



Greenness of procedures using NADES in the preparation of vegetal samples: Comparison of five green metrics

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ARTICLE INFO

Keywords:

Natural deep eutectic solvent (NADES)
National environmental methods index (NEMI)
Green analytical procedure index (GAPI)
Analytical Eco-scale
Analytical GREENness (AGREE)
White analytical chemistry (WAC)

ABSTRACT

The principles of green analytical chemistry have led to the development of analytical procedures that are increasingly sustainable. Different metrics have been created for the evaluation of greenness, although determination of the green nature of new analytical methods remains challenging, including for extraction methods that involve the use of natural deep eutectic solvents (NADES). In this study, the following five chemical metrics for the evaluation of greenness were considered: National Environmental Methods Index (NEMI), Green Analytical Procedure Index (GAPI), Analytical Eco-Scale, Analytical GREENness (AGREE), and White Analytical Chemistry (WAC). These methods were applied in evaluation of the environmental and sustainability characteristics of three different methods for the preparation of plant material samples: microwave-assisted extraction (MAE), ultrasound-assisted extraction (UAE), and microwave-assisted acid digestion (MW-AD). These methods employed different NADES as extraction solvents, as well as dilute nitric acid as an oxidizing agent, for the determination of As, Cd, Pb, and V by inductively coupled plasma mass spectrometry (ICP-MS). The NEMI metric found no differences between the MAE-NADES and UAE-NADES methods. The GAPI metric found differences between the MAE-NADES and UAE-NADES methods and identify the disadvantageous aspects of each step of the methods. The Analytical Eco-Scale and AGREE identified the MAE-NADES method as the greenest, while WAC-12 RGB indicated the UAE-NADES method as the greenest procedure. A detailed discussion is provided of the application of each metric, together with their differences and advantages.

Introduction

Green analytical chemistry (GAC) now plays an essential role in the development of analytical procedures considered to be sustainable. Currently, one of its main priorities is to reduce or eliminate the use of products, byproducts, and solvents toxic to human health and the environment [1,2]. To this end, optimized analytical procedures have been developed that enable reductions in the consumption of reagents and energy, while providing high sensitivity, precision, and accuracy, which are essential for the validation of analytical methods [3,4]. The analysis of elements present at low concentrations requires highly sen-

sitive analytical techniques, such as plasma-based methods (inductively coupled plasma optical emission spectroscopy (ICP-OES) and inductively coupled plasma mass spectrometry (ICP-MS)) [5], which necessarily require sample preparation procedures to be performed for solubilization of the analytes of interest [6].

Microwave-assisted acid digestion (MW-AD) is a common and efficient methodology adopted for the preparation of plant samples, which generally employs nitric acid (HNO₃) and other oxidants, with the advantage that closed vials are used, minimizing analyte losses and contamination [7]. However, this results in samples with high residual acidity, which can damage the sample introduction system and increase the

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<https://doi.org/10.1016/j.talo.2022.100131>

Received 31 May 2022; Received in revised form 14 July 2022; Accepted 16 July 2022
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likelihood of physical interferences in plasma-based analytical techniques. Furthermore, the use of concentrated HNO_3 does not comply with the GAC requirement to avoid the generation of corrosive and toxic residues [7].

Despite the effectiveness of using MW-AD with diluted HNO_3 for sample preparation [8,9], as an alternative in accordance with GAC, without compromising the efficiency of sample preparation by MW-AD, the development of green solvents remains one of the main objectives of GAC [10]. Deep eutectic solvents (DES) are a promising class of green solvents used in many different applications including electroanalysis, synthesis and modification of sorbent materials, and sample extraction [11]. DES are mixtures formed by interactions between a hydrogen bond donor compound (HBD) and a hydrogen bond acceptor compound (HBA) [12], combined with other intermolecular interactions such as van der Waals and electrostatic forces [13]. Natural deep eutectic solvents (NADES) can be obtained using abundant natural compounds as DES precursors, including organic acids, sugars, amino acids, and choline derivatives [14]. The main advantages of both classes of green solvents are biodegradability, low toxicity, low cost, simple preparation, negligible volatility, and especially their adjustable physicochemical properties (density, viscosity, and polarity) [15].

Both DES and NADES have been used in green sample preparation methods, focusing on achieving high precision in the results and prioritizing the sustainability of the processes. Among these methods, microwave-assisted extraction (MAE) and ultrasound-assisted extraction (UAE) can be highlighted. Both techniques have important advantages for GAC, notably short extraction times, low consumption of reagents and solvents, safe operation, and low consumption of electrical energy [16,17]. These methodologies have been used for the determination of organic analytes [18] and have shown promise for the determination of inorganic analytes in plant and biological tissue samples [19,20]. A crucial issue in GAC is the evaluation of the extent to which a process or product is environmentally friendly [21]. For this, green metrics can assist in measuring and comparing the sustainable characteristics of novel analytical procedures [2].

In this work, the green characteristics of different analytical procedures were evaluated using five selected metrics: National Environmental Methods Index (NEMI) [22], Green Analytical Procedure Index (GAPI) [23], Analytical Eco-Scale [24], Analytical GREENness (AGREE) [25], and White Analytical Chemistry (WAC) [26]. The selection of these metrics was based on their applicability to analytical sample preparation procedures, together with their relevance, as evidenced by the citations found in the Web of Science® (WoS) database (data reported up to July 7th 2022): NEMI (337), AMVI (43) [27], HPLC-EAT (80) [28], MCDA (176), Analytical Eco-Scale (530), GAPI (338), AGREE (148), and WAC (38). Some metrics available in the literature are specific to chromatographic procedures, examples being Analytical Method Volume Intensity (AMVI) and HPLC-EAT [2]. It was not possible to access the algorithm of the Ranking Organization Method for Enrichment Evaluations (PROMETHEE) tool, which has been used in multi-criteria decision analysis (MCDA) to select analytical procedures that have low environmental impacts [29]. It should be noted that the AGREE and WAC-12 RGB tools were only published recently, in 2020 and 2021, respectively, which was reflected in the low number of citations, compared to other more established metrics. Nonetheless, despite being recent techniques, their relevance could be seen from the significant number of citations obtained within a short period.

The main objective of the present work was to evaluate, using different metrics (NEMI, GAPI, Analytical Eco-Scale, AGREE, and WAC), the environmentally friendly characteristics of three different methods (MAE, UAE, and MW-AD) used to prepare samples of plant material, prior to the determination of trace elements by ICP-MS. The extracting solvents used were different NADES prepared using xylitol, malic acid, and citric acid, as well as nitric acid as an oxidizing solvent. The evaluation was performed using the green metrics presented above, which

were submitted to a critical comparison, identifying their main advantages and disadvantages.

Procedures

Reagents and samples

All the materials used in the syntheses were previously decontaminated for 24 h in acid baths containing 10% v v⁻¹ HNO_3 , followed by washing three times with ultrapure deionized water.

The NADES were synthesized using citric acid (99.5% purity, Sigma-Aldrich, MO, USA), DL-malic acid (99% purity, Sigma-Aldrich), xylitol (99% purity, Sigma-Aldrich), and ultrapure deionized water (18 M Ω cm) obtained from a Milli-Q purification system (ICW-3000, Merck KGaA, Darmstadt, Germany). The sample used was a forage grass (*Brachiaria brizantha* cv. Marandu; EMBRAPA, FO-01/2012) reference material produced by the Brazilian Agricultural Research Corporation (Embrapa).

The microwave-assisted acid digestion (MW-AD) experiments used HNO_3 (Sigma-Aldrich), previously purified using a sub-boiling distillation system (subCLEAN PTFE, Milestone, BG, Italy), and 30% v v⁻¹ H_2O_2 (Sigma-Aldrich).

Preparation of the NADES

Three different types of NADES were synthesized according to a ratio of 42:13:45 (% w w⁻¹), with combinations of the following components: citric acid/xylitol/water (XYL-CA), malic acid/xylitol/water (XYL-MA), and citric acid/malic acid/water (MA-CA). The mixture design combinations can be found in a previous work [20].

The preparation of the NADES was based on the method described by Dai et al. (2013) [13]. The mixtures were placed in a beaker fitted with a cap and were heated in a water bath for 2 h, at 50°C, under magnetic stirring at 220 rpm (AccuPlate Hotplate Stirrer, Labnet, Edison, NJ, USA). The synthesis time was adjusted in order to obtain a clear liquid. The resulting solvents were stored in desiccators, at room temperature [13,20].

Microwave-assisted acid digestion (MW-AD)

Masses of 150 mg of plant sample were digested in closed flasks, using 6.0 mL of HNO_3 (50% v v⁻¹) and 2.0 mL of H_2O_2 (30% v v⁻¹). The microwave (Multiwave PRO, Anton Paar GmbH, Graz, Austria) heating program employed three steps, with an initial ramp to 190°C (10 min), holding at 190°C (40 min), and cooling to 50°C (10 min). The volumes of the samples and blanks were made up with ultrapure water, followed by analysis using ICP-MS. The procedure was performed in triplicate [20,30].

Ultrasound-assisted extraction (UAE) with NADES

Masses of 90 mg of plant sample were transferred to polypropylene flasks containing 9 mL of NADES. The resulting suspensions were treated for 45 min in an ultrasonic bath (Model Q3.0/40A, Eco-Sonics, Indaiatuba, São Paulo, Brazil) operated at 40 kHz. Volumes of 6.0 mL of ultrapure water were then added and the mixtures were centrifuged at 4000 rpm for 10 min. The supernatants were separated by filtration and analyzed by ICP-MS. The procedure was performed in triplicate [19,20].

Microwave-assisted extraction (MAE) with NADES

Masses of 90 mg of plant sample and 9 mL volumes of NADES were placed in a closed vessel and subjected to microwave irradiation (Model Multiwave PRO, Anton Paar GmbH, Graz, Austria). The heating pro-

gram consisted of three steps, with (I) a ramp to 100°C (2 min), (II) holding at 100°C (18 min), and (III) cooling to 50°C (10 min). The suspensions were filtered and the supernatants were analyzed by ICP-MS. The procedure was performed in triplicate [20].

Metrics for evaluation of the sustainability of analytical methods

This section provides a description of the metrics used to assess the sample preparation methods that used NADES based on xylitol, malic acid, and citric acid as solvents for the extraction of As, Cd and Pb from plant samples. The extraction methods employed were MAE (MAE-NADES), UAE (UAE-NADES), and MW-AD (nitric acid). To evaluate the greenness of the methods, the energy consumption and waste generation values considered data referring to the extraction and the determination by ICP-MS. The following total values for energy consumption and waste generation were adopted, respectively, for each method evaluated, per sample: 10 mL and 0.3 kWh for MW-AD, 10 mL and 0.2 kWh for MAE, and 16 mL and 0.1 kWh for UAE.

NEMI

The NEMI tool employs a pictogram consisting of a circle divided into four sectors, as follows: 1 – PBT (persistent, bioaccumulative, and toxic); 2 – Hazardous; 3 – Corrosive; and 4 – Waste. Each region of the circle is filled in green or white colors, according to the criteria defined by Keith et al. (2007) [22], with a higher number of green sectors indicating greater sustainability of the method.

GAPI

The GAPI metric considers the green character of each step of the analytical process, including sampling, transport, storage, sample preparation, reagents, solvents, and other resources employed to obtain the final result. A pictogram is obtained, consisting of a pentagon with four other pentagons arranged on the edges, except at the base. Green, yellow, and red colors indicate low, medium, and high environmental impacts, respectively, for each step of the analytical method. Hence, a green field in the pictogram shows compliance of the procedure with the established requirements. Each field in the pictogram contributes to the sustainability assessment [23,31].

The five pentagrams are subdivided into 15 categories: A - Sample preparation, considering (1) type of sample collection, (2) preservation, (3) transport, (4) storage, (5) method type (direct or indirect), (6) extraction scale, (7) reagents consumption, and (8) additional treatments; B - Properties of the solvents and reagents, considering (9) quantity, (10) health hazards, and (11) safety hazards; C - Instrumental parameters, considering (12) energy expenditure, (13) occupational risks, (14) waste generation, and (15) waste treatment; and D - Quantitative indicator, where its presence in the circle in the central pentagon means that the method is quantitative, while its absence indicates that the method is qualitative [23].

Analytical eco-scale

The Analytical Eco-Scale technique is based on the allocation of a total of 100 points for an ideal green analysis. Penalty points are subtracted from this total, considering the various aspects of the analytical procedure. These include the quantity and risk (physical, environmental, and human health) of each reagent/solvent used; the waste energy of the equipment used for sample preparation and analysis; the risk associated with the analytical procedure (degree of hermetic sealing, generation of hazardous vapors); and the generation of chemical wastes, considering the volume produced, its risks, and its characteristics (recyclable, degradable, or with no intended treatment). In this way, a score is assigned to the proposed methodology, as shown in Eq. 1, in accordance with Gałuzka et al. (2012) [24].

Values higher than 75 indicate an excellent green analysis, while values in the range $75 < x < 50$ reflect an acceptable green analysis, and values below 50 indicate a poor green analysis [20].

$$\text{AnalyticalEco} - \text{Scale} = 100 - \text{total penalty} \quad (\text{Equation 1})$$

AGREE

The open source AGREE software (freely available at <https://mostwiedzy.pl/AGREE>), developed by the University of Vigo and the Gdansk University of Technology, enables data analysis and generation of a graph using scores on a scale from 0 to 1 for each of the 12 GAC principles. A green method is considered to be reflected by an overall score higher than 0.6. The performance for each of the 12 GAC principles is shown using a red-yellow-green color scale, indicating the strengths and weaknesses of the analytical method. The width of the segment corresponding to each individual GAC principle shows its impact on the final score for the method [25].

White analytical chemistry (WAC)

The White Analytical Chemistry (WAC) tool emerged as an extension of GAC, adopting the concepts of social and economic development, together with environmental protection, as three requirements for sustainable development. This method uses a graphical representation with three columns and associated color scales: 1) red, concerning the efficiency of the analytical method; 2) green, related to the environment and safety; and 3) blue, related to practical and economic aspects. These features constitute the metric called WAC-12 RGB, where it is considered that the color white, reflecting the synergistic effects of the three primary colors, indicates an ideal analytical method [26]. The WAC metric employs the RGB 12 algorithm, freely available as a spreadsheet, enabling rapid and simple evaluation of the whiteness of the method and its degree of sustainability. The 12 principles of WAC are arranged in three columns, each associated with four principles (Nowak et al., 2021) [26]: column 1 (red) - R1 (application scope), R2 (detection limits and quantification limits), R3 (precision), and R4 (accuracy); column 2 (green) - G1 (toxicity of reagents), G2 (number and amount of reagents and wastes), G3 (energy), and G4 (direct impacts); and column 3 (blue) - B1 (cost efficiency), B2 (time efficiency), B3 (requirements), and B4 (operational simplicity).

Results and discussion

According to Santana et al. (2020) [20], the MW-AD method provided recoveries in the range 80-120% for Pb and As in a sample of forage grass. The use of UAE with NADES achieved acceptable recoveries (80 and 120%) for As, Cd, and Pb. The use of UAE with the XYL-CA NADES provided acceptable recoveries for As, Cd, and Pb, while UAE with the MA-CA NADES only provided satisfactory results for determination of Pb in the plant material. The use of MAE with NADES led to good recoveries (80 and 120%) of As, Cd, and Pb. The use of MAE with the XYL-CA NADES showed satisfactory recoveries for As and Cd, while MAE with the MA-CA NADES provided satisfactory recoveries of As, Cd, and Pb. Therefore, for the application of the green assessment tools, the data for As and Pb were considered for MW-AD, while for MAE and UAE, the data for As (NADES XYL-CA) and Pb (NADES MA-CA) were used.

When the NEMI metric was used, the four quadrants were green for the MAE-NADES and UAE-NADES methods, while in the case of MW-AD, the quadrant related to the corrosivity criterion was not satisfactory, due to the use of nitric acid (Fig. 1). It was evident that NEMI was the least informative tool for evaluation of greenness, being limited, as pointed out by Gamal et al. (2021) [32], by the fact that in addition to being restricted to purely qualitative criteria, it does not provide a de-

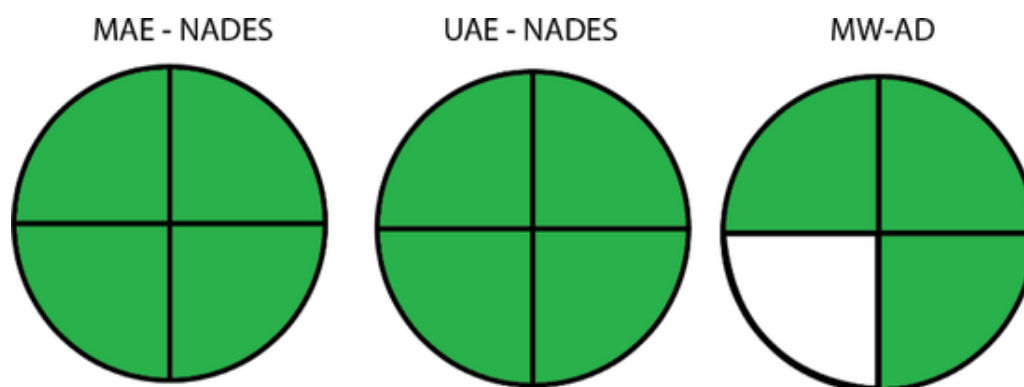


Fig. 1. Use of NEMI for comparison of the greenness profiles of three different extraction methods employing NADES and nitric acid as extracting solvents.

tailed visualization of the analytical process appraisal. Nonetheless, it was clear that the use of HNO_3 made the process less green, compared to the other two extraction methods.

The GAPI tool was proposed for the evaluation of new analytical protocols, considering the requirements of GAC [23,31]. Its use here enabled an assessment that was more comprehensive, compared to NEMI, with additional information provided about the methods, ranging from the sampling step to the final stages of analysis. Significant differences were observed among the MAE-NADES, UAE-NADES, and MW-AD methods. The pictograms for the MAE-NADES and UAE-NADES methods were similar (Fig. 2), but showed differences for parameters 12 and 14, related to energy expenditure per sample and the quantity of waste generated. The use of UAE led to a larger volume of waste (> 10 mL per sample), but required less energy during the preparation process. The methods employing NADES showed greater differences, relative to MW-AD, for parameter 7, solvent and reagent properties (parameters 10 and 11), and instrumental properties (parameters 12 and 14).

In the pictogram obtained for the MW-AD method, the greater number of red and yellow sectors, compared to the pictograms for the NADES methods, indicated disadvantages from the environmental perspective. Considering the axis corresponding to the reagents (parameters 10 and 11), the better performances of the MAE-NADES and UAE-NADES methods could be explained by the fact that they employed reagents of natural origin. In the case of the axis corresponding to the instrumental properties, the MAE and MW-AD methods consumed 15 times more energy per analysis (parameter 12), compared to UAE, with values ≤ 1.5 kWh and ≤ 0.1 kWh, respectively. When microwave radiation was used, this factor was compensated by the ability to digest from 4 to 12 samples in one analytical run, while the ultrasound method could only process one sample at a time in the equipment used in this work. The generation of waste (parameter 14) varied among the methods, with lower values for the microwave method (1–10 mL per sample) and higher values using ultrasound for extraction (> 10 mL per sample). It should be noted that the use of NADES as extracting solvent would enable possible recycling of the waste (parameter 15). The visual presentation of the GAPI metric is an efficient way to compare procedures,

allowing researchers to make their own decisions on the criteria considered important for green methods [23,31].

The Analytical Eco-Scale, which offers a semi-quantitative approach for greenness assessment, enables a more rigorous evaluation of the steps of the procedure [24]. The scores obtained were 76 per sample for MW-AD, 94 per sample for MAE-NADES, and 93 per sample for UAE-NADES. The MW-AD method had 24 penalty points, due to the use of HNO_3 (4 points) and H_2O_2 (4 points), generation of acid vapor (3 points), generation of acid residue (5 points), high energy expenditure per sample (2 points), lack of treatment of the acid residue (3 points), use of ICP-MS (1 point), and safety concerns (2 points). The MAE-NADES method had 6 penalty points, due to the use of NADES solvents (1 point), generation of waste up to 10 mL (3 points), use of ICP-MS (1 point), and moderate energy consumption ≤ 1.5 kWh (1 point). The UAE-NADES method had 7 penalty points, due to the use of NADES (1 point), use of ICP-MS (1 point), and generation of waste greater than 10 mL (5 points). Therefore, the UAE-NADES and MAE-NADES methods provided excellent green analyses, while the MW-AD method was classified as an acceptable green procedure.

The Analytical Eco-Scale was developed as a flexible tool, where the penalty attribution weightings can be modified according to the perception of the analyst [24]. This can lead to a certain degree of complexity in the use of this tool, since it requires a trained professional with understanding of the entire process and capable of a critical analysis that considers the evaluation criteria for each of the stages of an analytical method.

According to the AGREE tool, the MAE-NADES method was greener than UAE-NADES and MW-AD (Fig. 3). The energy consumption calculation used the predefined selection of the tool (which has an option for selection of methods saved in the software) for the most energy-intensive equipment used. A small advantage of MAE-NADES, compared to UAE-NADES, was related to the quantity of waste generated, analytical frequency, and number of analytes detected in the same analysis. The MAE-NADES method generated 10 mL of waste, with 12 samples extracted per run, and allowed simultaneous detection of up to 3 analytes. The UAE-NADES method generated 16 mL of waste, with 1

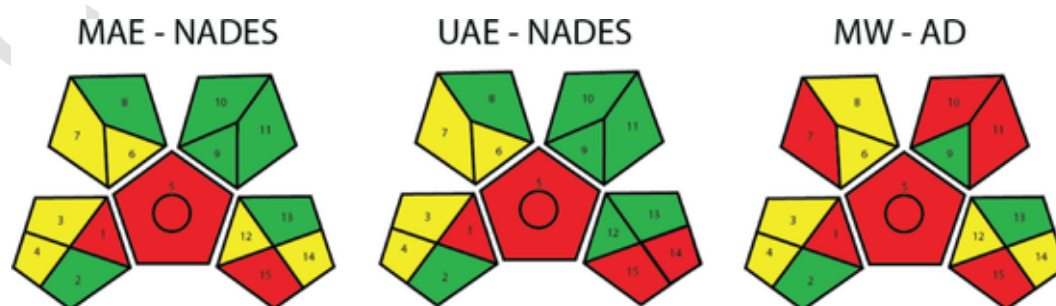


Fig. 2. Use of GAPI for comparison of the greenness profiles of three different extraction methods employing NADES and nitric acid as extracting solvents.

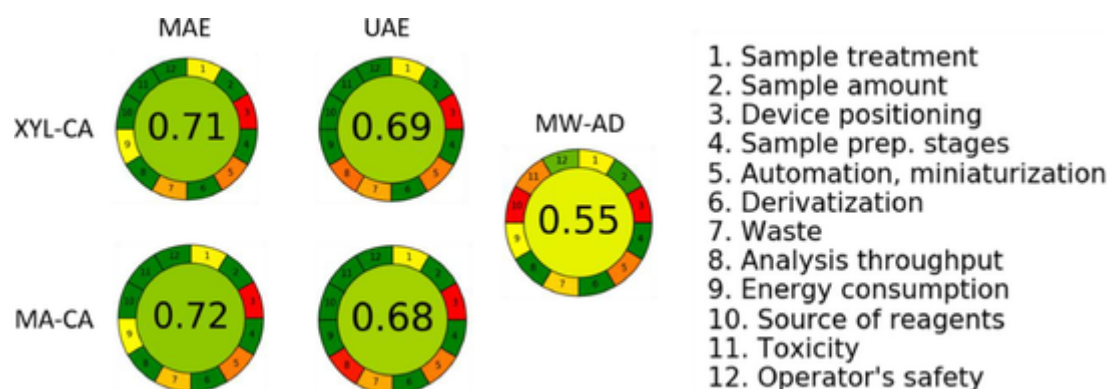


Fig. 3. Use of AGREE for comparison of the greenness profiles of three different extraction methods employing the NADES (XYL-CA and MA-CA) and nitric acid as extracting solvents.

sample extracted per run and simultaneous detection of up to 3 analytes. The MW-AD method had penalty points awarded due to the use of a hazardous reagent (nitric acid), generation of harmful waste, and concerns over operator safety. Comparison between the solvents for the same method showed a small difference in the scores, due to the difference in the number of analytes detected simultaneously (criterion 8). This difference was evident in the values obtained for the NADES XYL-CA (3 analytes) and MA-CA (1 analyte), for both extraction methods (Fig. 3).

The WAC-12 RGB tool provides a more detailed appraisal of the method steps, so evaluation of the solvents necessitated specific application of this metric for each analyte. For the purpose of comparison, the analytes As and Pb were selected, since they could be determined by the three methods and using the two NADES solvents. Application of the WAC-12 RGB tool resulted in the highest scores for the UAE method performed with the two different solvents. The use of UAE with the XYL-CA NADES was the greenest among the methods evaluated, with an overall score of 83.2. Graphs with the detailed scores for each method and solvent combination are shown in Fig. 4.

Use of the WAC-12 RGB tool to compare the MW-AD, MAE-NADES, and UAE-NADES sample preparation methods indicated that the UAE

method was greener and more sustainable, compared to the use of microwave irradiation. The individual analysis of the groups enabled identification of the criteria that contributed the most to differentiation among the methods. UAE-NADES presented higher scores in the three groups, so it was superior in terms of analytical and ecological criteria, as well as economic factors. MAE-NADES had lower scores in the red and green WAC groups, due to higher limits of detection and quantification, together with recovery values at the extremes of the acceptable range (from 80 to 120%). The MW-AD method clearly presented the lowest greenness, due to the use of nitric acid, as well as lower scores related to the sensitivity of the technique. The MW-AD and MAE-NADES methods had the lowest scores, which was due to their high cost, requirement for a qualified operator, moderate capacity for automation, and absence of portability.

Comparison of the use of the XYL-CA and MA-CA NADES showed that they were similar in terms of the environmental and economic parameters evaluated, with differences only in parameters of the blue group, related to the sensitivity and accuracy of the methods employed.

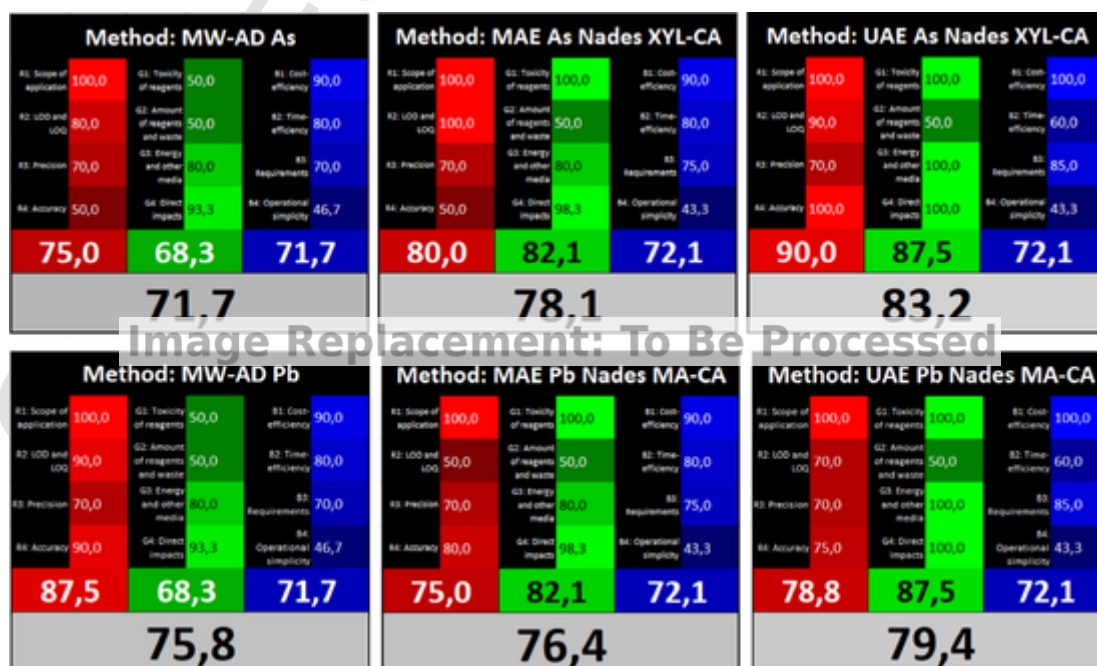


Fig. 4. Comparison of the greenness profiles obtained using the WAC-12 RGB tool applied to the three different extraction methods, with the NADES (XYL-CA and MA-CA) and nitric acid as extracting solvents.

4. Conclusions

The five metrics for evaluating the greenness of sample preparation methods, employing MAE and UAE with NADES solvents, and MW-AD with nitric acid, provided different results in terms of the greenest method. All the tools identified the MW-AD method as being the least green, which was mainly due to its use of nitric acid. Nonetheless, it should be noted that the high temperature and pressure employed in the MW-AD method increases the efficiency of decomposition of the organic matter present in samples such as plant material, while its use with dilute nitric acid solutions makes the process more user-friendly and with wide applicability to different matrices and analytes.

The NEMI metric found no differences between the MAE-NADES and UAE-NADES methods. The GAPI metric found differences between the MAE-NADES and UAE-NADES methods and identify the disadvantageous aspects of each step of the procedure. The Analytical Eco-Scale and AGREE tools identified the MAE-NADES method as being greenest, while use of the WAC-12 RGB tool resulted in the UAE-NADES method presenting the best score. This difference was mainly due to the effects of the energy consumption criterion in the calculations performed by the different tools. The use of WAC-12 RGB also enabled the identification of differences between the solvents, with the XYL-CA NADES being shown to provide greater greenness, sensitivity, and precision.

Comparison of the five metrics showed that NEMI was limited in its ability to evaluate the greenness of the sample preparation methods. It was the least efficient tool for evaluating and comparing the greenness of the MAE and UAE methods using NADES as extracting solvents, and MW-AD using nitric acid, since the criteria adopted were unable to identify differences at a sufficient level of detail.

The GAPI and Analytical Eco-Scale tools are easy to use and can provide more information concerning the environmental impacts of the methods evaluated. Since the former is semi-quantitative and the latter is quantitative, they can be used together as tools for method evaluation. In this way, the Analytical Eco-Scale enables quantification of the effects involving the amounts of reagents and samples, time, energy consumption, and volume of waste, while GAPI enables visual assessment and an overview of all the stages of an analytical procedure.

The AGREE software satisfies the two requirements mentioned above, since it is quantitative and also allows qualitative and visual analysis, in addition to considering all 12 of the GAC principles. The application of AGREE is somewhat more complex, compared to the methods described above, because, as an additional advantage, it allows the weighting of each evaluation criterion, according to the aims of the analyst.

The WAC-12 RGB tool can be understood as the most complete of the different metrics for evaluation of sample preparation methods, since it considers the fundamental tenets of sustainable development and includes criteria that integrate environmental protection, operator health, and the efficiency and cost-effectiveness of analytical techniques. Due to the greater quantity of information to be considered in the evaluation, as well as the need to allocate weightings to the different criteria, the use of this tool requires a qualified analyst able to understand the entire analytical procedure, in order to make the best decision in selection of greener methods.

With the exception of NEMI, all the tools presented here, with different levels of complexity, were found to be useful for evaluating and comparing the greenness of sample preparation methods, according to the objectives of the evaluator, and were able to include the main GAC principles. The tools differ from each other, since they contain more or less detail for each aspect evaluated. Therefore, it is up to the analyst to select the best metric to be used, according to the requirements and conditions. It should be noted that this is the first work to apply these five metrics for evaluation and comparison of the greenness of sample preparation methods employing NADES.

Green analytical chemistry plays an important role in the development of more sustainable analytical methods, with different metrics being available for assessment of the greenness of methods, employing a variety of parameters. Gaps exist in the metrics, although all those described here are suitable for use in assessment of the environmental impact of an analytical method, with selection of the best metric depending on the objective of the evaluation and the practical requirements.

Funding

The authors are grateful for the financial support provided by the Coordination for the Improvement of Higher Education Personnel (CAPES, grants #88887.485183/2020-00 and #88887136426/2017/00, and fellowships awarded to A.P.R.S., S.S.F., and T.G.S.G.), the National Council for Scientific and Technological Development (CNPq, grant #465571/2014-0), and the São Paulo Research Foundation (FAPESP, grants #2014/50945-4, #2015/08873-9, #2017/18531-3, #2019/22113-8, and #2021/14581-1). The authors thank the National Institute for Alternative Technologies of Detection, Toxicological Evaluation and Removal of Micropollutants and Radioactives (INCT-DATREM) for supporting this work.



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Declaration of Competing Interests

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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