at a flow rate of 1.5 mL min^{-1}, and each analysis was completed within 18 minutes. The column oven temperature was set to 30°C, and the injection volume was 10 μL. The ELSD system's gain and filter values were set to 7 and 10, respectively, with the evaporation temperature set to 40°C and N_2 pressure set to 350 kPa.

2.3. Calibration curves and statistical analysis

During the calibration phase, solutions containing Fr, Gl, and Sc were prepared and 10-point calibration curves covering a range of 50-2000 μg mL^{-1} were obtained. Triplicate analyses were conducted for each sugar compound using the prepared calibration sets. The calibration curve for the developed HPLC-ELSD method was generated by plotting the logarithm of the chromatogram peak areas against the logarithm of the standard solution concentrations of Fr, Gl, and Sc (Mojsiewicz-Pieńkowska, 2009).

The processing of data obtained from the HPLC-ELSD system was performed using LabSolution Shimadzu Corporation (Tokyo, Japan) software. All analyses were performed in triplicate (n=3) and the mean values were presented with standard deviation data. Microsoft Excel 2019 (Microsoft Corporation, USA) program was used for statistical analysis of the data.

Replicated analytical measurements (n = 3) for each ripening stage were subjected to one-way ANOVA to assess stage-dependent differences in Fr, Gl, and Sc levels. Tukey's HSD post-hoc test was performed for multiple comparisons using a significance threshold of α=0.05. Statistical analysis was performed using a reconstructed raw dataset generated from the experimental mean ± SD values, ensuring full compliance with ANOVA assumptions. Statistical calculations were carried out using GraphPad Prism version 6.0b.

2.4. Greenness Assessment Tools

The greenness profile of the applied HPLC-ELSD method was analysed using four different greenness metrics. NEMI, AES, GAPI, and AGREE were used to evaluate the environmentally friendly properties of the applied method.

The developed analytical method was evaluated for its environmental impact using the NEMI approach. Data on the persistent, bioaccumulative and toxic (PBT) status, hazardous chemical content, applied pH conditions, and waste generation of the chemicals used in the method were collected and assessed according to four primary acceptance criteria. Each criterion was examined to determine whether the method met the corresponding environmental requirement; the quadrant representing a fulfilled criterion was filled in green, whereas the quadrant for an unmet criterion was left blank (Keith et al., 2007).

The greenness of the proposed analytical procedure was further evaluated using the AES. In this assessment, penalty points were assigned based on parameters such as the type and amount of reagents used, energy consumption, occupational hazards, and waste generation. The total score was obtained by subtracting the sum of penalty points from the ideal value of 100, where higher scores indicate a greener analytical method (Gałuszka et al., 2012).

The assessment of the GAPI tool, which was downloaded from the following source: https://mostwiedzy.pl/pl/justyna-plotka-wasylka,647762-1/gapi. The tool facilitated the evaluation of the entire analytical workflow, providing a visual representation of the method's greenness across multiple parameters (Płotka-Wasylka, 2018).

AGREE is a tool designed to quantitatively evaluate the greenness of analytical methods based on the 12 principles of GAC, providing an overall score and visual representation of environmental friendliness. The AGREE tool (accessed via https://agree-index.anvil.app) was applied according to the recommended workflow (Pena-Pereira et al., 2020).

3. Results and Discussion

3.1. Sugar content analysis

The obtained HPLC-ELSD overlaid chromatogram of the standard solutions of Fr, Gl, and Sc (50-2000 µg mL^{-1}) under the optimized conditions is presented in **Figure 2a**, and obtained calibration curves of Fr, Gl, and Sc are shown in **Figure 2b**.
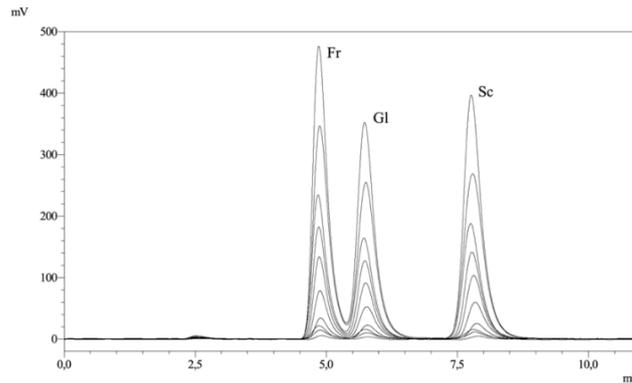

Figure 2a. Overlaid chromatogram of the fructose, glucose and sucrose standards (50-2000 µg mL^{-1})
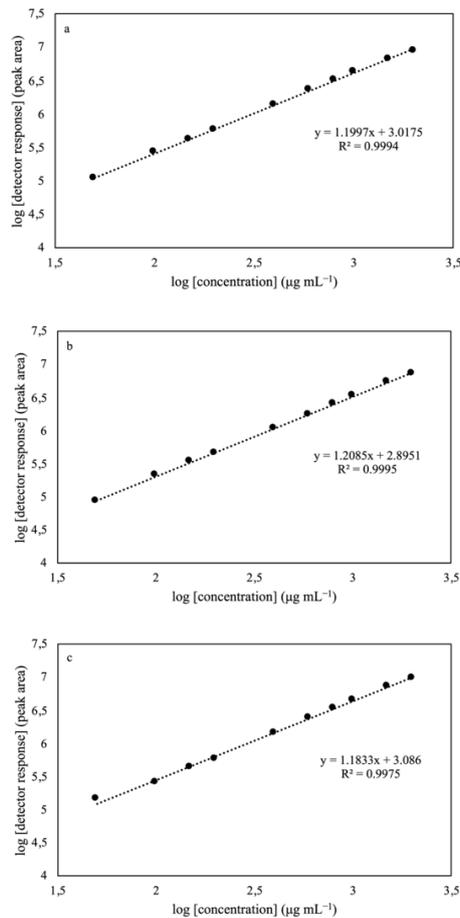

Figure 2b. Obtained calibration curves of fructose (a), glucose (b) and sucrose (c) using HPLC-ELSD (log [concentration] – log [peak area])

Regression equations of the calibration curve and coefficients of determination values of the analytes were reported for Fr, y=1.997x + 3.0175 (R²= 0.9994), for Gl, y=1.2085x + 2.8951 (R²= 0.9995) and for Sc, y=1.1833x + 3.086 (R²= 0.9975). The results showed that good linear relationships were observed within the test range and the resolution of the peaks were above (>2) which is recommended value for acceptable separation.

3.2. Evaluation of sugar amount change of climacteric foods during ripening

HPLC-ELSD analysis was performed on samples of bananas and tomatoes at four different stages of ripening (Green, Ripening, Ripe, Over-ripe) under the optimized conditions. Obtained chromatograms are shown in **Figure 3**a and **Figure 3b**. The amounts of Fr, Gl, and Sc (g/100g) obtained for the tomato and banana samples are presented in **Table 1**.

Table 1. Sugar content of samples at different ripening stages

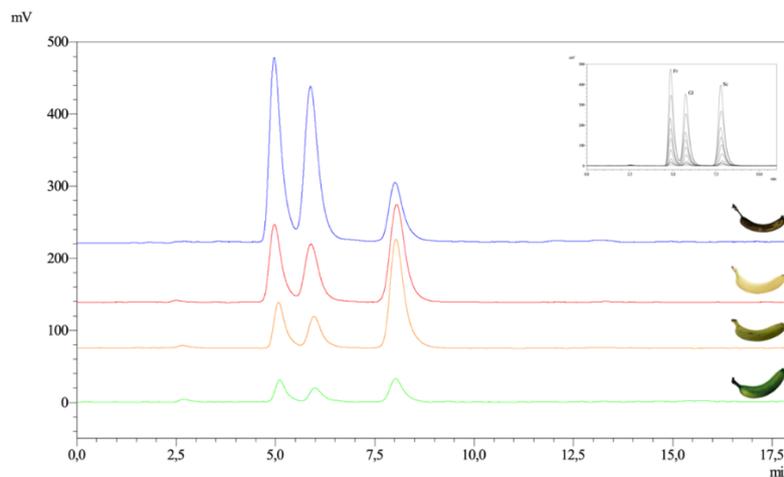| Sample	Stage	Image	Sugar content [g/100 g ± SD (n = 3)]		
			Fr	Gl	Sc
Banana	Green		0.87±0.01	1.27±0.02	1.11±0.01
	Ripening		1.66±0.02	2.57±0.04	4.41±0.02
	Ripe		2.86±0.02	4.83±0.01	4.36±0.01
	Over-ripe		5.76±0.04	11.13±0.11	2.65±0.06
Tomato	Green		4.21±0.01	6.48±0.01	0.74±0.01
	Ripening		4.78±0.04	6.48±0.01	0.38±0.01
	Ripe		4.82±0.01	6.52±0.02	ND
	Over-ripe		4.86±0.02	6.55±0.02	ND

Figure 3a. Overlaid HPLC-ELSD chromatogram of banana samples at different ripening stages
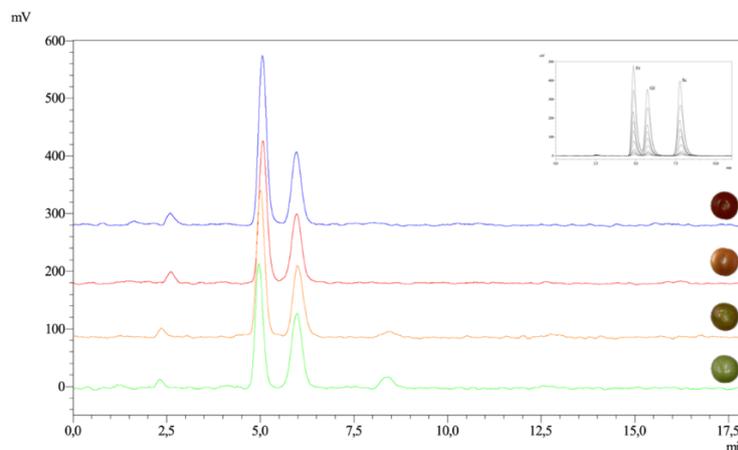

Figure 3b. Overlaid HPLC-ELSD chromatogram of tomato samples at different ripening stages

Statistical analysis was performed using one-way ANOVA followed by Tukey's post-hoc test ($p < 0.05$). In banana samples, Fr, Gl, and Sc contents showed highly significant differences among all ripening stages (ANOVA, $p < 0.0001$ for all sugars). Tukey pairwise comparisons indicated that every stage was significantly different from the others for Fr and Gl, demonstrating a strong and consistent increase during ripening. For Sc, all stage-to-stage transitions were also statistically significant ($p < 0.05$), corresponding to the typical pattern of Sc to monosaccharide conversion in climacteric fruits.

In tomato samples, although the numerical changes were small, both Fr and Gl showed statistically significant differences across stages ($p < 0.0001$ and $p = 0.0015$, respectively). Tukey comparisons revealed that the ripening stage was slightly but significantly different from other stages for Fr. For Gl, the over-ripe stage differed from the earlier stages, whereas green and ripening stages were statistically similar. Sc decreased significantly from green to ripening ($p < 0.05$) but was not detected in later stages, preventing further statistical analysis. These findings indicate that sugar transformations are pronounced in bananas, while tomatoes exhibit statistically detectable but very limited changes.

Upon examination of the sample chromatograms, no interference was detected. Analysis of the results for the banana samples indicated that the levels of fructose and glucose increased with increasing maturity, while sucrose content increased from the green stage to the ripening stage and then decreased in the subsequent stages (Ripe, Over-ripe). This observation supports the notion that starch is first broken down into sucrose and then further converted into fructose and glucose. The total sugar content of the banana samples was found to increase from 3.25 g/100g to 19.54 g/100g. These findings are consistent with those reported in the literature (Adão & Glória, 2005; Adewale et al., 2013). Upon examination of the tomato samples, it was observed that the levels of fructose and glucose increased with ripening, while sucrose content decreased and remained below the detection limit. The total sugar content of the tomato samples was found to increase with ripening. These results are consistent with those reported in the literature (Agius et al., 2018; Gao et al., 2019; Gautier et al., n.d.; Kaur et al., 2006).

NEMI is one of the early tools developed to assess the environmental impact of analytical methods. This system uses a circular symbol divided into four equal segments, with each segment representing a potential environmental hazard. When an analytical method meets certain environmental criteria, the corresponding symbol segment is coloured green. If a chemical used in the method is listed as persistent, bio-accumulative and toxic (PBT) on Environmental Protection Agency (EPA)'s Toxics Release Inventory (TRI) list, it is included in this assessment. In addition, a chemical is considered hazardous if it is on the TRI or the hazardous waste lists (D, F, P, or U) under the Resource Conservation and Recovery Act (RCRA). The method is considered corrosive if the pH of the analysis is below 2 or above 12. However, if the method generates more than 50 grams of waste, it is classified as a waste-generating method (Keith et al., 2007). The NEMI labelling of the applied HPLC-ELSD method is presented in **Figure 4a**. The results show that the methods meet the same green requirements such as PBT, corrosiveness, and waste except for the hazardous criteria section because MeCN does not fall under EPA PBT, D, F, U, or TRI hazardous chemical lists under the specified conditions and quantities.
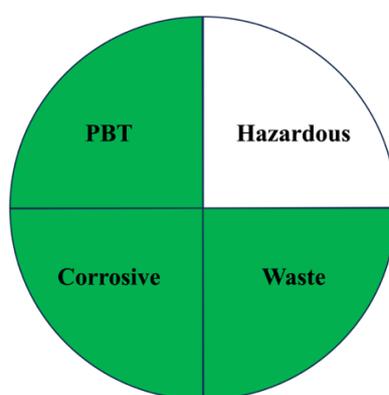

Figure 4a. NEMI pictogram for the applied HPLC-ELSD method

The AES quantifies the environmental impact of an analytical method through a scoring system, deducting penalty points from a maximum of 100. This approach provides a numerical gauge of eco-friendliness, with a perfect green analysis achieving a score of 100. Penalty points are assigned based on factors such as solvent and reagent usage, energy consumption, occupational hazards, waste generation, and waste management strategies. The resulting score reflects the method's overall environmental performance, with higher scores indicating greater sustainability and lower environmental impact. The AES formula, "AES = 100 - total penalty points (PP)," generates a numerical output categorized as excellent (>75 points), acceptable (>50 points), or inadequate (<50 points) green analysis (Gałuszka et al., 2012). A higher score means the analysis is more eco-friendly, showing it has minimized its environmental impact and is more sustainable. Conversely, a lower score indicates a greater negative impact on the environment. The AES points of the applied HPLC-ELSD method are shown in **Table 2**. AES points were found to be as 87 which is classified as excellent green analysis.

Table 2. Analytical Eco-Scale penalty points for HPLC-ELSD

Components	Pictograms	Signal word	Used in method	Penalty Point
Reagent				
MeCN		Danger	< 10 mL (g)	4
Water	No hazard pictogram	-	< 10 mL (g)	-
Instruments				
HPLC-ELSD	-	-	> 1.5 kWh per analysis	2
Sonicator	-	-	≤1.5 kWh per analysis	1
Occupational hazard			Analytical process hermetisation	0
Waste			>10 mL (g) per analysis	5
Total Penalty Point			$\sum$	12
Eco-Scale Score			$100 - \sum$	88

AGREE is a rating system that assesses the environmental sustainability of analytical methods. The results of the assessment are visualised in the form of a circular clock with both a score and a green-yellow-red colour scale for quick and easy understanding. Scores range from 0 to 1, and the closer to 1, the more environmentally friendly the method is. The twelve principles of the AGREE metric are as follows: 1: sampling process, 2: sample amount, 3: instrument layout, 4: sample preparation steps, 5: method miniaturization, 6: derivatisation, 7: waste generation and management, 8: analytical capacity, 9: energy consumption, 10: reagent type, 11: toxic reagent usage, and 12: operator safety. (Pena-Pereira et al., 2020).

The AGREE score for the HPLC-ELSD method used was 0.55 and the weakest criteria were items 3, 7, 9 and 11. Items 3 and 9, shown in red, represent instrument layout and energy consumption ≥1.5 kWh in a single analysis, respectively. Items 7 and 11 are assessed as close to red. Yellow areas represent intermediate scores on the 0-1 scale, typically neither fully green (ideal) nor fully red (unacceptable). For example, if a procedure partially meets a specific criterion, such as waste management (Item 7) or energy consumption (Item 9), but does not reach the ideal level, the corresponding segment is marked in yellow. The AGREE pictogram of the investigated analytical method is presented in **Figure 4b**.
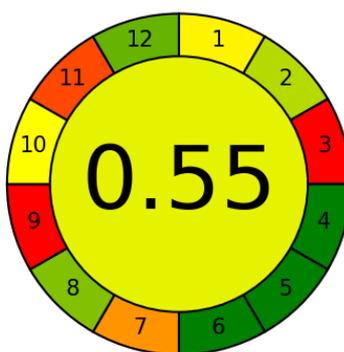

Figure 4b. AGREE pictogram for the HPLC-ELSD method

GAPI, a widely utilized tool for assessing eco-friendliness, was employed to evaluate the HPLC-ELSD analytical method. Represented by a pictogram featuring 15 subcategories, each stage is accompanied by three colour-coded criteria displayed on 5-pointed star symbols: red, yellow, and green. This visual representation offers insights into how well the analytical procedure aligns with green requirements (Płotka-Wasylka, 2018). The GAPI pictogram for applied HPLC-ELSD method is shown in **Figure 4c**. The pentagrams of the method reveal negative environmental impact due to the characteristics of its off-line sample collection, use of non-green solvents or reagents, energy consumption (≥ 1.5 kWh), ≥ 10mL of waste and no treatment for waste, in which case all are coded red.
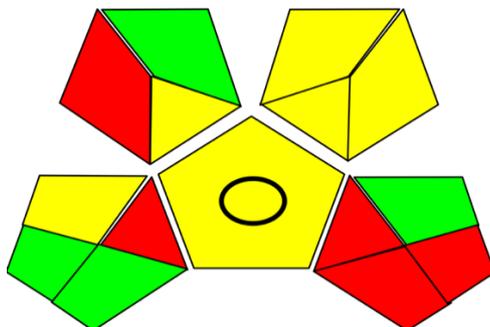

Figure 4c. GAPI pictograms for applied HPLC-ELSD method

4. Conclusions

Understanding the climacteric ripening process in fruits is important for the agricultural industry, as it can help growers and distributors to optimize the timing of harvest and transportation to ensure that the fruit reaches the consumer in the best possible condition. It can also help researchers to develop new methods for prolonging the shelf life of climacteric fruits, such as modified atmosphere packaging, controlled atmosphere storage, and the use of ethylene inhibitors. The ripening stage of fruits plays a significant role in the sugar levels and other nutritional aspects of fruits, as well as their sensory characteristics and overall quality. The ripening stage of fruits is closely associated with changes in sugar levels. During the ripening process, fruits accumulate sugars as a result of the breakdown of starch and other carbohydrates. The type and amount of sugar present in a fruit can influence its flavour, sweetness, and other sensory characteristics. This study allowed the determination of sugar contents in fruits at different ripening stages using HPLC-ELSD analysis. In addition, growing environmental concerns have directed researchers to examine the environmental impact of the conducted analyses. In analytical chemistry, it is important not only to determine the targeted substances but also to investigate their environmental impacts. In our study, the green profile of the applied HPLC-ELSD method was examined through four different greenness metrics along with the determination of major sugar levels varying with the ripening degrees of fruits.

References

Adão, R. C., & Glória, M. B. A. (2005). Bioactive amines and carbohydrate changes during ripening of "Prata" banana (Musa acuminata x M. balbisiana). *Food Chemistry*, 90(4), 705–711. https://doi.org/10.1016/J.FOODCHEM.2004.05.020

Adewale, I. O., Adefila, A., & Adewale, T. B. (2013). Changes in amylase activity, soluble sugars and proteins of unripe banana and plantain during ripening. *Annual Review & Research in Biology*, 34, 678-685.

Agius, C., von Tucher, S., Poppenberger, B., & Rozhon, W. (2018). Quantification of sugars and organic acids in tomato fruits. *MethodsX*, 5, 537–550. https://doi.org/10.1016/J.MEX.2018.05.014

Alferez, F., de Carvalho, D. U., & Boakye, D. (2021). Interplay between abscisic acid and gibberellins, as related to ethylene and sugars, in regulating maturation of non-climacteric fruit. *International Journal of Molecular Sciences*, 22(2), 669. https://doi.org/10.3390/ijms22020669

Chen, Y., Grimplet, J., David, K., Castellarin, S. D., Terol, J., Wong, D. C. J., Luo, Z., Schaffer, R., Celton, J. M., Talon, M., Gambetta, G. A., & Chervin, C. (2018). Ethylene receptors and related proteins in climacteric and non-climacteric fruits. *Plant Science*, 276, 63–72. https://doi.org/10.1016/J.PLANTSCI.2018.07.012

Filip, M., Vlassa, M., Coman, V., & Halmagyi, A. (2016). Simultaneous determination of glucose, fructose, sucrose and sorbitol in the leaf and fruit peel of different apple cultivars by the HPLC–RI optimized method. *Food Chemistry*, 199, 653–659. https://doi.org/10.1016/J.FOODCHEM.2015.12.060

Gałuszka, A., Migaszewski, Z. M., Konieczka, P., & Namieśnik, J. (2012). Analytical Eco-Scale for assessing the greenness of analytical procedures. *TrAC Trends in Analytical Chemistry*, 37, 61-72. https://doi.org/10.1016/j.trac.2012.03.013

Gao, Y., Tian, P., Li, J., Cao, Y., Xu, W., & Li, J. (2019). Transcriptional changes during tomato ripening and influence of brackish water irrigation on fruit transcriptome and sugar content. *Plant Physiology and Biochemistry*, 145, 21–33. https://doi.org/10.1016/J.PLAPHY.2019.10.025

Gautier, H., Diakou-Verdin, V., Bénard, C., Reich, M., Buret, M., Bourgaud, F., Poëssel, J. L., Caris-Veyrat, C., & Génard, M. (n.d.). How Does Tomato Quality (Sugar, Acid, and Nutritional Quality) Vary with Ripening Stage, Temperature, and Irradiance? *Journal of Agricultural and Food Chemistry*, 56(4), 1241-1250. https://doi.org/10.1021/jf072196t

Georgelis, N., Fencil, K., & Richael, C. M. (2018). Validation of a rapid and sensitive HPLC/MS method for measuring sucrose, fructose and glucose in plant tissues. *Food Chemistry*, 262, 191–198. https://doi.org/10.1016/J.FOODCHEM.2018.04.051

Grembecka, M., Lebiedzińska, A., & Szefer, P. (2014). Simultaneous separation and determination of erythritol, xylitol, sorbitol, mannitol, maltitol, fructose, glucose, sucrose and maltose in food products by high performance liquid chromatography coupled to charged aerosol detector. *Microchemical Journal*, 117, 77–82. https://doi.org/10.1016/J.MICROC.2014.06.012

Hussain, C.M., Hussain, G., & Keçili, R. (2025b). Disposable analytical chemistry: Design, application & sustainability. TrAC - Trends in Analytical Chemistry. https://doi.org/10.1016/j.trac.2025.118414.

Ivancic, T., Jakopic, J., Veberic, R., Vesel, V., & Hudina, M. (2022). Effect of Ripening on the Phenolic and Sugar Contents in the Meso- and Epicarp of Olive Fruits (Olea europaea L.) Cultivar 'Leccino.' *Agriculture* (Switzerland), 12(9), 1347. https://doi.org/10.3390/AGRICULTURE12091347/S1

Jaglan, P., Buttar, H. S., Al-bawareed, O. A., & Chibisov, S. (2022). Potential health benefits of selected fruits: apples, blueberries, grapes, guavas, mangos, pomegranates, and tomatoes. *Functional Foods and Nutraceuticals in Metabolic and Non-Communicable Diseases*, 359–370. https://doi.org/10.1016/B978-0-12-819815-5.00026-4

Kaur, D., Sharma, R., Wani, A. A., Gill, S., & Sogi, D. S. (2006). Physicochemical Changes in Seven Tomato (Lycopersicon esculentum) Cultivars During Ripening. *International Journal of Food Properties*, 9(4), 747–757. https://doi.org/10.1080/10942910600575716

Keith, L. H., Gron, L. U., & Young, J. L. (2007). Green analytical methodologies. *Chemical reviews*, 107(6), 2695-2708. https://doi.org/10.1021/cr068359e

Kim, H. Y., Farcuh, M., Cohen, Y., Crisosto, C., Sadka, A., & Blumwald, E. (2015). Non-climacteric ripening and sorbitol homeostasis in plum fruits. *Plant Science*, 231, 30–39. https://doi.org/10.1016/J.PLANTSCI.2014.11.002

Klee, H., genetics, J. G.-A. review of, & 2011, undefined. (2011). Genetics and control of tomato fruit ripening and quality attributes. *Annual Reviews*, 45, 41–59. https://doi.org/10.1146/annurev-genet-110410-132507

Kou, X., Feng, Y., Yuan, S., Zhao, X., Wu, C., Wang, C., & Xue, Z. (2021). Different regulatory mechanisms of plant hormones in the ripening of climacteric and non-climacteric fruits: a review. *Plant Molecular Biology*, 1-21. https://doi.org/10.1007/s11103-021-01199-9

Kou, X., & Wu, M. (2018). Characterization of climacteric and non-climacteric fruit ripening. *Methods in Molecular Biology*, 1744, 89–102. https://doi.org/10.1007/978-1-4939-7672-0_7/COVER

Liu, R. H. (2003). Health benefits of fruit and vegetables are from additive and synergistic combinations of phytochemicals. *American Journal of Clinical Nutrition*, 78 (3). https://doi.org/10.1093/AJCN/78.3.517S

Ma, C., Sun, Z., Chen, C., Zhang, L., & Zhu, S. (2014). Simultaneous separation and determination of fructose, sorbitol, glucose and sucrose in fruits by HPLC–ELSD. *Food Chemistry*, 145, 784–788. https://doi.org/10.1016/J.FOODCHEM.2013.08.135

Maduwanthi, S. D. T., & Marapana, R. A. U. J. (2017). Biochemical changes during ripening of banana: A review. *International Journal of Food Science and Nutrition*, Vol.2 (5), pp. 166-169. http://dr.lib.sjp.ac.lk/handle/123456789/7777

Mojsiewicz-Pieńkowska, K. (2009). On the issue of characteristic evaporative light scattering detector response. *Critical Reviews in Analytical Chemistry*, 39(2), 89–94. https://doi.org/10.1080/15389580802570218

Nogueira, L. C., Silva, F., Ferreira, I. M. P. L. V. O., & Trugo, L. C. (2005). Separation and quantification of beer carbohydrates by high-performance liquid chromatography with evaporative light scattering detection. *Journal of Chromatography A*, 1065(2), 207–210. https://doi.org/10.1016/J.CHROMA.2004.12.074

Payasi, A., Biochemistry, G. S.-J. of food, & 2010, undefined. (2010). Ripening of climacteric fruits and their control. *Journal of food Biochemistry*, 34(4), 679-710. https://doi.org/10.1111/j.1745-4514.2009.00307.x

Pena-Pereira, F., Wojnowski, W., & Tobiszewski, M. (2020). AGREE—Analytical GREEnness metric approach and software. *Analytical chemistry*, 92(14), 10076-10082. https://doi.org/10.1021/acs.analchem.0c01887

Płotka-Wasylka, J. (2018). A new tool for the evaluation of the analytical procedure: Green Analytical Procedure Index. *Talanta*, 181, 204-209. https://doi.org/10.1016/j.talanta.2018.01.013

Ruiz-jimé Nez, J., Priego-capote, F., & Dolores Luque De Castro, M. A. (2010). Qualitative and Quantitative Sugar Profiling in Olive Fruits, Leaves, and Stems by Gas Chromatography-Tandem Mass Spectrometry (GC-MS/MS) after Ultrasound-Assisted Leaching. *J. Agric. Food* Chem, 58, 12292–12299. https://doi.org/10.1021/jf102350s

Singh, B., Singh, J. P., Kaur, A., & Singh, N. (2016). Bioactive compounds in banana and their associated health benefits - A review. *Food Chemistry*, 206, 1–11. https://doi.org/10.1016/J.FOODCHEM.2016.03.033

Sezgin, B., Arli, G., & Soyseven, M. (2025). Multi-metric greenness, performance and applicability evaluation of a green HPLC-ELSD method for polysorbate 80 determination in various pharmaceuticals compared with AI-assisted scoring systems. Microchemical Journal, 218. https://doi.org/10.1016/j.microc.2025.115710.

Soyseven, M., Sezgin, B., Acar, E., & Arli, G. (2023). Green, Rapid, Robust Determination of Aspartame in Beverages by High-Performance Liquid Chromatography-Evaporative Light Scattering Detection (HPLC-ELSD): Method Development and Validation. *Analytical Letters*, 56(11), 1773–1783. https://doi.org/10.1080/00032719.2022.2146129

Soyseven, M., Sezgin, B., & Arli, G. (2022). A novel, rapid and robust HPLC-ELSD method for simultaneous determination of fructose, glucose and sucrose in various food samples: Method development and validation. *Journal of Food Composition and Analysis*, 107. https://doi.org/10.1016/J.JFCA.2022.104400

Subedi, P. P., & Walsh, K. B. (2011). Assessment of sugar and starch in intact banana and mango fruit by SWNIR spectroscopy. *Postharvest Biology and Technology*, 62(3), 238–245. https://doi.org/10.1016/J.POSTHARVBIO.2011.06.014

Tsai, Y. H., Tsai, C. W., & Tipple, C. A. (2022). A validated method for the analysis of sugars and sugar alcohols related to explosives via liquid chromatography mass spectrometry (LC-MS) with post-column addition. *Forensic Chemistry*, 28. https://doi.org/10.1016/J.FORC.2022.100404

Yumbya, P., Ambuko, J., Hutchinson, M., Owino, W., Juma, J., Machuka, E., & Mutuku, J. M. (2021). Transcriptome analysis to elucidate hexanal's mode of action in preserving the post-harvest shelf life and quality of banana fruits (Musa acuminata). *Journal of Agriculture and Food Research*, 3. https://doi.org/10.1016/J.JAFR.2021.100114