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Teaching with simulation tools to introduce the basics of analytical chemistry instrumentation

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Introduction

Lectures on instrumental methods in analytical chemistry courses are typically complemented by practical work to facilitate understanding and to provide students a know-how that will be useful in the workplace. Hands-on laboratory work experience is crucial whether one is working directly in the laboratory (e.g., as a technician) or supervising a team working in the laboratory. In this manuscript, we report on the case of second-year pharmacy students enrolled at the University of Bordeaux, France, who will achieve the title of "Docteur en pharmacie" (PharmD) in a minimum of 6 years. Many of these students will then apply fundamental analytical principles in industry, in hospitals, or in academic research.

Unfortunately, the number of hours devoted to practical work is limited in our pharmacy curriculum: in pharmaceutical sciences, chemistry is part of a broad multidisciplinary curriculum, in which the number of hours dedicated to analytical chemistry is lower in comparison to chemistry students. It should be noted that in the French pharmacy system, there is little leeway as courses must take place in the first 3 years of the curriculum. Until the 3rd year, the courses aim to build a common base of knowledge that is essential to the pharmacy professions. Additionally, the analytical chemistry classes are heavily concentrated in the second year of the curriculum at our institution (they are sometimes spread across the second and third year at other French universities). Due to the high number of students and the low number of instruments in our laboratory, the students have little time devoted to the manipulation of each technique within this intensive program. This limited number of instruments is correlated to the limited budget, which decreases both the opportunities and duration for student experimentation.

As a result of all the above, the time spent per student on each analytical technique is very limited. In our case, the laboratory possesses two UV/Vis spectrometers, two high-performance liquid chromatography (HPLC), and one gas chromatography (GC) instruments and two liquid–liquid extraction (LLE) stations associated with a third UV/Vis spectrometer. Each student has the opportunity to perform a single 3-h practical session on each of these techniques and must share the use of the instruments with one to five other students. Some experiments are lengthy (e.g., HPLC equilibration and run times), which further limits opportunities for students to practice.

As a result, students have difficulties when attempting to solve analytical chemistry problems that are not a straightforward application of the course or that do not follow a solution process they have memorized. Another consequence, which is also linked to the dense program that is offered to them, is that students find it difficult to build on their prior knowledge. For example, they often fail to use their knowledge of molecule polarity and acid/base character when they tackle LLE or HPLC.

Remote teaching in instrumental chemical analysis courses is gaining popularity among students and teachers, especially after most universities moved towards distance learning during the COVID-19 pandemic [1, 2]. In this context, one of the main challenges is teaching experimental laboratory practices remotely. Activities complementary to hands-on experiences (virtual lab simulations and technical videos used in combination with hands-on lab experiences) demonstrated improved cognitive and affective learning [3]. Computer-based teaching tools have the advantage to be more accessible, cheaper, and safer compared to at-home

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laboratory experiments [2]. In any cases, we need to verify student accessibility to Internet services, hardware, and software before we can engage them in remote learning.

Using analytical instrument simulators offer several advantages:

- Opportunity for each student to modify analytical parameters and thus optimize analysis in a faster way
- No exposure to chemicals
- Minimal wear and tear on instrumentation
- Useful for online analytical chemistry education
- Save money

Several scientific publications have already proposed software [4] or Excel spreadsheets [5–7] as tools to simulate chromatographic separations. Some are commercial, for example, Drylab (Molnar-Institut, Germany) and ChromSword (Iris Tech) [8], which may not be compatible with the budget of a state university. Free alternatives exist [9, 10]. Although they are of excellent quality, they are best suited to help advanced students and skilled chemists and designed only to optimize chromatographic separations. For LLE or UV/Vis [11, 12], there are few equivalent applications. Some simulation applications and spreadsheets have been developed for a variety of analytical instruments, but these modules generally operate independently. We needed a tool that would allow students to establish connections between several instruments used concurrently or sequentially and relevant to pharmacy courses, which prompted us to develop our own software.

Here, we present an open-source web application (that can also be downloaded and used offline with a web browser) developed in 2018–2019 and implemented since 2019 at the Pharmacy Department of the University of Bordeaux for students to practice LLE, UV/Visible spectroscopy, and reversed-phase HPLC (RP-HPLC) [13]. The overall goal was to allow students to better understand these techniques through virtual experiments without constraints of time or instrument availability and with a reduced need for mathematical skills. The application was also designed to encourage students to decompartmentalize their knowledge through multi-technique experiments, in which the results of a given experimental module influence the others. Finally, we sought to achieve sufficient modularity to allow instructors to prepare customized questions and practical case studies in order to develop active learning activities [14]. Although this tool was first developed only for a use face-to-face in a computer room, we also present its use during the COVID-19 pandemic in a remote learning.

Software description

Our software was developed thanks to a collaboration between analytical chemistry lecturers in pharmaceutical sciences and the pedagogy and innovation support center (*Mission d'Appui à la Pédagogie et à l'Innovation*, MAPI) both belonging to the University of Bordeaux.

This application is divided into four separate modules: liquid–liquid extraction, UV/Vis spectroscopy, isocratic and gradient elution in RP-HPLC, and chromatographic resolution, which cover a large portion of the techniques taught during the year (Fig. 1).

Each module is accessible from the main menu page or the header of any page. The latter is useful to quickly switch modules without having to first go back to the main page. The header bar includes a target icon aimed at importing, saving, and exporting input parameter values described in the sections below.

Each set of parameters can be recorded by clicking on the target and applies to the currently used module (up to two simultaneously). Parameter sets are named automatically with the module name and date but can be renamed by users. All currently recorded parameter sets can be exported and latter imported, as a JSON file (see the Guide in Supporting information for more details).

There are two main uses for this tool:

1. Many of the input parameters may be defined by the educators to set up the initial conditions of the exercises they want to set up. However, asking students to fill in these values themselves can be time-consuming and error-prone. Educators can therefore generate a single JSON file containing the parameters for all exercises (each set can be named unambiguously), so that students only have to select the set corresponding to the current exercise.
2. Users can save the experimental conditions they have set up themselves so that they can quickly switch between them, which is useful for comparing or optimizing these conditions.

Three modules dedicated to UV/Vis, extraction, and RP-HPLC share the same standards and sample solutions, allowing to perform sequential experiments. All solutions are composed of up to four analytes, called A, B, C, and D, whose physicochemical properties can be altered in the relevant modules.

There are four standards containing a single analyte (useful for quantitation by external standardization or to acquire pure UV/Vis spectra) and a fifth containing a mixture of A and B (useful to set up a separation).

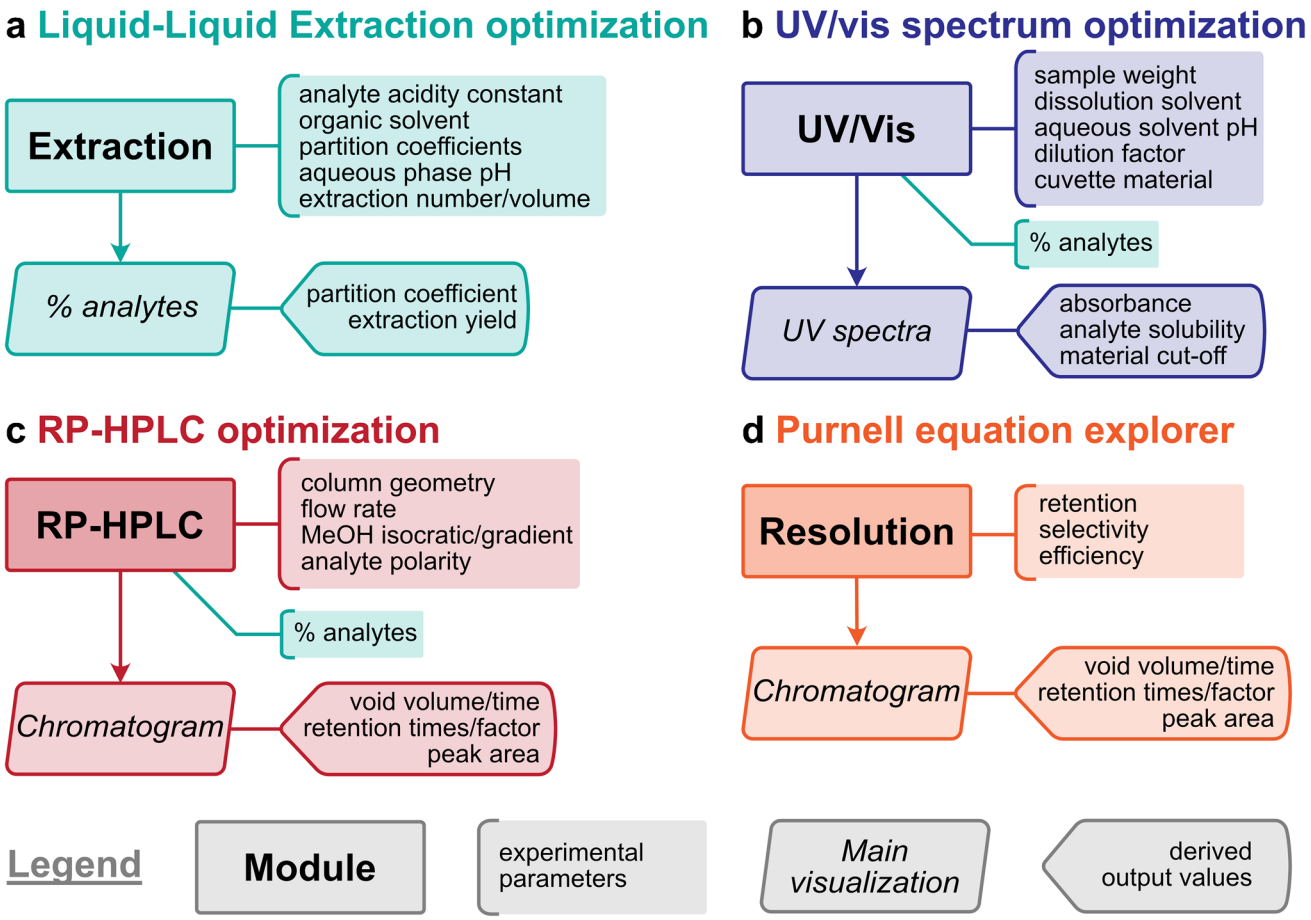

Fig. 1 Schematic description of the simulation tool. Each module appears in a distinct panel: module LLE (**a**); module UV/Vis (**b**); module RP-HPLC (**c**), and module resolution (**d**). The main visualization, derived output values, and tunable experimental parameters are given in different shapes (see legend). The percentage of analytes extracted (% analytes) by LLE (**a**) can be used as an input for the UV/Vis and RP-HPLC modules (**b**) and (**c**)

Twenty samples are available, containing distinct amounts (and unknown to students) of all four compounds. Each student can therefore be assigned a specific sample (to the limit of twenty students).

Some parameters of these analytes and solutions are not directly alterable by students in the user interface, namely, the proportion of analytes, the partition coefficients in different solvents, and the molar absorption coefficients at each wavelength (from 400 to 200 nm) for analytes in their acidic or basic forms. These parameters can be changed by teachers through the creation of a custom dataset, using the supplied template (a Microsoft Excel file in the Supplementary material) and Excel-to-JavaScript conversion tool (see the Guide in supporting information for more details in the Supplementary material).

The output of the extraction module (% analytes) (Fig. 1) can become an input for the UV/Vis and RP-HPLC modules. To do this, a checkbox can be activated in the header bar, producing a split screen in which the extraction module is displayed to the left of UV/Vis or RP-HPLC module. The software is adapted to narrow screens, in which case the modules are stacked vertically.

Any change in the extraction module is dynamically transferred to the other module. It is therefore possible to perform sequential experiments consisting of LLE followed by RP-HPLC or UV/Vis, in which only the analytes extracted by the former are further analyzed by the latter.

"Extraction" module

The aim of the LLE module is to study the influence of key parameters on extraction yields and optimize the selective extraction of analytes (Fig. 1). Here, the extraction is explicitly performed in the aqueous-to-organic phase direction, but

the reversed direction can be implicitly studied by reverting the yields from the output.

Students can select any of the ten available organic solvents and change the pH of the aqueous phase and the number of extractions. Figure 2 shows examples of the extracted fraction obtained for the four analytes depending of the (i) selected solvents (chloroform or diethyl ether (Fig. 2b vs d)), (ii) pH of the aqueous phase (pH 6 or pH 13 (Fig. 2a vs b)), and (iii) number of sequential extractions (one or five extractions (Fig. 2a vs c)). The volume of organic solvent (V_{org}) and the volume of the aqueous sample to extract (V_{aq}) can also be adjusted. The latter may be fixed by educators to set up exercises, letting students optimize the extraction using the former four parameters. The pH can be changed using a slider, allowing to observe trends in extraction yields upon continuous change of the input (Fig. 2a vs b).

The acid–base character of each analyte (A to D) may be specified, for a single acid–base function (i.e., multi acid/base molecules cannot be considered). Specifically, users can change the pK_a and indicate which forms are neutral or ionized (i.e., for a carboxylic acid, the acid form is neutral; for an amine, the basic form is neutral). This input is directed towards educators rather than students to set up exercises but may also be exploited by the latter.

The partition coefficients K_D are fixed (but can modified by educators using custom datasets; see above) for each triad analyte/water/organic solvent, as are the miscibility of organic solvents with water.

The main output of this module is the extraction yield ρ for each analyte, calculated with Eq. (1) where n is the number of extraction and the distribution coefficient D is obtained from Eq. (2), knowing the pK_a of the analyte and the pH (and therefore [H^+]) of the aqueous phase:

$$\rho = 1 - \frac{1}{\left(1 + D\frac{V_{org}}{V_{aq}}\right)^n} \tag{1}$$

$$D = \frac{K_D}{1 + \frac{K_a}{[H^+]}} \text{ and } D = \frac{K_D}{1 + \frac{[H^+]}{K_a}} \text{ for a weak acid and base, respectively} \tag{2}$$

The yields are shown as both a number (0 to 1 which corresponds to 0% to 100% yield) and a bar graph (Fig. 2). The latter is particularly useful to optimize the extraction of any given analyte as students can visually assess the yields of all four analytes at once.

These yields can be sent as input to the UV/Vis and RP-HPLC modules to modify the amounts of each analyte, hence allowing sequential experiments with RP-HPLC or UV/Vis experiments, as mentioned above.

The output also includes values of D, K_D, and their decimal logarithms. Because extraction yields can be determined experimentally by measuring the absorbance of the aqueous phase before and after extraction, the module also outputs absorbance values (the initial being fixed at 1.00). This gives the opportunity to link the simulated results with the practical experience students obtained thus far.

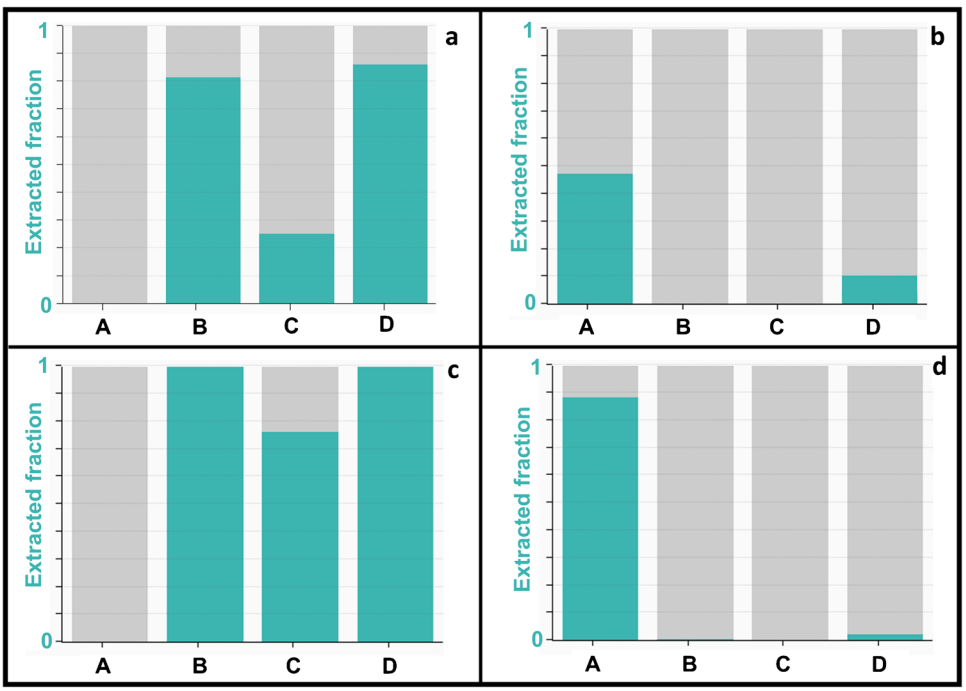

Fig. 2 Extracted fraction (green) of the different molecules (A, B, C, D) depending on the condition set. **a** Single extraction, using chloroform and an aqueous phase at pH 6; **b** single extraction, using chloroform and an aqueous phase at pH 13; **c** multiple sequential extractions $n = 5$ using chloroform and an aqueous phase at pH 6; **d** single extraction, using diethyl ether and an aqueous phase at pH 13. Partition coefficients for A, B, C, and D are 0.9, 4.4, 4.1, and 6.2 in chloroform and 7.7, 9.2, 0.1, and 1.4 in diethyl ether, respectively. pK_a are 9.41, 9.38, 4.95, and 11.25 for A, B, C, and D, respectively. A molecule is set as a basic, whereas B, C, and D are set as acids

"UV/Vis" module

With the UV/Vis spectrum acquisition module, students can explore the different parameters that must be optimized to obtain a satisfactory UV/Vis spectrum such as solvent, pH, and dilution of the analytical solution and cuvette nature. The module can also be used as a toolbox to quiz students on related specific concepts (e.g., molar absorption coefficients and linearity of the Beer–Lambert law).

The initial input is the choice of sample or standard solution and the acidity constant of each analyte. The former influences the relative amounts of analytes, and the latter may alter their absorption coefficients at any given wavelength. The relative amounts of analytes may be further modified by considering the extraction yields in the context of extraction, and this output can be implemented in UV/Vis experiments.

Sample preparation from raw materials is made possible by a second set of input values, including the mass of weighed sample, nature (water, methanol, ethanol, acetone, dichloromethane), and volume of solvent for dissolution into a stock solution and the volume of sampled stock and final sample for further dilution. For aqueous solutions, the pH can be modified to explore the influence of acid/base equilibria on absorbance spectra.

Two instrumental parameters can be modified: the cuvette material (quartz, glass, or plastic) and, optionally, the noise. The cuvette pathlength l is kept constant at 1 cm. The molar absorption coefficient ε is defined at each wavelength λ for each analyte (A–D) and is the average of the basic and acidic values defined in the dataset weighted by their pH-dependent abundances.

The main output is the UV spectrum, produced from the parameters above. It is generated by using Eq. (3) at each discrete wavelength, where i is A–D and C is the analyte molar concentration, which depend on the selected solution and, optionally, on the extraction module output:

$$A_\lambda = \sum_i \left(\varepsilon_{\lambda,i} l C_i\right) \tag{3}$$

Following the choice of dissolution solvent, a textual output indicates whether all analytes are indeed solubilized. This obviously also influences the spectrum but may not be directly interpretable by students since other parameters may lead to similar results (e.g., very low analyte concentrations). This textual output therefore replaces the visual output they might have in real life, e.g., the presence of solid particles in the solution, and assists them into selecting adequate experimental conditions.

The spectrum may be overly noisy with absorbance values too low (low analyte concentration) (Fig. 3c) or, on the contrary, reach an absorbance plateau with values too high (here, $A_{max} = 2$; high analyte concentration (Fig. 3b), cuvette material cut-off or use of non-UV transparent solvent (Fig. 3a)), which constitute important feedbacks for students. If appropriate concentrations and settings (cuvette material and solvent) are entered, the generated spectrum is suitable for quantification (Fig. 3d).

"RP-HPLC" module

The aim of the RP-HPLC module is to optimize the separation of two to four analytes by RP-HPLC in isocratic or gradient mode. Together with the resolution module, it allows students getting a better understanding of the principles of HPLC separation. It is also amenable to quantification by external calibration taking advantage of the standard solutions containing pure A–D.

Users can select numerous input values, some of which may be fixed by educators to set up exercises. Most of the numerical values can be changed using sliders so that students can apply continuous change to the input and observe the corresponding result dynamically in the output. Any solutions and standards defined can be selected. The relative amounts of analytes may be altered by the LLE yield if both modules are used simultaneously. The logP of the four analytes can be modified.

The column geometry (length, particle diameter) can be changed (Fig. 4a, c and d). It can be fixed to challenge students to optimize the separation in a given setup or, on the contrary, can be used to explore the influence of these geometric parameters on the separation. Figure 4a, c and d illustrates the change in particle diameter and column length on hold-up time, efficiency, and resolution.

The nature of the organic solvent is fixed (methanol), but the flow rate and composition of the mobile phase can be changed. Figure 4a and b exhibits a flow rate change. Both isocratic and gradient elution are possible (compare Fig. 4a and e). In the latter case, the initial and final percentages of methanol as well as the duration of the gradient can be altered.

Optionally, the noise level can be changed, which is valuable to demonstrate the issues arising from low-concentration analyte or wide peaks. The detector time step can also be modified to demonstrate its importance on peak shape.

Students can visually assess the quality of the separation from the chromatogram, on which the void time is also explicitly displayed. The retention times of individual analytes are reported in a table; hence, co-eluting peaks that are not distinguishable on the chromatogram may still be detected.

Several values characterizing the separation are provided, in particular the gradient slope, efficiency, void

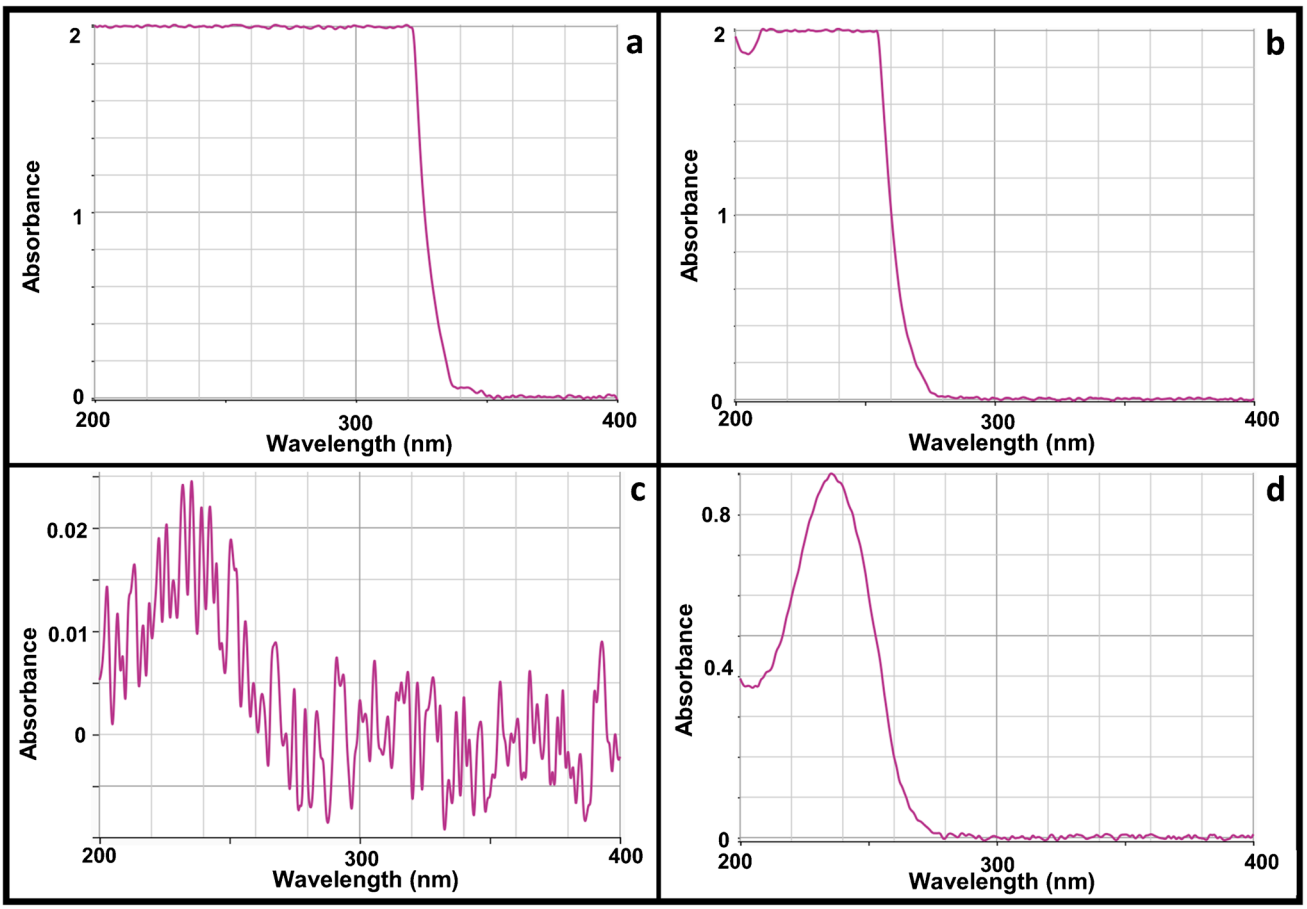

Fig. 3 UV spectra generated depending on the set conditions. **a** Non-transparent cuvette or solvent used; **b** too high concentration of the analyte; **c** too low concentration of the analyte; **d** appropriate conditions for quantification of the analyte (cuvette material, solvent used, and concentration)

time and volume, and the minimum resolution between two consecutive peaks.

For quantification purpose, the peak areas are also reported. Students can therefore switch between standard and sample solutions to determine the amounts of analytes using a single-point external standardization.

"Resolution" module

This module uses the Purnell equation to provide students with a dynamic visualization of the influence of key chromatographic parameters on the resolution between two peaks, as they may not have a good appreciation of this from the equations alone. This module is entirely independent from others: it does not allow exporting the output to other modules nor importing the input from another module. However, students can refer to it when using the RP-HPLC module, for example, to better understand why low retention, selectivity, or efficiency leads to deteriorated resolutions.

The user must choose a separation factor (α) between the two peaks, a retention factor (k) and plate number (N), and can optionally modify the noise level. The input interface is a set of three sliders, allowing students to continuously modify N, k, or α values, giving them a good sense of their influence on the resolution. The column geometry (150×4.6 mm) and peak areas are kept constant, so that only N, k, and α have an impact on the resolution (Fig. 5).

The graphical output is composed of a parallel-scale nomogram of the resolution (R_s) and the corresponding chromatogram for two analytes.

The module produces the nomogram following the Purnell equation (Eq. (4)):

$$Rs = \frac{\sqrt{N}}{4}\frac{k}{k+1}\frac{\alpha-1}{\alpha} \tag{4}$$

The nomogram highlights how much the resolution is changed (e.g., the influence of k is large for small values but rapidly plateaus, while that of efficiency is more gradual and

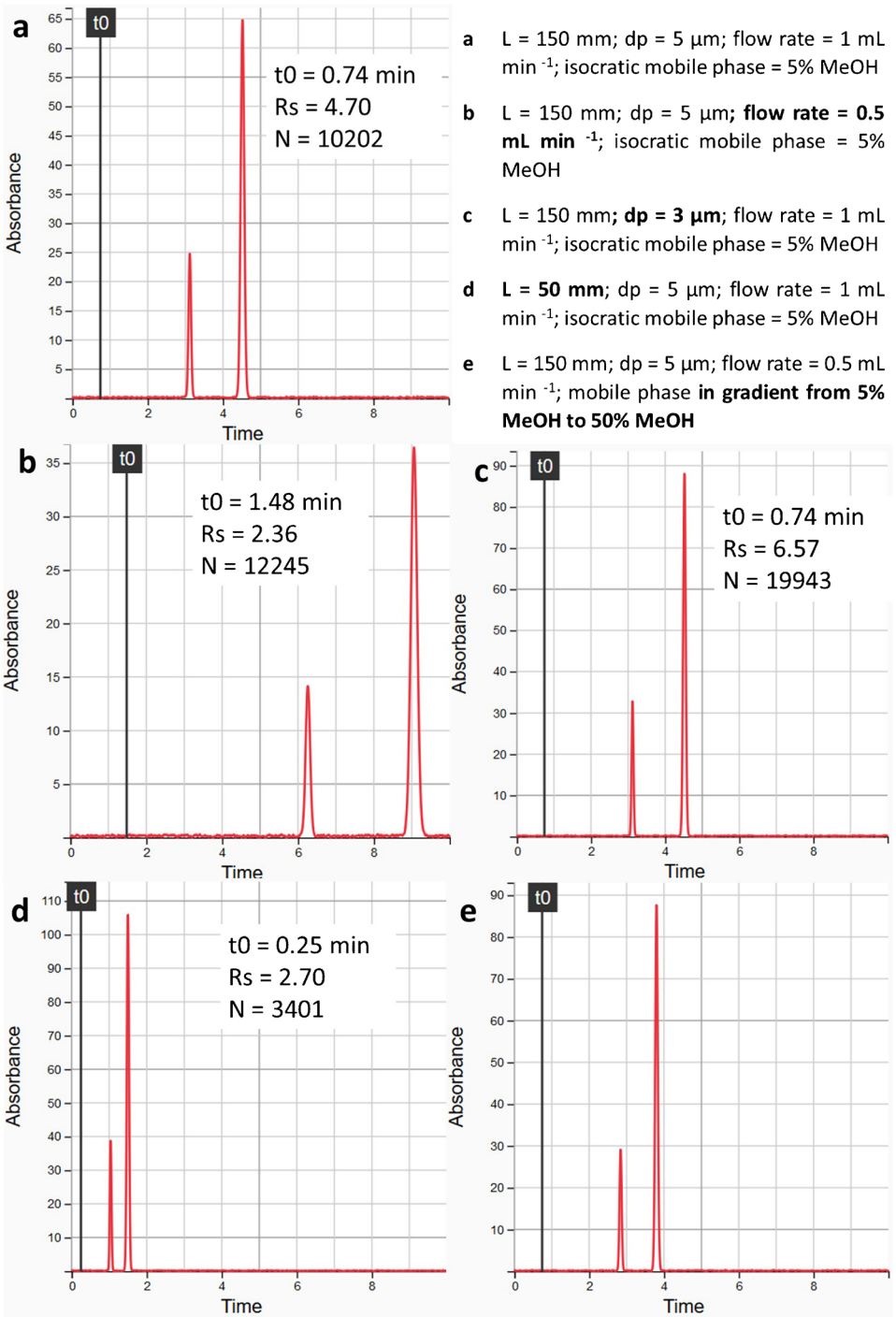

Fig. 4 Chromatograms generated depending on the conditions set. Different flow rates (**a**) and (**b**); 3-mm internal diameter column with different dimensions: (**a**) L = 150 mm and dp = 5 µm, (**c**) L = 150 mm and dp = 3 µm, and (**d**) L = 50 mm and dp = 5 µm; different elution modes: isocratic (**b**) vs gradient elution (**e**)

extends to larger values). The chromatogram (specifically the retention times and peak widths) is generated from these values. It is very useful for students to visualize the practical influence of those key parameters (e.g., a decrease of efficiency is detrimental to the resolution because the peaks widen).

A fourth slider for the noise input is valuable for students to observe its influence on peak detection, in particular in cases where peaks are particularly wide (high retention and/or low efficiency).

Several numerical values are also provided to the user: resolution, duration of the analysis (based on the retention time of the second peak), column geometry, void time, and volume (constant).

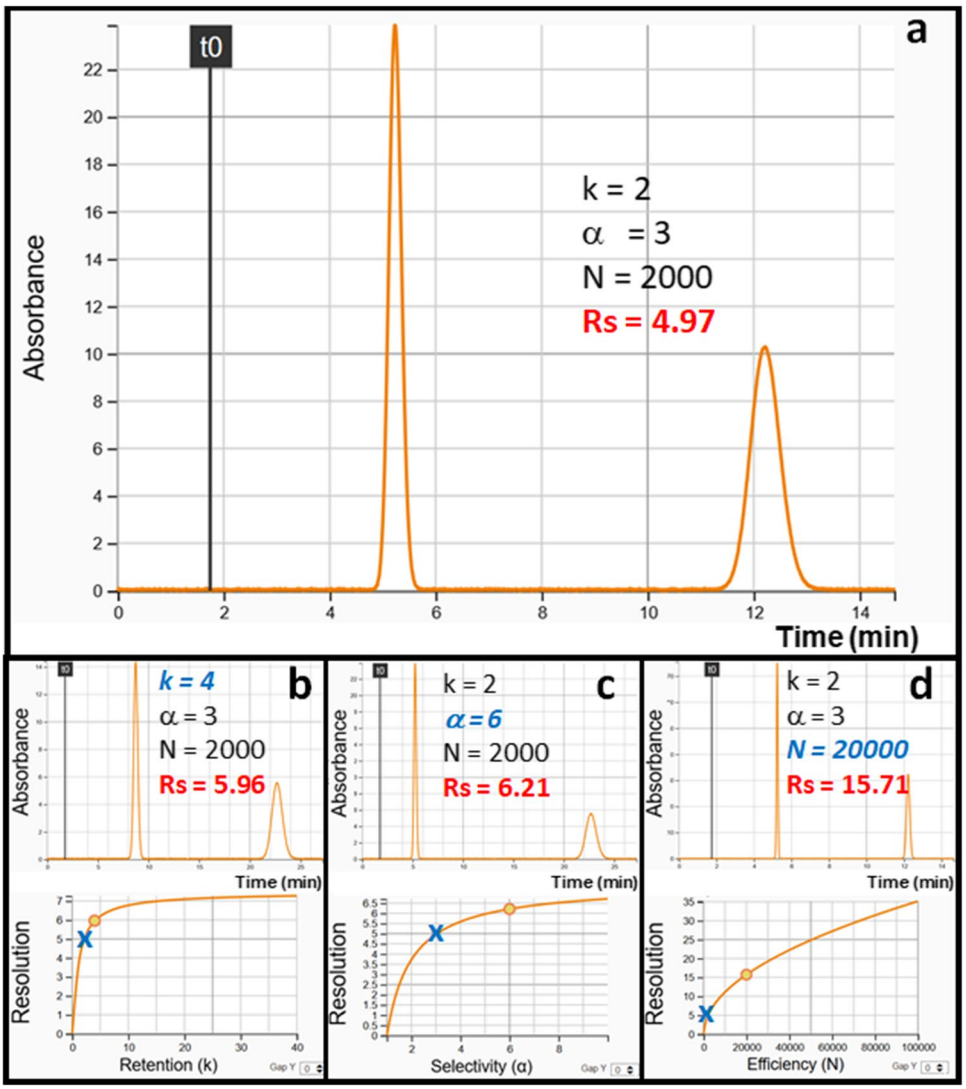

Fig. 5 Chromatograms generated depending on *k*, α, and *N* used in Purnell equation. In the middle panels, impact on chromatograms of changes in *k* (**b**), α (**c**), and *N* (**d**) compared to the reference condition (**a**); in the bottom panels (**b**), (**c**), and (**d**), the blue crosses correspond to the resolution values of the reference condition (**a**) and orange circles to tested condition (**b**), (**c**), and (**d**), respectively. Column geometry (150×4.6 mm) and peak areas are kept constant

Practical work protocol

Our experience with this software was exclusively with second-year pharmacy students, who graduate after a minimum of 6 years of study. It was therefore aimed at students with only basic knowledge in analytical chemistry. Several analytical instrumentation techniques are taught at this level. The application of the course is divided into two types of sessions: ten tutorials in class (90 min each) and six laboratory sessions of 3 h, the latter four sessions being dedicated to instrumentation. Students in pairs are assigned one out of four techniques (RP-HPLC, LLE, UV/Vis, or GC) at every given session and rotate for the following sessions. The software was used as a final seventh laboratory session in a computer room (IT session). The protocol was based on questions (hosted on Moodle) to familiarize oneself with the software and to simulate laboratory experiments of increasing complexity. The same configuration was used as in the laboratory, i.e., in groups of 16 students and for a duration of 3 h.

The application was mostly used with A and B being active pharmaceutical ingredients (API) and C and D being excipients, but other configurations are possible.

At the beginning of the simulated session, the student had to open the "lesson" activity on Moodle, in which questions are grouped by topic in modules. This page includes step-by-step instructions (also available as a short video) allowing students to set up the activity without the help of a teacher. The objective is to promote student autonomy in their learning so that they can progress at their own pace. The different steps were as follows:

1. Opening the toolbox in another window.
2. Downloading the file containing the initial question settings (eight in total) from a link in the Moodle activity page.

3. Uploading this file in the toolbox using the dedicated red arrow on the header bar. The student had the ability to reload this file at any time during the class to reset the settings to the initial ones, if necessary.
4. Choosing a first module (UV/Vis, extraction, HPLC, or resolution) to start working on, knowing that they must keep the "quantification" activity for the end.

For each module, the students had to answer several questions (4 to 6) using the toolbox to experiment, the aim being to practice and understand, not to recite the course by heart. After the last question of any module, students were directed to the menu page to choose a next module to work on. Hereafter, we present in more details the example of the "UV/Vis" module.

The 5 questions of the "UV/Vis" module are related to various aspects of the analytical technique: the molar absorption coefficient (assignment of values to specific wavelengths based on a spectrum, calculation from a spectrum and solution description, impact of the pH), the influence of cuvette material, and sample concentration on spectrum quality. After loading the initial settings, the students could experiment in the toolbox to determine the correct answer. Specifically, they could modify the sample mass, dissolution volume, dilution factor, and the cuvette material and observe their influence on the UV/Vis spectrum to reach a conclusion. If their answer was correct, the next question appeared. However, if parts or all of the answer was wrong, students were given the opportunity to try again.

After completion of the first four modules, a sample was randomly assigned to each student to complete the "quantification" activity. These samples are qualitatively the same but contain different quantities of the following molecules: two API, tramadol (compound A), and acetaminophen (compound B) with two excipients, magnesium stearate (compound C) and lactose (compound D) contained in a tablet. Several external standards were also available in the toolbox (containing pure A, B, C, and D). The aim of the activity was to quantify both API by gradient RP-HPLC, by external calibration on a single point, after an LLE step. The molecules were selected to ensure that both the RP-HPLC and LLE steps are necessary: the extraction cannot remove all excipients, and RP-HPLC cannot separate all four analytes given the experimental constraints given to the students (in particular the run time). Some parameters were indeed fixed (e.g., single extraction, column geometry, HPLC analysis duration) in order to obtain a realistic result, while others could be modified to optimize LLE and HPLC analyses. As previously presented, the toolbox can display the "extraction" and the "RP-HPLC" modules side-by-side, allowing the students to dynamically observe the impact of LLE yields on the composition of the sample injected in HPLC. The student could then leverage the extraction yields and HPLC peak areas from the standards and sample solutions to determine the API content in their assigned sample. As for the previous activities, if the answer was not correct, the student could try again before ending of the lesson.

We supervised the students in a way that encouraged active learning and understanding of the concepts taught during the year, in the same way as our "traditional" laboratory courses. Specifically, we asked students to read the instructions and questions carefully and to answer them in autonomy. We strongly encouraged students to ask questions that would help them understand the underlying concepts rather than questions that would get the right answer from the teacher. When their first attempt at an answer was not correct, students were invited to explain their choices to a supervising teacher, thus ensuring that they would not pass the question without understanding it. We refrained from interacting with the whole class; instead, we discussed with one or a small group of students to adapt to their specific needs and level of understanding. This method is also more suited to the different pace of the students and the non-linear nature of the class, as students could respond to the different modules in any order they wished. The course content was short enough to allow ample time for discussion between students and teachers.

The students were finally invited to answer a non-compulsory evaluation of the semester, which forms the basis of the student feedback presented below.

We have 4 years of experience in using this tool with this protocol or variations thereof, starting in 2018/2019. In reality, the initial protocol was only applied as described above in 2018/2019, during which the students worked in pairs, sharing the same computer, and in 2021/2022 (not covered in the quantitative feedback below).

During the COVID-19 lockdown (year 2019/2020), only the first laboratory sessions dedicated to instrumentation were carried out. The students were therefore only able to experiment with one of the four techniques planned in the rotation. Only short videos (filmed as part of the overall effort to modernize our teaching) describing the principle of the instrumentation, their use, and the planned experiments were made available to the students. The simulated session could not be conducted at the university either, so it was made available online for students to participate from home. In view of the difficult situation, we offered this last simulation session as an option to the students, and the grade obtained was only taken into account if it improved their overall grade.

We were able to bring back the face-to-face simulation session in 2020/2021, but students had to work individually to maintain social distance. This approach was reconducted in 2021/2022.

From the teachers' point of view (three participants), it was found that this approach allowed students to make a final synthesis of the teaching in analytical chemistry, which helped them compared to the previous years. Below, we present the point of view of students.

Student feedback

A survey on the teaching of analytical chemistry for second-year pharmacy students was carried out at the end of the 2018/2019 and 2020/2021 academic years. Students were able to respond anonymously (and optionally) to a set of rating-scale (strongly agree, somewhat agree, somewhat disagree, strongly disagree) and multi-select multiple choice questions. Two of the questions were specific to the simulated laboratory session and more specifically to its usefulness (see questions in Table 1). More than 50% of the students responded ($n = 159$ and 150 in 2019 and 2021, respectively), which was considered to be an adequate sampling of the whole class.

Generally, positive feedback was observed, with a majority of students agreeing that the session was useful, with 76% and 59% in 2018/2019 and 2020/2021, respectively.

The decrease observed in 2020/2021 can be attributed to the conditions under which the activity took place and in particular to the fact that (i) students had to work individually and (ii) that a mark automatically provided by Moodle was displayed at the end of the activity and did not reflect the mark effectively given. Marks were indeed curved based on the students' understanding and behavior during the class and in particular their academic honesty. Working in pairs generally evens out the students' level. When asked to work individually, some students found themselves unable to complete the assignment correctly, resulting in a higher than usual rate of low grades. Both aforementioned aspects generated stress that distracted students from the true purpose of the activity, which may have led some to evaluate it negatively. This is an important observation that should be considered when preparing this session in future years. Thus, in 2021/2022, we kept students working individually but made our expectations clearer (with a focus on understanding), which seems to have resulted in less stress and geared the students towards more productive sessions.

When asked how the activity was useful, students gave similar answers both years. About two-thirds of the students felt that it provided a synthesis of the laboratory sessions (69% and 72%, respectively) and that it was likely to provide practice (64% and 72%) and a way to review the coursework for the exam (67% and 62%). A majority also agreed that it allowed them to practice concepts learned in class and to better understand the impact of instrumental parameters on analysis.

The only marked difference between the 2 years was a decrease in the number of students who felt that the activity was a good summary of the course itself (from 60 to 41%), probably for the reasons mentioned above.

At the end of the survey, the students were given the opportunity to comment. A total of 17 and 12 comments were made in 2018/2019 and in 2020/2021, respectively. The comments covered different aspects: understanding, clarity, feeling, scoring, suggestions, and criticism. The most positive comments were made on understanding, clarity, and feeling, whereas the harshest criticisms were on the

Table 1 Results of the surveys carried out with the students in 2019 and 2021

	Date of the survey	2019	2021
Participation	%	55	56
	n	159	150
To understand instrumentation in analytical chemistry, the format of the final practical work in the computer classroom is useful	Strongly agree	41	35
	Somewhat agree	35	24
	Somewhat disagree	20	19
	Strongly disagree	4	22
The final practical work in the computer room allowed you:*	A summary of the course	60	41
	A synthesis of practical work	69	72
	A revision	67	62
	A training	64	72
	Putting into practice the different concepts established in analytical chemistry	53	57
	An understanding of the impact of instrumental parameters on the analysis	52	57
	Other	2	0

*several choices are possible

final mark. It is possible that some of these students usually rely on their lab partners. As a result, the IT session of 2020/2021 recorded both the lowest average mark and largest standard deviation of the dataset (Table 2) compared to the marks obtained from instrumental sessions carried out in laboratory (i.e., called UV/Vis, RP-HPLC, GC, and LLE).

For the year 2019/2020, among the instrumental sessions in the laboratory, only one out of the four was carried out; three were canceled due to lockdown. The simulated session was conducted asynchronously, online, and only by a fraction of the students who volunteered. They also had to work alone but were not forbidden to communicate with each other. They had at their disposal an assistance chat for technical issues and a forum for questions related to the actual course content. Despite these more challenging conditions (also accounting for the various consequences of COVID-19), students scored similarly to the previous years. Possible explanations for this include (i) a slight adaptation of the marking in their favor and (ii) volunteers having scored higher in previous classes, on average, than students that opted out. Figure 6 is a comparison of mark distributions for students who volunteered ("yes") or not ("no") to the online IT session. Although relatively limited in size (0.30–0.96 with a 95% confidence), this difference in score is statistically significant (Table 3).

Table 2 Results of the marks carried out with the students

Mark[a] means ± SD					
Year	UV/Vis	RP-HPLC	GC	LLE	IT
2018/2019	14.7 ± 2.2	15.2 ± 2.6	14.6 ± 1.6	14.9 ± 1.9	15.3 ± 2.6
2019/2020	14.8 ± 1.5	14.9 ± 1.8	14.7 ± 1.9	14.5 ± 1.8	15.2 ± 2.6
2020/2021	14.1 ± 1.9	14.8 ± 1.7	14.5 ± 1.8	14.7 ± 1.7	13.0 ± 4.4

[a]The student grading in France is between 0 and 20. To pass a minimum of 10 is expected

Concluding comments

This simulation tool for teaching analytical chemistry instrumentation is the first to integrate multiple instrumental techniques. Of the four modules, three are analytical methods (LLE, UV/Visible, and RP-HPLC) which are interconnected. The students can observe the consequence of the extraction on the analysis in UV/Visible or in HPLC. The intensity of the spectral bands and the number of chromatographic peaks as well as their intensity are directly linked to the composition of the sample extracted in LLE. The fourth, based on Purnell's equation, illustrates how the resolution evolves and can be optimized as a function of retention (k), selectivity (α), and efficiency (N). The tool was designed to allow activities to demonstrate an analytical approach to undergraduates, i.e., students new to these techniques. The main reason of its development was to help the students discover these instrumental techniques given the available teaching time and resources. In this way, students can change any parameters and immediately observe the result without any of the time constraints or stress, as is often the case when handling expensive laboratory equipment. Another reason for this development is that the existing solutions are too expensive or for higher-level students.

The software has been used for the last 4 years in the last laboratory course of a series of seven, as part of the

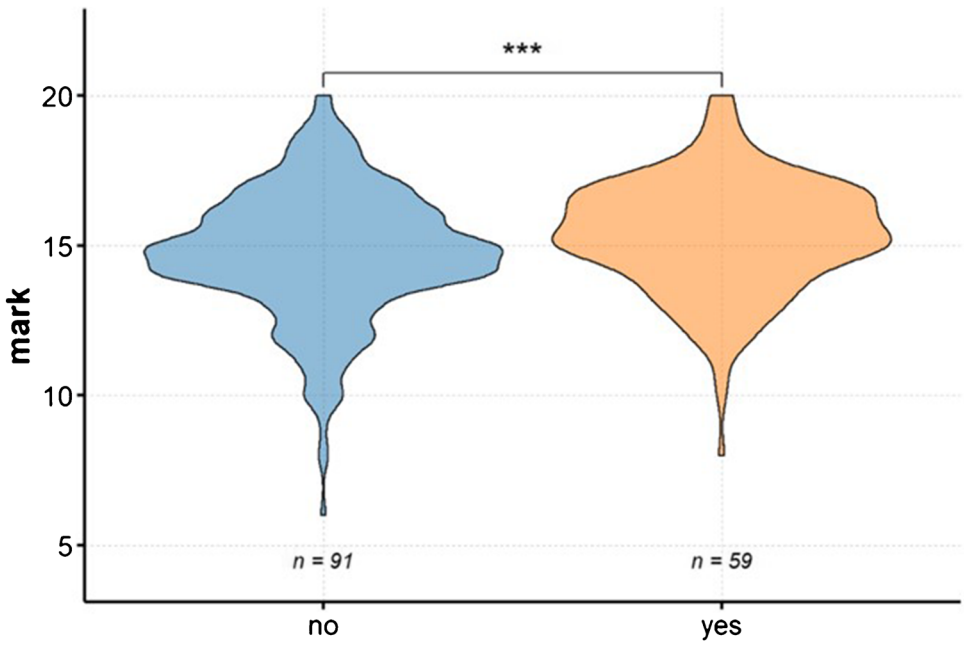

Fig. 6 Violin plots for the mark comparison between volunteered and not volunteered students for the IT session ($p < 0.001$)

Table 3 Statistical significance of the mean marks between volunteered and not volunteered students for the online IT session during 2019/2020

Volunteers	Median	Mean	SD	*t* statistic	Degrees of freedom	*p* value	Mean difference	Confidence interval (95%)
No	15.00	14.78	2.13	3.43	478.66	0.001	0.61	0.30–0.96
Yes	15.25	15.39	1.85					

analytical chemistry courses taught to second-year pharmacy students. Overall, student feedback confirms its usefulness as an effective synthesis of coursework and training activity that has helped them in their understanding of analytical chemistry. The scoring feedback of the session in Moodle has generated stress among students, steering them away from learning. Student feedback on this topic was considered for the latest installment (2021/2022). Notably, no particular criticism was noted for the toolbox itself.

The software was clearly an asset in 2020, helping us continue teaching the practical aspects of analytical chemistry despite the COVID-19 lockdown. More generally, it offers many activities that may not be possible to conduct in the laboratory given the time and resource limitations. Moreover, since the students themselves experiment with changing the parameters of the different techniques in combination, the development of a learning activity based on this tool is part of the active learning approaches for teaching instrumental analytical chemistry [14]. We however believe that it should only be considered as an additional teaching activity that should not replace the bench experience.

This software called "SAN Tools" is a web application, available in English [13] and French [15] in open-source, and the source code can be downloaded at https://gitub.u-bordeaux.fr/mapi/san-tools for offline use or installation on one's own server.

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Declarations

Conflict of interest The authors declare no competing interests.

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