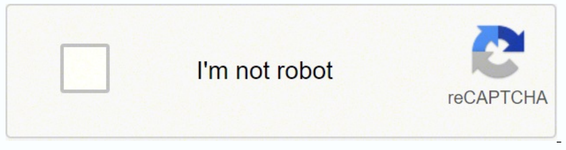

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The color intensity was captured using a smartphone and analyzed through a free App named Photometrix®. ROIs were decomposed in the following color channels: red (R), green (G), blue (B), hue (H), saturation (S), value (V), lightness (L), and intensity (I), providing the pixel’s average value [24].The proposed approach was investigated as a portable and disposable volumetric platform for quantitative analysis. Advances in microfluidic paper-based analytical devices for food and water analysis. Bioanal. This study was supported by INCTBio (grant 465389/2014-7), CAPES (grant 3363/2014), CNPq (grant 448089/2014-9) and FAPEG. [Google Scholar] [CrossRef] [PubMed]Martinez, A.W.; Phillips, S.T.; Butte, M.J.; Whitesides, G.M. Patterned paper as a platform for inexpensive, low-volume, portable bioassays. Electrophoresis 2014, 35, 2309–2324. Nova 2014, 37, 1171–1176. Figure 3. Soc. Filter paper model JP40 was obtained from Quanty (São José dos Pinhais, Paraná, Brazil).Paper microzones were prepared in quantitative filter paper JP40 by wax printing [27]. [Google Scholar] [CrossRef] [PubMed]Dungchai, W.; Chailapakul, O.; Henry, C.S. Use of multiple colorimetric indicators for paper-based microfluidic devices. [Google Scholar] [CrossRef] [PubMed]Busa, L.S.A.; Mohammadi, S.; Maeki, M.; Ishida, A.; Tani, H.; Tokeshi, M. 41–45. Licensee MDPI, Basel, Switzerland. As it can be noted, the color intensity ranged from ca. As it can be noted in Figure 2b, a better correlation for pH values between 2 and 11 can be inferred. The analytical performance of the proposed devices was explored with acid–base titrations examples, where jaboticaba peel extract was used as a natural pH indicator. S.A.N. and W.K.T.C. conceived and designed the experiments; S.A.N., L.R.S., N.K.L.S. and P.H.F.R. performed the experiments, analyzed the data and contributed reagents/materials/analysis tools; S.A.N. and W.K.T.C. wrote the paper. 2014, 406, 5613–5630. A 2016, 1434, 50–56. After printing, devices were passed three times in a laminator heated to 150 °C to completely melt the wax and thus create effective hydrophobic barriers on paper substrates. Angew. The authors declare no conflict of interest. Acid–base reactions were monitored using a natural pH indicator prepared from jaboticaba peel extract, which provides a color gradient over a wide pH range. [Google Scholar] [CrossRef] [PubMed]Cate, D.M.; Adkins, J.A.; Mettakoonpitak, J.; Henry, C.S. Recent developments in paper-based microfluidic devices. Paper as a platform for sensing applications and other devices: A review. Briefly, five jaboticaba fruits were collected, corresponding to a mass of ca. [Google Scholar] [CrossRef]Yu, H.; Tan, Y.F.; Cunningham, B.T. In Smartphone fluorescence spectroscopy. For the readings using universal pH strips, each device was introduced inside sample solution requiring a volume of ca. [Google Scholar] [CrossRef] Figure 1. Titration curves performed on wax printed paper microzones showing examples of (a) NaOH versus HCl and (b) HCl versus NaOH titrations. Similar results were achieved for zones defined with larger diameters; however, they require a greater volume of sample or reagents. As it can be seen in Figure 3a, the data recorded with the proposed device are in good agreement with the values achieved using the universal pH strip. The data associated with the color intensity versus pH is presented in Figure 2b. Gomez, Carlos D. For experimental practices, this low RSD suggests that the microzones can be wax printed, spotted with colorimetric indicator, and then explored by different student groups or even classes of experimental chemistry over the course of 1 day, for example. The colorimetric responses for both examples demonstrated in Figure 4 were recorded in different color channels. Besides, the use of a smartphone to capture images followed by analysis in a free app offers simplicity to all users. The stability over longer times was not evaluated because the printed platform can be quickly prepared by students at the beginning of each experiment.Before proceeding acid–base titrations on paper zones, the colorimetric response to each pH value was evaluated to establish a color pattern as reference. Figure 1 displays the fabricated device layout.Acid–base titrations were performed on wax printed microzones using jaboticaba peel extract (Myrciaria califlora) as a natural pH indicator. Paper spray for direct analysis of complex mixtures using mass spectrometry. Int. For titration, microzones were also spotted with 5 µL of the natural indicator, as previously mentioned. For both examples displayed in Figure 4, five successive titrations were performed in five different microzones. Some apps—which are not very widespread in publications associated with microfluidics—are dedicated to colorimetric detection (e.g., Colormeter® and Photometrix®). In all examples, images were obtained after the addition of each aliquot of titrant solutions. Images were directly captured using the Photometrix® App, available for free download on the Google Play Store. On the other hand, in the report proposed by Karita and Kaneta [25], µPADs were designed to detect specific samples in different concentration levels. The authors would like to acknowledge Gilson A. The latter offers instrumental simplicity and portability, once digital images can be recorded through popular electronic devices like smartphones, digital cameras, or scanners [6,21].The use of smartphones for analytical applications in conventional and miniaturized scales has significantly increased in recent years [6,20,22,23]. [Google Scholar] [CrossRef]Shiroma, L.Y.; Santhiago, M.; Gobbi, A.L.; Kubota, L.T. Separation and electrochemical detection of paracetamol and 4-aminophenol in a paper-based microfluidic device. The natural indicator was prepared according to the procedure described by Guimarães and co-workers [28]. [Google Scholar] [CrossRef] [PubMed]Martinez, A.W.; Phillips, S.T.; Carrilho, E.; Thomas, S.W.; Sindi, H.; Whitesides, G.M. Simple telemedicine for developing regions: Camera phones and paper-based microfluidic devices for real-time, off-site diagnosis. Once the good agreement between the pH values measured by universal pH strips and paper zones spotted with natural indicator was demonstrated, the pH resolution was also investigated. As previously reported [25], the use of synthetic indicators like phenolphthalein, methyl orange, and bromocresol purple offer color changes in a narrow pH range, which is dependent of the pK of each indicator. Karita and Kaneta reported acid–base titrations on µPADs using an array with ten microzones for reaction and detection interconnected to a central zone for sample inlet [25]. ACS Appl. Differently from the mentioned references [25,26], the current study makes use of a smartphone equipped with a free App to monitor acid–base titrations on wax printed paper zones. 0.18 arbitrary units (a.u.) when the pH was raised from 6.1 to 7.0. The data presented in Figure 4 reveal good ability to differentiate pH values in increments of 0.1. This parameter can be defined as the pH resolution of the proposed approach. The color intensity for each pH was calibrated and used to monitor titrations involving (i) strong acid versus strong base; (ii) strong base versus strong acid; and (iii) weak acid versus strong base. It is important to note that the titration on paper microzone does not require more than 20 µL. In this report, we describe the use of a free smartphone application to monitor acid–base titrations on wax printed paper microzones. 2013, 405, 7573–7595. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (. Adhesive tape was fixed to the device bottom to avoid solution leakage. The acid acetic concentration levels were calculated based on the required volume to reach the endpoint of the titration, which was found through the first derivative of the titration curve (data not shown). [Google Scholar] [CrossRef]De Souza, F.R.; Duarte, G.F.; Garcia, P.D.; Coltro, W.K.T. Evaluation of digital image capture devices for colorimetric detection on printed microzones. [Google Scholar] [CrossRef] [PubMed]Wang, L.J.; Sun, R.R.; Vasile, T.; Chang, Y.C.; Li, L. In 2015, Myers and co-workers reported an iodine titration on µPAD [26]. In comparison with universal pH strips, the resolution achieved with paper microzones is quite interesting and advantageous. The use of paper-based microzones prepared by wax printing for applications involving acid–base titrations opens a new gate to be explored in the chemistry field, including the possibility to perform experiments in analytical chemistry laboratories or even basic chemistry for high school. For 1-mm-diameter, wax particles promote partial or total blockage of the zones. To estimate the stability, microzones were spotted with the indicator and the color intensity was recorded between 10 min and 24 h. The required volume of samples and reagents was ca. Photometrix® is an application for the univariate calibration and exploratory analysis of multivariate data from the decomposition of acquired digital images. The obtained curves present similar profiles to those obtained using standard volumetric methods (data not shown). Ed. 2007, 46, 1318–1320. High-throughput optical sensing immunoassays on smartphone. However, the main challenges associated with colorimetric detection by smartphone are external light control and the technical ability of the user to capture images, which can both compromise the reliability of the analytical response [20,21]. [Google Scholar] [CrossRef] [PubMed]Santhiago, M.; Nery, E.W.; Santos, G.P.; Kubota, L.T. Microfluidic paper-based devices for bioanalytical applications. Recent examples of applications using µPADs have demonstrated the ease in coupling with different detectors, including mass spectrometry [12,13], chemiluminescence [14], fluorescence [15], electrochemical methods [16,17,18], and colorimetric detection [6,19,20]. Figure 3. The latter was recently developed by Helfer and co-workers [24], and it is compatible with Android and Windows platforms. Methods 2016, 8, 6506–6511. During the investigation, the use of zones with diameter between 2 and 4 mm compromises the quality of captured image due to the poor focus adjust. It is important to highlight that the proposed methodology demonstrated the capacity to perform quantitative analysis with high reliability and without statistical difference from a conventional methodology applied worldwide. The endpoint of titration was determined by naked eye based on color changes which are associated with the detection of excess amounts of base or acid. Ferrão for their technical support on data analysis performed by using Photometrix®. Braz. This platform offers great advantages, including biocompatibility, low cost, global affordability, and especially the capability to perform tests under lateral or vertical flow with reduced consumption of reagents and samples [1,2,3,4]. After calibrating the colorimetric response, two examples of acid–base titrations were tested. The proposed methodology arises as a new strand to be exploited in the diffusion of the analytical chemistry education field as well as an alternative for quantitative analysis with extremely simplified instrumentation. Chim. [Google Scholar] [CrossRef] [PubMed]Nery, E.W.; Kubota, L.T. Sensing approaches on paper-based devices: A review. [Google Scholar] [CrossRef] [PubMed]Ge, L.; Yu, J.H.; Ge, S.G.; Yan, M. 2015, 87, 19–41. Before titrations, color intensity versus pH was calibrated to be used as a reference in titrations as (i) strong acid versus strong base; (ii) strong base versus strong acid; and (iii) weak acid versus strong base. [Google Scholar] [CrossRef] [PubMed]Silva, M.C.; de Souza, V.B.; Thomazini, M.; da Silva, E.R.; Smaniotto, T.; de Carvalho, R.A.; Genovese, M.I.; Favaro-Trindade, C.S. Use of the jabuticaba (Myrciaria cauliflora) depulping residue to produce a natural pigment powder with functional properties. The analytical response was obtained by using color information from channel S. Lastly, Fabrício R. 0.35 to ca. [Google Scholar] [CrossRef]Miller, J.N.; Miller, J.C. Statistics and Chemometrics for Analytical Chemistry, 5th ed.; Pearson Education Limited: Harlow, UK, 2005; pp. Basically, twelve standard solutions were prepared in a pH range between 1 and 12 and sequentially measured using both universal pH strip and paper zones spotted with natural indicator. Considering the volume usually consumed in a standard volumetric titration (10–50 mL), the waste generation is extremely minimized, thus positively contributing to the green chemistry. As recently observed in the literature, the combination of smartphones and microfluidic devices has promoted a real explosion in the number of publications, and it is a global trend—especially in laboratories with limited resources or restricted access to sophisticated infrastructure. Additionally, the proposed method allows the determination of the titration endpoint and the monitoring of the color intensity in a wide pH range.The feasibility of acid–base titrations on paper and colorimetric monitoring via smartphone was investigated in the quantitative analysis of acetic acid in three commercial vinegar samples. [Google Scholar] [CrossRef]Meredith, N.A.; Quinn, C.; Cate, D.M.; Reilly, T.H.; Volckens, J.; Henry, C.S. Paper-based analytical devices for environmental analysis. For colorimetric analysis, the best focus for image capture was achieved using zones defined with 5 mm diameter. [Google Scholar]Silva, P.B.M.; Oliveira, K.A.; Coltro, W.K.T. Colorimetric detection of glucose in biological fluids using toner-based microzone plates. Presentation of (a) pH values measured with universal pH strip and wax printed paper zones previously spotted with natural pH indicator and (b) pH resolution measured on paper zones. The founding sponsors had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript, and in the decision to publish the results.Coltro, W.K.T.; Cheng, C.M.; Carrilho, E.; de Jesus, D.P. Recent advances in low-cost microfluidic platforms for diagnostic applications. Recently, some research groups have explored the paper platform to perform acid–base and redox titrations based on colorimetric measurements. In the titrations of acid versus base and base versus acid on paper, the colorimetric analysis through Photometrix® revealed that the required volume to reach the endpoint of the titration was ca. The fruits were immersed in 100 mL of 96% ethanol (Vetec, Duque de Caxias, Rio de Janeiro, Brasil) for 6 h. Ed. 2010, 49, 877–880. Quim. Color intensity was captured by smartphone and analyzed through the Photometrix® App. 2010, 82, 2463–2471. The mentioned instrumental and economic advantages open new gates that can directly impact analytical chemistry knowledge diffusion at all levels, not requiring sophisticated infrastructure for volumetric analysis. 2008, 80, 3699–3707. This application was selected due to the simplicity of the matrix and experiment to demonstrate principles of quantitative analysis for students of different levels or courses. Figure 2. The achieved curves in each titration were overlapped to show a good reproducibility offered by the proposed method. The obtained extract was filtered and kept in a dark flask to minimize light influence, which degrades anthocyanins. [Google Scholar] [CrossRef] [PubMed]Adkins, J.; Boehle, K.; Henry, C. de Souza and M. 2010, 82, 3–10. Electrochemical paper-based microfluidic devices. 2009, 81, 7091–7095. [Google Scholar] [CrossRef]Karita, S.; Kaneta, T. The region of interest (ROI) containing 64 pixels × 64 pixels was selected for each image. 2014, 4, 1334–1340. Chromatogr. Acta 2010, 674, 227–233. [Google Scholar] [CrossRef] [PubMed]Myers, N.M.; Kernisan, E.N.; Lieberman, M. An array of twelve microzones of 5 mm diameter each was wax printed on filter paper. [Google Scholar] [CrossRef] [PubMed]Carrilho, E.; Martinez, A.W.; Whitesides, G.M. Understanding wax printing: A simple micropatterning process for paper-based microfluidics. All images were captured with ambient light, keeping the smartphone at distance of 10 cm from the wax printed paper device. Since the first publications from Whitesides’ group [5,6], µPADs have been explored for clinical [1,3], biological [7], food [8], environmental [9], chemical sensing [10], and forensic [11] applications. The achieved data were linearly fitted and presented a coefficient of determination equal to 0.99. [Google Scholar] [CrossRef] [PubMed]Martinez, A.W.; Phillips, S.T.; Whitesides, G.M.; Carrilho, E. The lamination step was performed at rate of 60 cm/min. 2016, 88, 5145–5151. Regarding the selection of sample, we chose vinegar because the determination of the acetic acid concentration in this sample is a common practice to teach principles of quantitative analysis for students of different levels or courses.Hydrochloric acid (Synth, Diadema, São Paulo, Brazil), sodium hydroxide (Vetec, Duque de Caxias, Rio de Janeiro, Brazil), potassium hydrogen phthalate (Synth, Diadema, São Paulo, Brazil), ethanol (Vetec, Duque de Caxias, Rio de Janeiro, Brazil), sodium tetraborate (Synth, Diadema, São Paulo, Brazil), phenolphthalein (Synth, Diadema, São Paulo, Brazil), and methyl orange (Isofar, Duque de Caxias, Rio de Janeiro, Brazil) were used without any purification. [Google Scholar] [CrossRef] [PubMed]Guimarães, W.; Alves, M.I.R.; Antoniosi-Filho, N.R. Antocianinas em extratos vegetais: Aplicação em titulação ácido-base e identificação via cromatografia líquida/espectrometria de massas. Although the obtained values ranged from 5% to 8% compared to the concentrations on the conventional method, the proposed methodology presented high analytical reliability. The titrations of an NaOH solution versus HCl solution as well as the titration of an HCl solution versus NaOH solution were performed using equimolar concentrations (0.1 mol/L each). It is important to note that the quantitative analysis performed on wax printed microzones was based on the required volume to reach the endpoint of the titration. After evaluating the feasibility of the proposed methodology, the concentration level of acetic acid was obtained in three vinegar samples. Then, 1.2 µL aliquots of the titrant solution (0.1 mol/L acid or base solution) were successively added into the microzones, and a digital image was recorded between each addition. Table S1 (available in the supplementary material) presents the composition of all analyzed solutions. Consequently, it is possible to conclude that the waste generation is minimal. However, it needs to be kept in a dark and closed flask to avoid contamination or ethanol evaporation, which would lead to indicator pre-concentration and, consequently, changes in the color intensity. Anal. In addition to the reduced consumption of reagents, the required time to complete a titration on paper microzones as well as the requested instrumentation are other advantageous features in comparison with standard titration. In (a,b), the color intensities were recorded after adding 1.2 µL aliquots of HCl and NaOH, respectively. 2014, 86, 8805–8813. Bioanalysis 2014, 6, 89–106. Simple and sensitive paper-based device coupling electrochemical sample pretreatment and colorimetric detection. Interfaces 2015, 7, 8345–8362. Basically, the app’s goals are to capture information about color intensity and provide chemometric analysis. Better resolution was observed for diameters higher than 2 mm. For pH values lower than 2 and higher than 11, the color intensity did not exhibit appreciable changes. As previously mentioned, titration can be performed in a single zone, and it does not require either change of indicator or its concentration. Analyst 2016, 141, 1874–1887. In addition, natural indicators have toxicity lower than synthetic indicators, causing less environmental impact. Diagnostics for the developing world: Microfluidic paper-based analytical devices. RSC Adv. [Google Scholar] [CrossRef] [PubMed]Oliveira, K.A.; Damasceno, D.; de Oliveira, C.R.; da Silveira, L.A.; de Oliveira, A.E.; Coltro, W.K.T. Dengue diagnosis on laser printed microzones using smartphone-based detection and multivariate image analysis. [Google Scholar] [CrossRef] [PubMed]Silva, T.G.; de Araujo, W.R.; Munoz, R.A.A.; Richter, E.M.; Santana, M.H.P.; Coltro, W.K.T.; Paixao, T. [Google Scholar] [CrossRef] [PubMed]Helfer, G.A.; Magnus, V.S.; Böck, F.C.; Teichmann, A.; Ferrão, M.F.; da Costa, A.B. Photometrix: An application for univariate calibration and principal components analysis using colorimetry on mobile devices. Technol. 5 µL, as expected. [Google Scholar] [CrossRef] [PubMed]Liu, J.J.; Wang, H.; Manicke, N.E.; Lin, J.M.; Cooks, R.G.; Ouyang, Z. Then, 5 µL of different solutions prepared at pH range from 1 to 12 were added on microzones prior to image capture. The obtained titration curves showed the same behavior as the conventional titration curves. In addition, the use of a natural pH indicator over a wide pH range allowed to report standard volumetric applications usually used in chemistry laboratories with acceptable performance. LWT Food Sci. In (b), the color intensity was determined in phosphate buffer solutions prepared in a pH range between 6.1 and 7.0. For pH determination on paper zones spotted with natural indicator, see Figure 2. When spotted on paper, the indicator lifetime is considerably reduced due to some factors that influence its stability, such as light and temperature [29,30]. Mater. Micromachines 2016, 7, 86. Figure 4. 34.4 g. For the titration of base versus acid, the channel S was selected. Scheme of paper devices for acid–base reactions and procedure for colorimetric reading by smartphone. 5 months if stored either at room temperature or when kept refrigerated. In (a), microzones were first spotted with 5 µL of the natural indicator. Instituto de Química, Universidade Federal de Goiás, Campus Samambaia, Goiânia, GO 74690-900, Brazil Instituto Nacional de Ciência e Tecnologia em Bioanalítica, Campinas, SP 13084-971, Brazil Author to whom correspondence should be addressed. While reaction zones were preloaded with different concentrations of primary standard solutions, detection zones were spotted with colorimetric indicator. This channel provides information about the color hue in the ROI, allowing, for example, to distinguish red from yellow [24]. This channel measures the color saturation (i.e., the amount of color that is present in the ROI) [24]. 2014, 86, 12108–12114. In summary, the monitoring of acid–base titrations on paper platforms with a smartphone equipped with a free App showed instrumental and operational simplicity, low cost, and extremely attractive benefits for the environment. Helfer and Marco F. Table 1. The acid–base titration on paper-based devices is outstanding, since any titration can be completed within 5 min using 20 µL volumes. In both examples, titrations were carried out using equimolar solutions (0.1 mol/L). Considering that the calculated values for t-test (2.58–2.94) were below the theoretical critical value (tcrit = 3.18), it can be inferred that both methodologies did not differ statistically from one another at a confidence level of 95%. In this study, twelve microzones with 5 mm diameter each, distributed in two rows of six microzones each, were drawn on Corel Draw software and printed on paper by using a wax printer (Xerox ColorQube 8570, Xerox Corporation, Rochester, NY, USA). Presentation of (a) an optical micrograph showing the color changes on printed zones and (b) color intensity analysis over different pH values. 1000-fold lower than the amount usually employed in standard volumetric methods. Comparison of the acetic acid levels achieved in vinegar samples through classical acid–base titration and the proposed method using wax printed paper-based microzones (n = 3). The proposed method was able to monitor not only the endpoint of the titration, but also the color intensity over the wide pH range. The achieved results were compared to the values determined by conventional acid–base titration. For this reason, we decided to use zones with 5 mm diameter and capture the image at a distance of 10 cm.When compared to a previous report from Karita and Kaneta [25], the use of a natural indicator composed of anthocyanins extracted from jaboticaba to monitor acid–base titrations is advantageous as it promotes a color gradient through a wide pH range, varying from magenta to different shades of green. Acta 2012, 725, 44–50. Two limitations of the method reported by Karita and Kaneta are related to the poor color uniformity and stability after drying, as well as the dependence of the indicator concentration for a better visualization of the endpoint of titration [25]. Through the wax printing method, zones with diameter between 2 and 8 mm can be produced. Lab on paper: Iodometric titration on a printed card. The obtained concentrations through both methods are shown in Table 1. J. Based on the colorimetric analysis, the relative standard deviation (RSD) ranged from 1.3% to 3.0%. [Google Scholar] [CrossRef]Dai, Y.T.; Rozema, E.; Verpoorte, R.; Choi, Y.H. Application of natural deep eutectic solvents to the extraction of anthocyanins from catharanthus roseus with high extractability and stability replacing conventional organic solvents. CNPq is also acknowledged for the scholarship and researcher fellowships (grants 311744/2013-3 and 308140/2016-8) granted to the LRS and WKTC, respectively. Figure 2. Electroanalysis 2016, 28, 1420–1436. The calculated concentrations of acetic acid in three samples ranged from 3.87% to 3.93%, and the proposed methodology did not significantly differ from classic acid–base titration at a confidence level of 95%. The feasibility of the proposed approach for performing quantitative analysis of acetic acid concentrations in vinegar samples was successfully demonstrated. This may be attributed to the indicator stability in extreme pH values. Chem. Standard solutions were prepared in a pH range between 1 and 12. For each titration, microzones were first spotted with 5 µL of the natural indicator. In this way, the use of a wax printed zone for monitoring acid–base titrations is simpler than the µPAD proposed by Karita and Kaneta [25]. [Google Scholar] [CrossRef] [PubMed]Thom, N.K.; Lewis, G.G.; Yeung, K.; Phillips, S.T. Quantitative fluorescence assays using a self-powered paper-based microfluidic device and a camera-equipped cellular phone. 2017, 28, 328–335. 2014, 55, 203–209. Acid-base titrations using microfluidic paper-based analytical devices. The reliability of the colorimetric response recorded with the natural indicator was compared to the pH values measured using a universal pH strip. 2017, 28, 197–201. As observed in the optical micrograph depicted in Figure 2a, the pH variation for values between 1 and 12 leads to a color change from magenta to green. Prior to titrations, the calibration of the colorimetric response over the pH range was performed by adding 5 µL aliquots of different standard solutions into the microzones spotted with the indicator. This application presents a user-friendly interface and it can be obtained for free in the Google Play Store. According to the presented data, the differences between values of the proposed method and classical method were lower than 8%, which is considered satisfactory for analytical purposes. [Google Scholar] [CrossRef] [PubMed]Mahadeva, S.K.; Walus, K.; Stoeber, B. Microfluidic paper-based devices (µPADs) have received considerable attention from the scientific community. The instrumental simplicity and requirements enable their use for experimental practices in high school or undergraduate courses. Development, characterization, and application of paper spray ionization. After the addition of 30 µL aliquots of solution at the sample inlet zone, the fluid was transported through the channel by lateral flow reaching the reaction and detection zones. 2015, 87, 3764–3770. According to the data presented in Figure 4a,b, the addition of acid or base aliquots promoted noticeable changes in the colorimetric response. In this case, the higher resolution of the proposed device opens the possibility to monitor reactions that promote changes in a narrower pH range. 2016, 88, 8302–8308. In their study, the authors developed µPADs for detecting NaOH in two concentration ranges (0.1–1.0 mol/L and 0.01–0.10 mol/L), which required different amounts of indicator to make the detection of the titration endpoint possible. Analytical information about the color intensity can be achieved in real-time, thus reducing the analysis time associated with the image processing and pixel’s intensity on similar platforms using scanner as detector [32]. Images were captured with a Samsung Galaxy smartphone model J5 equipped with a 13 MP resolution camera (Samsung Electronics, Suwon, Gyeonggi, Korea). Hathcock for the support on statistical analysis and English translation, respectively. Garcia and Nam-Trung Nguyen Micromachines 2017, 8(5), 139; Received: 30 January 2017 / Revised: 19 April 2017 / Accepted: 28 April 2017 / Published: 2 May 2017 This study describes the use of a smartphone for monitoring acid–base titrations on wax printed paper microzones. In comparison with a previous report from Karita and Kaneta [25], the titration on paper microzones requires a longer time to be completed, and it demands a lower titrant volume. 1 mL to obtain coloration and allow the comparison with a pH scale defined in a color gradient as reference. Basically, different buffer solutions composed of sodium phosphate were prepared in the pH range between 6.1 and 7.0 with increments of 0.1. The recorded data are depicted in Figure 3b. It is well-known that universal pH strips are limited to distinguishing pH variations of 1 unit. Lab-on-paper-based devices using chemiluminescence and electrogenerated chemiluminescence detection. As proof-of-concept, the acetic acid concentration level was determined in three different vinegar brands and compared to the values achieved by a standard titration method. [Google Scholar] [CrossRef] [PubMed]Mettakoonpitak, J.; Boehle, K.; Nantaphol, S.; Teengam, P.; Adkins, J.A.; Srisa-Art, M.; Henry, C.S. Electrochemistry on paper-based analytical devices: A review. Two important features are related to the sample volume required for reading and the pH resolution. On the other hand, the titration of acid versus base was monitored in channel H. Paper-based assays were carried out without any previous sample treatment; i.e., vinegar sample aliquots were added to the microzones and analyzed without dilution. Nova 2012, 35, 1673–1679. [Google Scholar] [CrossRef] [PubMed]Wang, H.; Liu, J.J.; Cooks, R.G.; Ouyang, Z. Electrophoresis 2015, 36, 1811–1824. For this purpose, 5 µL of the natural indicator was added to the zones and allowed to dry at room temperature for 5 min. The advantages associated with paper-based devices as well as the popularity of smartphones make their application in any laboratory or research center with limited-resources possible.Microzones were initially fabricated with diameter ranging between 1 and 8 mm. Figure 1. This example is usual in most of the experimental practices of general and analytical chemistry courses. The indicator lifetime in solution is ca. The data found by both methodologies were statistically compared through the Student’s t-test [31]. The decomposition generates the pixel’s intensity in eight channels extracted from different color models [24]. Academic Editors: Frank A. Color intensity was captured through a scanner and analyzed in graphics software. The proposed procedure is performed within 5 min, and it requires only a micropipette and a smartphone. SamplesStandard MethodPaper Zones#A4.25% ± 0.07% (v:v)3.93% ± 0.19% (v:v)#B4.14% ± 0.02% (v:v)3.87% ± 0.18% (v:v)#C4.11% ± 0.02% (v:v)3.92% ± 0.11% (v:v) © 2017 by the authors. The labels depicted in (a) indicate the used microzones for each pH value. Figure 4. In (a), all measurements were recorded for standard solutions prepared in a pH range between 1 and 12. The required volume to fill the entire zone in the paper-based devices is 5 µL, contributing to the minimal generation of waste over multiple assays.

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