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EVALUATION OF THE ANALYTICAL PERFORMANCES OF TWO RAMAN HANDHELD SPECTROPHOTOMETERS FOR PHARMACEUTICAL SOLID DOSAGE FORM QUANTITATION

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Abstract

This paper addresses the issue of pharmaceutical solid dosage form quantitation using handheld Raman spectrophotometers. The two spectrophotometers used are designed with different technologies: one allows getting a more representative sampling with the Orbital Raster Scanning technology and the other one allows setting acquisition parameters. The goal was to evaluate which technology could provide the best analytical results. Several parameters were optimized to get the lowest prediction error in the end. The main objective of this study was to evaluate if this kind of instrument would be able to identify substandard medicines. For that purpose, two case-study were explored. At first, a full ICH Q2 (R1) compliant validation was performed for moderate Raman scatterer active pharmaceutical ingredient (API) in a specific formulation. It was successfully validated in the ± 15% relative total error acceptance limits, with a RMSEP of 0.85% (w/w). Subsequently, it was interesting to evaluate the influence of excipients when the API is a high Raman scatterer. For that purpose, a multi-formulation model was developed and successfully validated with a RMSEP of 2.98% (w/w) in the best case. These two studies showed that thanks to the optimization of acquisition parameters, Raman handheld spectrophotometers methods were validated for two different case-study and could be applied to identify substandard medicines.

Keywords : Raman handheld spectrophotometer Quantitative performances, Accuracy profile method validation, Uniformity of dosage unit Pharmaceutical tablets

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1. Introduction

Vibrational spectroscopic techniques have known a significant increase of interest since last decades because of their several advantages such as the rapidity of analysis, the non-destructive and non-invasive properties. These techniques and especially near- and mid-infrared spectroscopies, have been widely developed in the agri-food sector in the past decades because of the strong analysis demand during harvest or in the industry. Indeed, the high amount of sample to analyze induce the need to have a technology able to do fast and low-cost analysis. In this context, the development of portable and handheld spectrophotometers allowed to do real time and on-site quality control [1–3]. Many papers are also related to the quantitative aspect of handheld and benchtop near-infrared (NIR) spectrophotometers [4–10] which get more and more places in industries, allowing reduce the use of destructive techniques which are much expensive and time consuming.

Regarding the pharmaceutical industry, since the FDA's guidance of 2004 [11], the process analytical technology is more and more present [12,13] in order to do in-line and in-situ monitoring thanks to probes or optical fibers using vibrational spectroscopy. Benchtop spectrophotometers (spectroscopy and hyperspectral imaging) have no more to prove their efficiency for qualitative and quantitative analyses [14–18] but the problem is their high cost and the cumbersome of these kind of instruments. That is why, there is a significant increase of interest regarding portable and handheld spectrophotometers since 2010 [19]. More and more portable and handheld spectrophotometers are proposed on the market for various applications, at different sizes and prices. However, these kinds of equipment are designed for qualitative information, such as the identification of raw material, pharmaceutical product, quality control [20,21] and also final product identification [22]. Moreover, because Raman spectroscopy provides nice resolved spectra, it is easy to interpret results and the use of chemometrics is not always needed. Indeed, it is rather easy to match spectra with a database thanks to hit quality index or correlation coefficient for example. However, the use of chemometrics can be useful in more complex cases, for example for falsified medicines applications [23–25]. Because handheld spectrophotometers were built for qualitative information, there is a few studies [26–29] which have been evaluated the potential of the handheld spectrophotometers for quantitative purposes.

Regarding the falsification and low-quality standard medicines production, which is one of the major health concern of the century, it is a burden which is expanding to the whole world with approximately 10% of falsified pharmaceutical medicines found in low and middle-income countries in 2018 [30]. It has to be noticed that the European Union has known an important turning point thanks to the Falsified Medicines Directive (Directive 2011/62/EU) [31] which was accepted by the European Council and the European Parliament in 2011. In accordance with this legal text, since the February 9, 2019, the outer packaging of medicines has to get an unique identifier, an anti-tampering device on and a EU-wide logo, also called serialization [32]. Thanks to this new directive, it is hoped to decrease the number of falsified medicines on the global market. However, because not all countries are ruled by

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the same legal texts, there are still a lot of weaknesses in the legal supply chain allowing the appearance of falsified products. In order to help to tackle this burden, several vibrational spectroscopic techniques and especially handheld devices, have been developed since a couple of years. For example, it had been demonstrated the high potential of handheld NIR and Raman spectrophotometers to dismiss falsified medicines with a high specificity [25]. The TruScan™ RM was evaluated in several studies for the detection of falsified medicines, directly on drugs and also after several experimental steps [33,34]. However, no studies have been conducted so far regarding the development of quantitative model on handheld spectrophotometers, which could be an interesting tool for substandard medicines detection.

The global objective of this study was to compare the quantitative performances of two handheld Raman spectrophotometers with different technologies and specifications. The TruScan™ RM (Thermo Scientific) has been chosen because it allows setting the acquisition parameters (laser power, number of co-adds and acquisition time) [35]. The effect of this possibility on the quantitative performances of the device was investigated. Regarding the IDRaman Mini™, this small spectrophotometer is equipped with the Orbital Raster Scanning (ORS) technology that broadens the surface analyzed increasing the representativeness of the spectrum. The two chosen devices allow the investigation of both the influence of the sampling and acquisition parameters. For that purpose, two cases of application have been evaluated. The first study will be focused on a moderate Raman scatterer active pharmaceutical ingredient (API) in a typical pharmaceutical formulation and the other one, a high Raman scatterer in multi-formulation from the market. The first objective of the study was to evaluate the feasibility to quantify the active pharmaceutical ingredient (API) with a total error comprised between ± 15% following the ICH Q2 (R1) guidelines [36].

2. Materials and methods

2.1. SAMPLES

2.1.1. IBUPROFEN FORMULATIONS

A D-optimal experiments design (Table S1) was established, with a set of fifteen runs, each made of different proportion of ibuprofen (TCI, Belgium), microcrystalline cellulose (Sigma-Aldrich, Belgium) (MCC) and mannitol (Sigma-Aldrich, Belgium) and a fixed proportion of other common excipients. Seven levels of concentration of ibuprofen were obtained (70–130%).

Samples used for the calibration and the validation sets in the Raman study were randomly chosen from the production. Three tablets per run were analyzed to constitute the calibration set (45 tablets). The validation step was conducted using four independent series of three tablets (180 tablets).

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2.1.2. PARACETAMOL FORMULATIONS

Tablets of the calibration set were manufactured in order to mimic a typical and simple composition. In order to cover a large concentration range, seven level of concentration, from 50% (w/w) (API/Excipients) to 97% (w/w) (API/Excipients) were produced. The mix of excipients was realised by using Explotab® (JRS Pharma), magnesium stearate and lactose in the proportion (27:7:66). The paracetamol used in the study is a paracetamol PVP3 (3% of povidone) (VWR International, Belgium). The powders were pressed using a Specac hydraulic press based on a 10 mm pellet die and pressed with 2 tons during 1-min long.

Commercial paracetamol tablets were bought at local pharmacies in Belgium and France to enrich the calibration set and constitute the validation set. Twenty different formulations were chosen, with several strengths and shape (Table 1). Several associations of paracetamol with other API were also used in order to show the generalization of the proposed method and to obtain lower paracetamol/other compounds ratio.

2.2. HANDHELD RAMAN SPECTROPHOTOMETERS

2.2.1. TRUSCAN™ RM

The first handheld spectrophotometer used in the study is the Thermo Scientific TruScan™ RM Handheld Analyzer. It has a 785 nm excitation laser (250 mW) covering the range 250–2875 cm^{-1} Raman shifts and has a spectral resolution of 8–10.5 cm^{-1} (FWHM) across range. The TruScan™ RM was used in the point and shoot analysis mode. The principle of the technology is based on the optimization of the signal to noise ratio, so that the analysis stops when this ratio is considered optimal inducing different acquisition parameters for each sample. Recently, a new upgrade known as TruTools™ was released allowing the operator to set acquisition parameters, develop qualitative and quantitative chemometric models and embed them into the spectrophotometer.

The analyses were first performed using the automatic acquisition parameters. Then, six different combinations of acquisition parameters were evaluated. Finally, the influence of the sampling was evaluated measuring 1 spot or 3 spots at different locations on each tablet. The 3 spots were averaged to get a unique spectrum.

For simplicity, the TruScan™ RM in automatic mode will be called Tr-A and the TruScan™ RM with the TruTools will be called Tr-T.

2.2.2. IDRAMAN MINI

The second handheld spectrometer used in the study is the IDRaman Mini™ version 1, from Oceans Optics. It has a 785 nm excitation laser (100 mW), covering the spectral range 400–2300 cm^{-1} with a spectral resolution of 12–14 cm^{-1} (FWHM) across range. This spectrophotometer has an Orbital Raster Scanning (ORS) technology increasing the measured surface by moving the laser spot. This technology

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is designed to enhance the representativeness of the sampling while keeping sharp spectral features. The baseline correction option was turned off. For each formulation, the influence of the sampling was also evaluated by doing 1 spot or 3 spots at different locations of the tablets. The 3 spots were averaged to get a unique spectrum.

For simplicity, the IDRaman Mini™ will be called IDM.

2.3. DATA PROCESSING

All computations were carried out with Matlab R2018b (The Mathworks) with the PLS Toolbox (version 8.6.2, Eigenvector Research).

Because preprocessing can influence results in multivariate analysis, the parameters were optimized for each instrument by means of the root mean square error of cross-validation first (RMSECV) and secondly by the root mean square error of prediction (RMSEP). The cross-validation algorithm used was venetian blind, with 10 data splits, and 1 sample per blind.

Final preprocessing for the ibuprofen spectra were the Savitzky-Golay 1st derivative (polynomial order: 2, window size: 15) followed by standard normal variate (SNV) and mean centering for IDM. A smoothing filter (window size: 15) followed by a standard normal variate (SNV) baseline correction and mean centering was used for TruScan (Tr-A and Tr-T) respectively.

The paracetamol spectra were first reduced to the 582 to 1761 cm^{-1} spectral range then preprocessed by the Savitzky-Golay 1st derivative (polynomial order: 2, window size: 35) for the IDM. For the Tr-T and TrA, the selected spectral range was 284–1794 cm^{-1} preprocessed by a Savitzky-Golay 1st derivative (polynomial order: 2, window size: 15) followed by mean centering.

The method validation for the ibuprofen method was conducted by using E-noval 4.0b (Pharmalex Belgium). The acceptance limits were set at ± 15% of relative total error following the European Pharmacopoeia general monograph 2.9.40 on the uniformity of dosage units. The beta-expectation tolerance intervals were set at 95%. Four series of validation samples were analyzed and each series encompasses three independent repetitions. The devices were shut down and restarted between each series.

2.4. REFERENCE METHODS

2.4.1. IBUPROFEN FORMULATIONS

The reference concentration values of the tablets used in the study were obtained using a previously validated NIR method using a FT-NIR MPA from Bruker Optics (Germany). The reference values for this method were acquired by HPLC. The spectra were collected with the Opus software 6.5 (Bruker Optics). Each spectrum was measured in the transmission mode and is the average of 32 scans with a resolution set at 8 cm^{-1} over the range from 12,500 to 4000 cm^{-1}. The accuracy profile and the validation data

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obtained (β = 95%, acceptance limits: ± 15% total error) are provided in the supplementary data Fig. S1. This NIR model was then used to predict the reference value (% (w/ w)) of the new ibuprofen tablets used in this study.

2.4.2. PARACETAMOL FORMULATIONS

Reference values of the ratio paracetamol/excipients were computed by weighting each tablet. The % (w/w) of paracetamol for the commercial formulations was determined by averaging the weight of ten tablets per formulation and computing the ratio between the average weight and the announced paracetamol amount. Indeed, it has been decided to take the announced dosage as a reference. So that, in order to minimize the error possibly provided by this manipulation, the Raman spectra of ten tablets were averaged to get only one spectrum for each formulation.

3. Results and discussion

3.1. IBUPROFEN FORMULATIONS

3.1.1. OPTIMIZATION OF THE ACQUISITION PARAMETERS FOR THE TR-T

Handheld spectrophotometers were initially designed for qualitative analyses or raw materials. Their acquisition parameters are automatically optimized to obtain a sufficient signal to noise (S/N) ratio. That implies that if the sample is a high Raman scatterer (e.g. paracetamol), the analysis time will be short but if the sample is a weak Raman scatterer or fluorescent (e.g. cellulose derivatives), the acquisition time and number of co-adds will be increased.

In the case of ibuprofen tablets, there is a balanced signal between API and excipients. Therefore, the best S/N ratio will be optimized for each formulation leading to poor quantitative performances. Indeed, for formulations with a higher ibuprofen or mannitol content (good Raman scatterer), the analysis time will be reduced compared to formulations with higher MCC content (poor Raman scatterer and fluorescent). That is why the setting of parameters is interesting, because it enables keeping the quantitative information linked to spectral intensities.

It has been decided to test six different acquisition parameters, with the most intense laser power available (250 mW): 2000 ms/1 co-add, 2000 ms/2 co-adds, 2000 ms/5 co-adds, 5000 ms/1 co-add, 5000 ms/2 co-adds and 5000 ms/5 co-adds to evaluate the best configuration for the quantitation of ibuprofen in tablet forms.

Nine tablets with different API concentration level (Table S1: run 6, 11, 12) were analyzed for each combination of parameters. A principal component analysis was performed and the evolution of the percentage of detector fill was evaluated to help to choose the best acquisition time. As it can be seen on Fig. 1-A, there is a separation between each configuration. It reveals that there is a better

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repeatability for lower acquisition time and higher number of co-adds. In order to choose the best combination of parameters, the spectra obtained with 5000 ms were removed and a new PCA was realised. As it can be seen on Fig. 1B, there is a high variability between repetitions regarding the 2000 ms/1 co-add and 2000 ms/2 co-adds combination compared to the 2000 ms/5 co-adds combination. So that, the higher the number of co-adds, the better repeatability will be. In addition, when comparing the results between the 2000 ms/5 co-adds and the 5000 ms/5 co-adds configurations, there is a significant difference in terms of spectral signature. Indeed, looking at the (Figs. S2-A and S2-B), it appears that there is some noise for the 5000 ms time acquisition, especially on the lower part of the spectrum (< 800 cm^{-1}). Smoother spectra are obtained for 2000 ms whatever the Raman shift range. This is probably explained by the higher percentage of detector saturation for the higher acquisition time (Fig. S2-C). In a nutshell, after the evaluation of several number of co-adds and the acquisition time, the best combination of acquisition parameters for this application is the 2000 ms/5 co-adds.

3.1.2. OPTIMIZATION OF THE SAMPLING

The potential influence of the number of spots measured per tablets on the Raman spectral signature was evaluated. Because the spot size is only about 2 mm diameter for the TruScan and the tablets are 1 cm wide, the sampling representativeness of a single spot per tablet may not be sufficient. That is why two sampling procedures were compared: single spot versus averaged three spots at different locations. In addition, for each individual spot a regression model was built to compare the variability amongst the different sampling. PLS models were built with each sampling procedure and the final sampling procedure was chosen based on the model's RMSECV. The actual vs predicted API content for each PLS models are presented in Fig. S3 and the performances are described in Table S2. The preprocessed and the raw spectra for each instrument are plotted in Fig. 2 and Fig. S4 respectively.

As it can be seen on Fig. S3 and Table S2, the best results in terms of calibration and cross-validation error for the two instruments are logically the three averaged spots. Indeed, there is variability between the three positions measured with each instrument. This is highlighted by the relative difference between the RMSECV per location with values of 9%, 22%, 14% of maximal relative difference for Tr-A, Tr-T and IDM respectively. Averaging the spots provides a more representative and repeatable measure of the tablet improving the quantitative performances. Indeed, the cross-validation error and the calibration error have been improved for both Tr-T and Tr-A. Regarding the IDM device, despite ROS technology, the information obtained is better when three spots are averaged, with a decreased cross-validation error. The error of calibration has been increased but seems to be more representative of the calibration set with a slighter difference between the RMSEC and RMSECV. The prediction of the four validation datasets was also performed on each individual spot model to ensure that the averaged spot was the best strategy with external validation data. Looking at the RMSEP values confirmed the conclusions drawn from RMSEC and RMSECV. A decrease from 1.35 to 1.18% (w/w) and 0.92 to 0.85% (w/ w) and for the Tr-A and Tr-T respectively is observed. In case of IDM, the evolution of the RMSEP is not significate, from 1.00 to 0.98% (w/ w), which means that the ROS technology is rather efficient. Moreover, when comparing both instruments, it appears that the

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Tr-T is more suitable for the prediction of the unknown samples. Comparing Tr-T and Tr-A, the calibration and cross-validation error are comparable, but the best RMSEP is for the Tr-T, 0.85% (w/w) compared to 1.18% (w/ w). The same trend is observed for the R^2 of prediction. The prediction of the four validation sets with the final models are shown in Fig. S5.

In a nutshell, thanks to these preliminary analyses, the methodology chosen for the quantitative analysis of ibuprofen formulations are: 3 averaged spots acquired with 2000 ms and 5 co-adds for Tr-T and automatic acquisition mode for Tr-A and IDM.

3.1.3. VALIDATION OF THE METHOD FOLLOWING THE ICH Q2 (R1)

The different PLS-R models developed for each instrument were validated following the ICH Q2 (R1) guidelines and the total error concept. Indeed, the RMSEP is an averaged error over the whole concentration range and it does not give an evaluation of the prediction for each concentration level [37]. Results are provided in Fig. S5 and in Table 2.

The very first requirement of any validation is to check the specificity of the method. In spectroscopy, the specificity may be evaluated by looking at the loadings or regression vector from the PLS model. In Fig. S6, it can be seen that, for each instrument, the loading of the first latent variable is highly correlated to the spectrum of the ibuprofen raw material. This comparison ensures that the compound of interest (ibuprofen) is responsible of the major part of the co-variance modeled by the PLS-R model.

Several parameters were then evaluated, such as, trueness, intraassay precision, between-assay precision. The acceptance limits were set to ± 15% following the Ph. Eur. Chapter on uniformity of content [38] and the beta-expectation tolerance intervals were computed for β = 95%. When comparing results obtained from the Tr-T and the Tr-A (Table 2, Figs. 3-A and 3-B), it appears that the trueness, the intra-assay precision, the between-assay precision and the accuracy are significantly improved by fixing the acquisition parameters. Indeed, only Tr-T exhibits β-expectation tolerance intervals comprised in the acceptance limits. It means that 95% of the future measured samples will fall in this interval. In addition, regarding the risk profile (Fig. S7), one can see that the risk of having future samples outside the acceptance limits is much smaller for Tr-T leaving more room for future process variation with 1.3% maximum of risk error compared to 3.0% and 8.7% for the IDM and the Tr-A respectively for the first level of concentration.

Regarding the results of the IDM compared to the Tr-T in Table 2, it can be seen that the two methods are rather equivalent in terms of intra-assay precision. However, when regarding the trueness, the Tr-T is less biased. Looking at the intra- and between-assay precision per equipment, one may see that the values are almost the same meaning that shutting down and restarting the equipment has almost no influence. The error linked to the measurement seems to be minimal. The major part of the random error is due to the sampling representativeness (as expected based on the results from results of paragraph 3.1.2.).

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Consequently, regarding the ICH Q2 (R1) guidelines, Tr-T was the sole equipment validated between ± 15% of relative total error.

3.2. PARACETAMOL FORMULATIONS

This part of the study is focused on the potential influence of excipients regarding multi and co-formulation issues. It has to be noticed that paracetamol is a high Raman scatterer and often highly dosed, so that the signal of excipients has a minor influence on the final sample spectrum. Therefore, the possibility to build a PLS-R model was investigated to quantify the ratio API/excipients that could be applied on different commercial paracetamol formulations. However, because paracetamol formulations are generally dosed between 80 and 92% (w/ w) and co-formulation dosed to less than 70% (w/w), the co-formulation samples were chosen to mimic the low-dosed samples. The difficulty of this study was about the presence of four co-formulations of paracetamol, with other API: caffeine, tramadol, pseudoephedrine or pseudoephedrine chlorhydrate combined with triprolidine. The study was carried out using Tr-A, Tr-T and IDM.

3.2.1. DEVELOPMENT OF THE MODEL

As a first step, a quantitative model was developed on each spectrophotometer with homemade tablets and predicted commercial tablets. The advantage of using homemade tablets was the possibility of varying the API/excipients ratio to cover a large range in a convenient and easy way. However, homemade tablets were not sufficient to predict correctly the test samples. Indeed, the physicochemical properties of the homemade samples and the commercial samples were too different in chemical composition to enable prediction of new unknown samples. In addition, the homemade tablets provided a too homogeneous dataset leading to erroneous predictions of unknown commercial tablets. Therefore, in order to handle this issue, it was decided to use the Kennard – Stone algorithm to select 30% of the test set (commercial tablets) and add it to the homemade calibration set to include more spectral variability. The preprocessed and the raw spectra for each instrument are plotted in Fig. 4 and Fig. S8 respectively. The final composition of each calibration and validation sets are described in Table 1 and the results of the calibration models in Table 3 and Fig. 5. When looking at the calibration performances, the three equipment exhibited comparable performances in terms of R^2_{CV} (0.79, 0.81, 0.83) and RMSECV (5.82, 5.35, 5.27% (w/w)) for the IDM, Tr-T and TrA respectively.

3.2.2. VALIDATION OF THE MODEL

The prediction results obtained with the external validation datasets are shown in Fig. 5 and are resumed in Table 3. Looking at the validation results, the Tr-T has a RMSEP and a R^2 of prediction (2.98% (w/ w)/0.94) inferior to the IDM (3.73% (w/w)/0.80) and to the Tr-A (4.19% (w/w)/0.76). It is worth noting that the co-formulations are as well predicted as the paracetamol only formulations. This point tends to confirm that it is possible (for some specific cases) to build a unique regression model to get

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a quantitative information with a relatively low error of prediction about the API/ratio in market commercial samples. This is a relevant information for two principal reasons. First, since the co-formulations were as well predicted as the paracetamol formulations, it may be possible to detect substandard medicines in an efficient way even with a possible difference of excipients. Secondly, within the context of evaluation of quality of medicines, it may be interesting to follow this methodology to reduce the time and hence, the financial cost of the calibration development step.

4. Conclusion

This study shows that handheld Raman spectrophotometer may be used to obtain quantitative information from different kind of pharmaceutical formulations. Several important points have been raised especially regarding the methodology, which has to be evaluated before any measurement on tablets.

Indeed, for each studied formulation, the possibility to set acquisition parameters has allowed to improve the quantitative performances comparing to automatic parameters. Moreover, it has been shown that sampling is an important parameter to investigate. Indeed, the information obtained with one spot is not always sufficient to acquire representative data of the whole tablet. In our study, the analysis of three spots clearly improved the quantitative performances. However, this parameter should be investigated for each specific case depending on the particle size and homogeneity of the sample.

Once these parameters optimized, a PLS regression model have been validated to assay ibuprofen with one spectrophotometer following the ICH Q2 (R1) guidelines and the total error concept.

This study has also shown the potential of handheld Raman spectrophotometers to quantify a high Raman scatterer and highly dosed API in several formulations using a unique regression model. This approach avoids the construction of a quantitative model for each formulation before going on the field. Once again, it has been shown that setting acquisition parameters have a strong influence on the spectra and consequently on the prediction performances of unknown samples. It was possible to quantify the API/excipient ratio even with co-formulation with another minor API because these co-API were not high Raman scatterer nor highly dosed. Consequently, these models could possibly be applied to evaluate the quality of a broad range of formulations with certain co-formulations and could be applied in case of real substandard medicines.

Credit author statement

Laureen Coic: Writing - Original Draft, Writing - Review & Editing, Software, Validation, Formal analysis, Visualization. Pierre-Yves Sacré: Conceptualization, Writing - Review & Editing. Amandine Dispas: Writing - Review & Editing. Elodie Dumont: Writing - Review & Editing. Julie Horne: Writing - Review & Editing. Charlotte de Bleye: Writing Review & Editing. Marianne Fillet: Conceptualization, Funding

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acquisition, Writing - Review & Editing. Philippe Hubert: Supervision, Funding acquisition, Project administration, Writing - Review & Editing. Eric Ziemons: Supervision, Funding acquisition, Project administration, Conceptualization, Writing - Review & Editing.

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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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.talanta.2020.120888.

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Tables

Table 1

Presentation of paracetamol samples: brand, dosage, additional API, expiry date, and the dosage relative to each sample. The formation of calibration-validation set is also presented.

	Name	Brand	Dosage (mg)	Additional API	Batch number	Expiry date	% (w/w)
Calibration	P1	Dafalgan	1000	–	R4162	4/1/18	91.83
	P3	Paracetamol EG	1000	–	65,132	12/1/19	82.80
	P4	Paracetamol EG	500	–	54,011	10/1/18	86.49
	P5	Paracetamol TEVA	1000	–	B16032A	2/1/19	84.08
	P7	Perdolan	500	–	GHL4000	7/1/19	84.64
	P10	Tramadol paracetamol EG	37,5/325	Tramadol	C7069	1/1/20	68.05
	P15	Algostase	1000	–	18J09	10/1/22	92.22
	P16	Paracetabs	1000	–	ABN8009A	4/1/21	83.76
	P17	Claradol	500/50	Caffeine	NOR112	6/1/21	72.80
	P19	Dolirhume	500/30	Pseudoephedrine chlorhydrate	1154	6/1/21	75.00
	P20	Doliprane	500	–	119	8/1/21	91.53
	P21	Zaldiar	37.5/325	Tramadol	00066G	9/1/17	73.18
Validation	P2	Dafalgan	500	–	T2164	6/1/19	86.59
	P6	Dafalgan	500	–	18,428	2/1/22	84.44
	P8	Panadol	500	–	M74L	5/1/19	84.52
	P9	Panadol retard	665	–	6150351	2/1/18	91.13
	P11	Panadol plus	500/65	Caffeine	40F273B	7/1/20	72.75
	P12	Paracetamol Mylan	500	–	609D141	3/1/19	83.46
	P13	Paracetamol Sandoz	500	–	1760005B	4/1/20	84.55
	P14	Pe-tam	500	–	52,434	12/1/21	79.39
	P18	Actifed	500/60/2.5	Pseudoephedrine chlorhydrate + triprolidine	8AV1681	12/1/19	66.79
	P22	Tramadol/Paracetamol Teva	37.5/325	Tramadol	7294065	6/1/18	70.29
	P23	Tramadol/Paracetamol EG	37.5/325	Tramadol	D7122	2/1/20	67.54
	P24	Tramadol/Paracetamol EG	37.5/325	Tramadol	D7128	2/1/20	67.29

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Table 2

Results of the ibuprofen method validation following the ICH Q2 (R1).

	Concentration level (% (w/w))	Tr-T	Tr-A	IDM
Trueness				
Relative bias (%)	13.92	2.80	−0.0100	−4.20
	17.41	0.800	−2.56	−2.70
	19.69	0.860	−0.080	−0.830
	22.68	−0.790	−2.03	−1.60
	25.60	0.530	−1.50	−1.00
Intra-assay precision				
Repeatability (RSD %)	13.92	5.20	8.50	5.50
	17.41	4.40	4.40	6.10
	19.69	4.40	5.20	5.10
	22.68	3.30	6.02	4.50
	25.60	3.40	4.80	3.30
Between-assay precision				
Intermediate precision (RSD %)	13.92	5.20	8.50	5.50
	17.41	4.40	4.40	6.10
	19.69	4.50	5.20	5.10
	22.68	3.70	6.00	4.50
	25.60	3.40	4.80	3.30
Accuracy				
Relative β -expectation tolerance limits (%)	13.92	[-7.74, 13.35]	[-17.29, 17.27]	[-15.43, 7.06]
	17.41	[-8.53, 10.12]	[-11.78, 6.67]	[-15.59, 10.17]
	19.69	[-8.31, 10.04]	[-10.84, 10.67]	[-11.38, 9.73]
	22.68	[-9.04, 7.46]	[-14.74, 10.68]	[-11.03, 7.82]
	25.60	[-6.42, 7.48]	[-11.27, 8.38]	[-7.66, 5.57]

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Table 3

Results of the PLS model performances and results of prediction for each instrument for the paracetamol formulations.

		Tr-T	Tr-A	IDM
Calibration	Number of spectra	19	19	19
	Number of latent variables	4	4	4
	R2Cal	0.90	0.97	0.95
	R2CV	0.81	0.83	0.79
	RMSEC (% (w/w))	2.97	2.10	2.81
	RMSECV (% (w/w))	5.35	5.27	5.82
	CV bias (% (w/w))	0.61	0.61	0.55
Validation	Number of spectra	12	12	12
	R2Pred	0.94	0.76	0.80
	RMSEP ((% (w/w))	2.98	4.19	3.73
	Prediction bias (% (w/w))	1.44	−0.41	0.08

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Figures

Fig. 1. Principal component analysis on spectral data obtained with different acquisition parameters (ACQ) with Tr-T. A) Score plot representing the variability existing between the several acquisition parameters. Each marker represents one combination of parameters (acquisition time + number of co-adds). B) Score plot after removing the 5000 ms-acquired spectra.

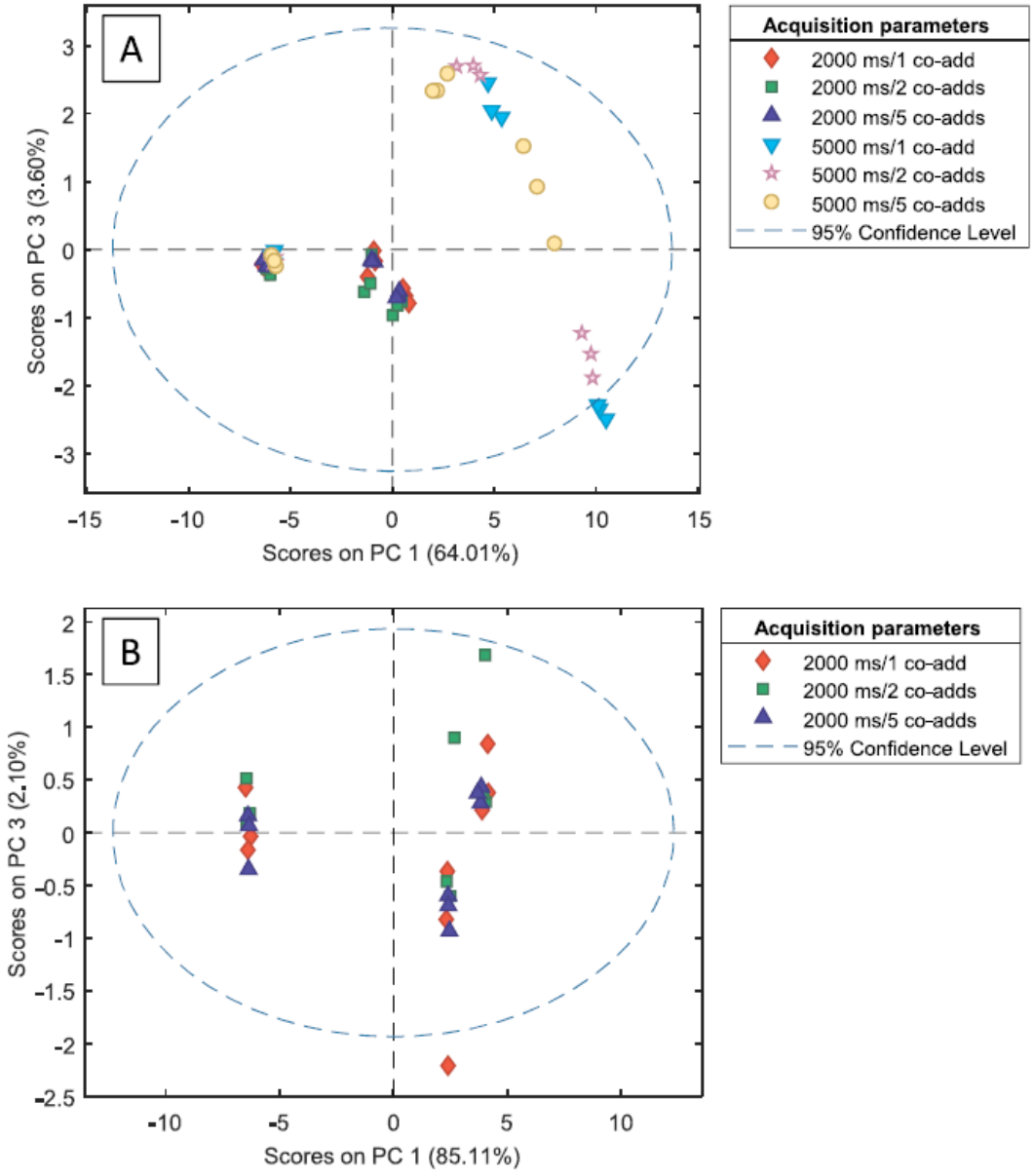

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Fig. 2. Preprocessed spectra of three ibuprofen formulations. A) Tr-T, B) Tr-A and C) IDM.

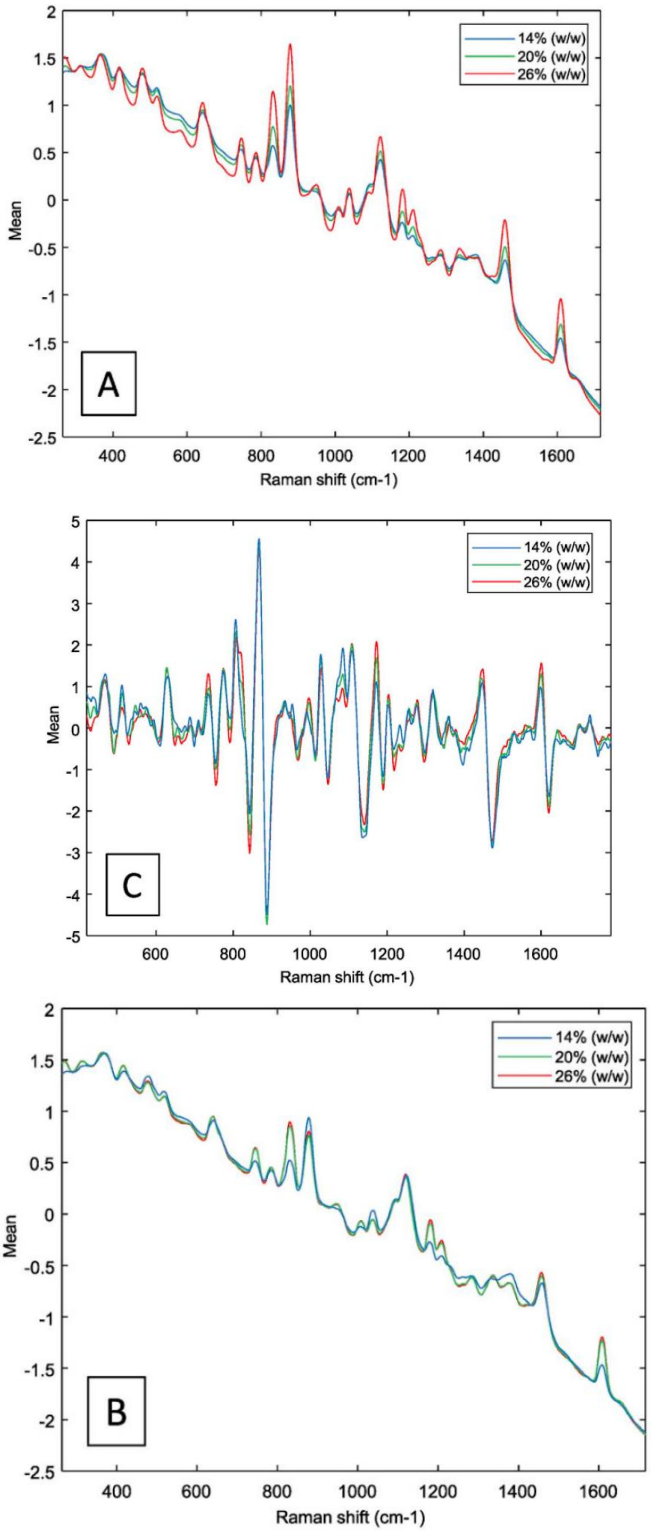

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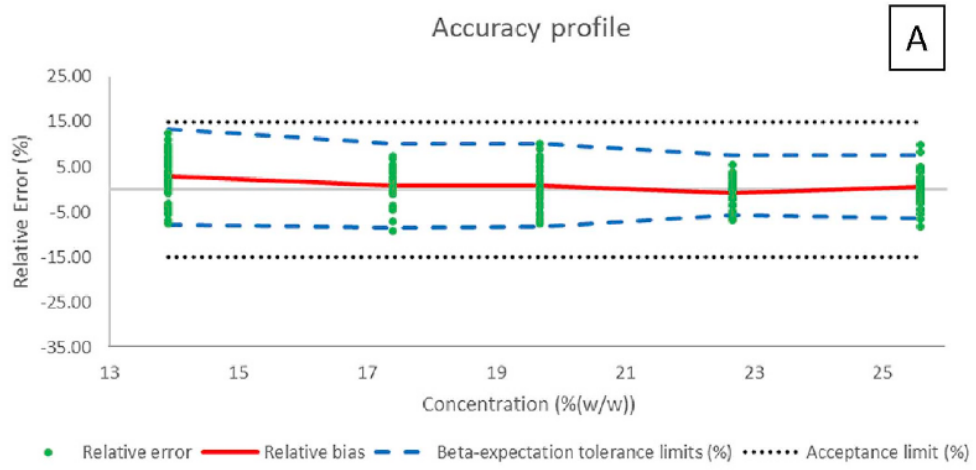
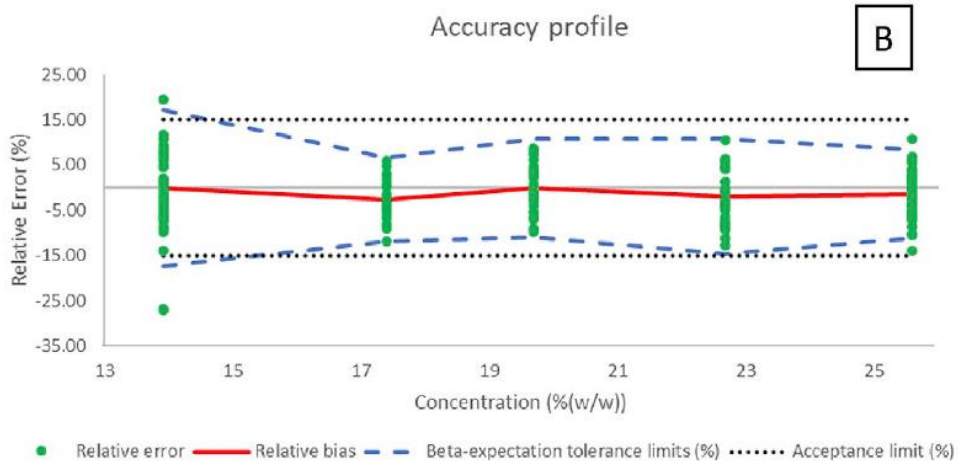
Fig. 3. Accuracy profiles for ibuprofen formulations for each instrument. A) Accuracy profile for the Tr-T. B) Accuracy profile for the Tr-A. C) Accuracy profile for the IDM.





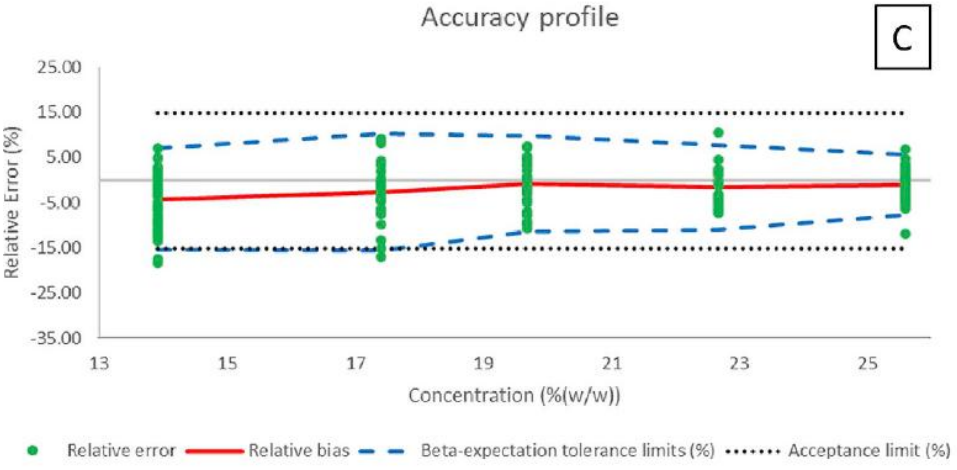


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Fig. 4. Preprocessed spectra of paracetamol formulations. A) Tr-T, B) Tr-A and C) IDM.

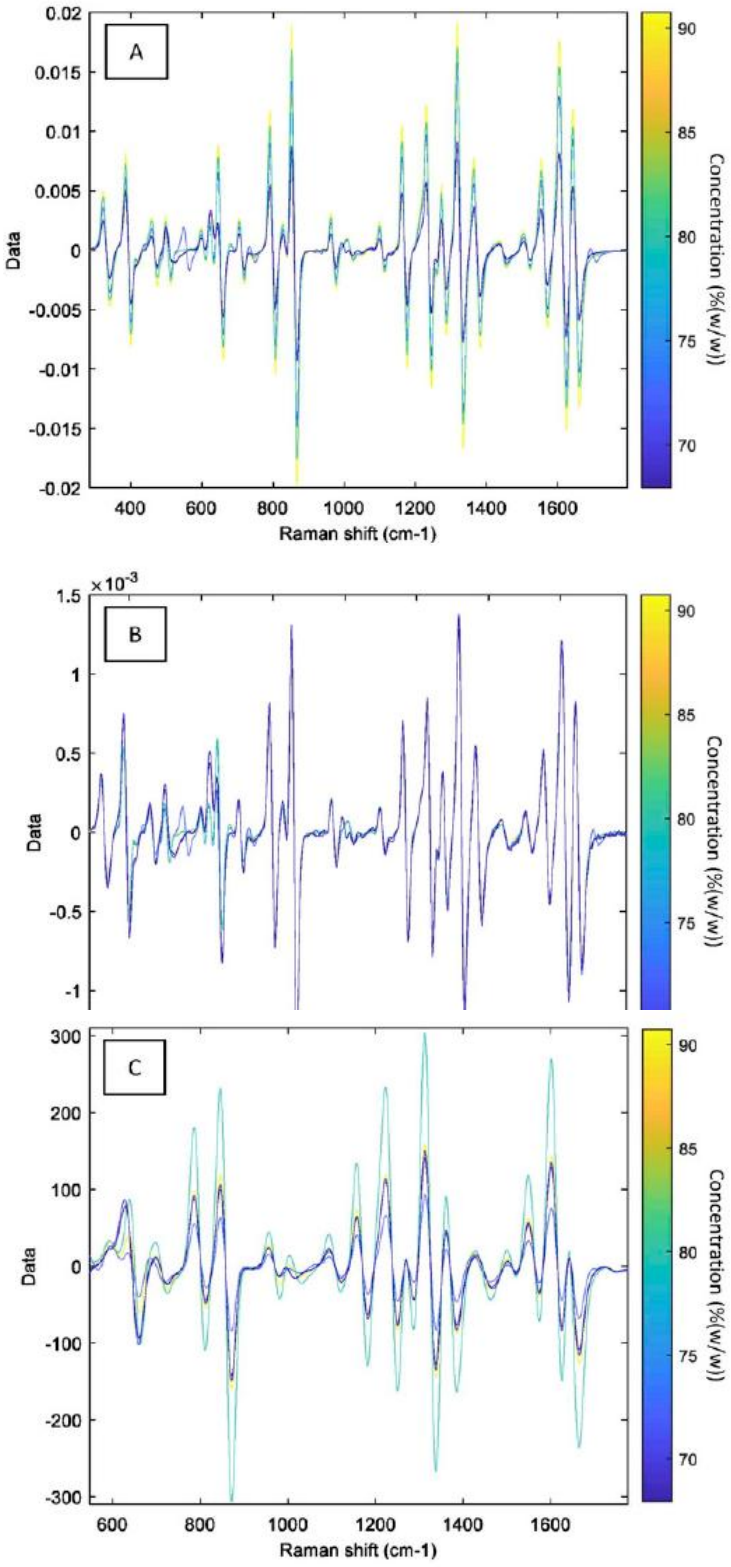

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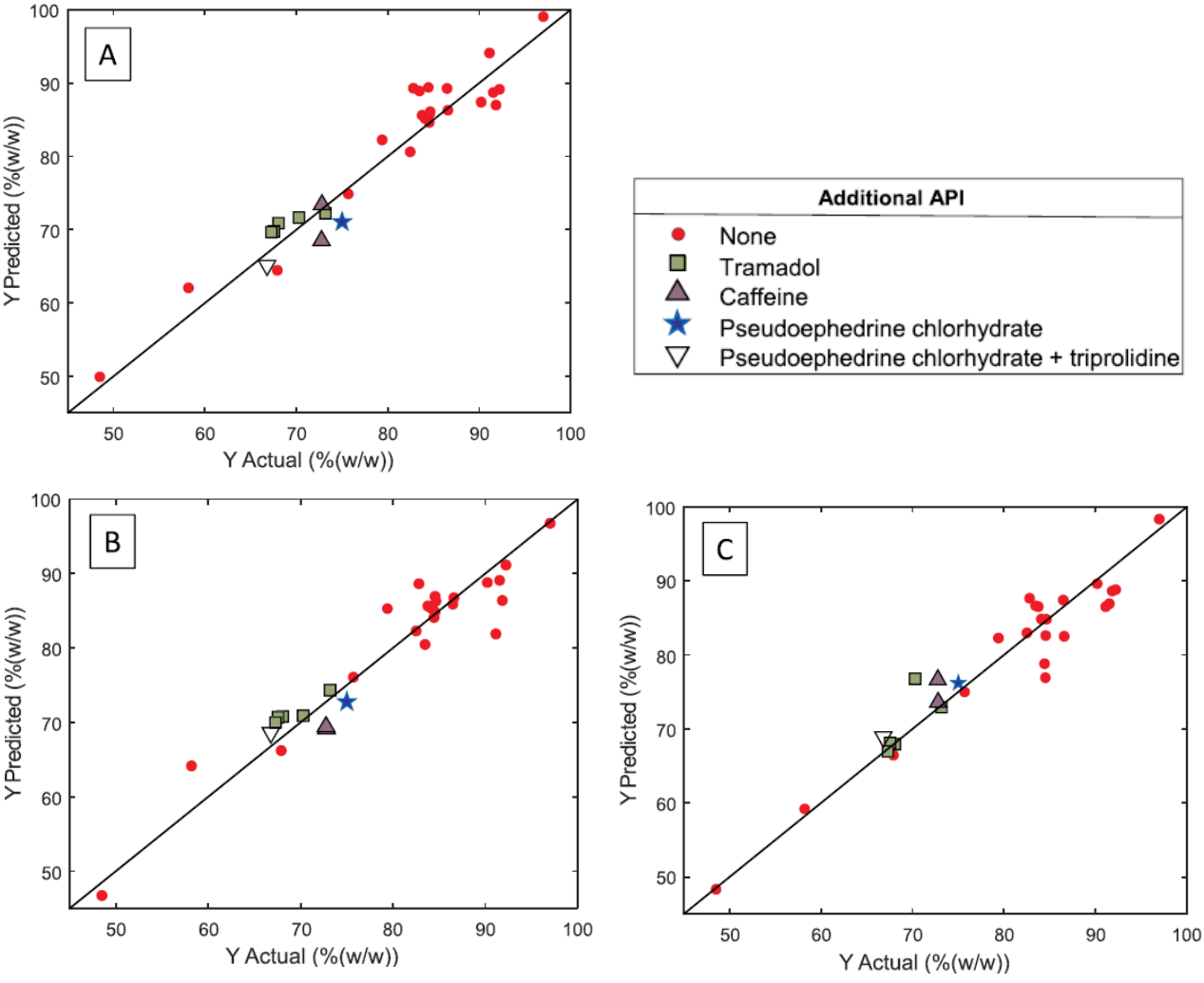
Fig. 5. Results of the prediction of the validation set for each instrument. Co-formulations are plotted with different color and/or marker. A) Prediction curve of the Tr-T. B) Prediction curve of the Tr-A. C) Prediction curve of the IDM. Y actual is the API/excipient ratio based on the claimed label. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)



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