Available online at www.sciencedirect.com
ScienceDirect
Journal of Pharmaceutical and Biomedical Analysis 45 (2007) 356–361
JOURNAL OF PHARMACEUTICAL AND BIOMEDICAL ANALYSIS
www.elsevier.com/locate/jpba

Short communication

Validation of manufacturing process of Diltiazem HCl tablets by NIR spectrophotometry (NIRS)

C. Bodson [a,*], E. Rozet [b], E. Ziemons [b], B. Evrard [a], Ph. Hubert [b], L. Delattre [a]

[a] *Laboratory of Pharmaceutical Technology, University of Liege, 1 Avenue de l'hôpital, 4000 Liège, Belgium*
[b] *Laboratory of Analytical Chemistry, University of Liege, 1 Avenue de l'hôpital, 4000 Liège, Belgium*

Received 6 April 2007; received in revised form 13 May 2007; accepted 14 May 2007
Available online 21 May 2007

Abstract

The goal of this study was to apply the Process Analytical Technology FDA's initiative in pharmaceutical tablets manufacturing. Near Infrared Spectrophotometry (NIRS) was used as a non-destructive, very fast technique requiring no sample preparation.

Direct compression powder blends containing Diltiazem HCl as a model drug were pressed into tablets for the calibration and the validation steps.

First, a partial least squares model was built to calibrate the NIR spectrometer. Then, this model was validated and compared with a validated UV spectrophotometry reference method. For this comparison, the Bland and Altman's statistical method was applied.

The manufacturing process was validated by producing three batches at three different concentration levels. The NIR analysis of these batches was performed during 3 days.

This study shows that NIRS can be used to validate the whole manufacturing process and not only as an analytical method for tablets assay. NIRS is an interesting tool to show possible variations during the manufacturing process which could lead the finished product to fall outside of specifications.

Keywords: NIR spectroscopy; Validation; Manufacturing process; Tablet assay; PAT

1. Introduction

Pharmaceutical controls are time-consuming and require a lot of sample preparation. Process Analytical Technology (PAT) implementation in the pharmaceutical industry would reduce these time consuming operations. One of the most interesting outcomes that PAT offers is the real time release once the last manufacturing step is finished [1]. The real time release is the ability to evaluate and ensure the quality of in-process and final product based on process data [2].

The present manufacturing process time compared to the time spent on quality testing after manufacturing is very low, so real time release offers some significant benefits for the manufacturer. PAT applications involve that sophisticated quality controls are moving from the laboratory to the process or manufacturing plant [3].

To perform this implementation, the use of analytical techniques capable of providing accurate results in a simple and a rapid manner is necessary [4]. During this study, the near-infrared spectroscopy (NIRS), which is a technique that meets these requirements has been used. NIRS is a nondestructive technique that permits determination of chemical and physical properties [5]. Other advantages include speed, simplicity and no sample preparation as required by conventional analytical methods. The NIR region spans the wavelength range 12,500–4000 cm^{-1}. NIRS is also remarkably versatile. If sample contains bonds such as C–H, N–H, or O–H, and if the concentration of the analyte exceeds about 0.1% of the total composition, then it is very likely to yield acceptable results.

One example of studies in which quantitative analysis has been performed using the NIRS technique is the intact tablet assay [6–9]. Usually, these publications present a comparison between the NIRS and a conventional technique. The validation is only focused on the analytical technique. The aim of this study is the validation of the whole manufacturing process by

* Corresponding author. Tel.: +32 4 3664301; fax: +32 4 3664302.
E-mail address: cbodson@ulg.ac.be (C. Bodson).

0731-7085/$ – see front matter © 2007 Elsevier B.V. All rights reserved.
doi:10.1016/j.jpba.2007.05.015

assaying the final dosage form with NIRS in transmission mode. The idea was to demonstrate that NIRS could be used as a real time release system if the analyzed final product meets suitable quality criteria.

In our work, the studied formulation was a direct compression formulation using Diltiazem HCl as the model drug.

2. Materials and methods

2.1. Chemicals

Diltiazem hydrochloride (Pharm.Eur.5th) was purchased from Roig Farma® (Barcelona, Spain). Lactose monohydrate (DCL 15®) was provided by DMV International (Veghel, The Netherlands). Microcrystalline cellulose (Avicel PH 102®) was supplied by FMC (Brussels, Belgium). Colloidal silicon dioxide (Aerosil 200®) and Magnesium Stearate were obtained from Alpha Pharma (Braine-l'Alleud, Belgium).

All solvents used in the reference method were of analytical grade.

2.2. Tablets manufacturing

For the NIRS equipment calibration, three different tablet concentration levels were manufactured: 5, 10 and 15% (w/w). Blends were mixed in a high shear mixer Gral-10® (Collette, Wommelgem, Belgium) during 5 min without Magnesium Stearate at 400 rpm. The lubricant was then added and mixed during 1 min.

Blends were directly tableted using a rotary press (RO/2, GEA-Courtoy, Halle, Belgium). Flat faced tablets were obtained using round punches with a diameter of 7 mm. Tablets weight was fixed approximately at 140 mg, respectively, for each concentration and the tablet hardness was fixed at ~90 N. The active ingredient amounts inside the tablets for the different batches were 7, 14 and 21 mg, respectively.

2.3. FT-NIR equipment

Intact tablets were analyzed by transmission mode with a multipurpose analyzer (MPA®) Fourier transform near infrared spectrometer (Bruker Optics, Brussels, Belgium) equipped with a room temperature-indium gallium arsenide (RT-InGaAs) external detector positioned above the tablet. The spectra were collected with the Opus software 5.0 (Bruker Optics, Brussels, Belgium). Each spectrum was the average of 32 scans and the resolution was 8 cm^{-1} over the range from 12,500 to 4000 cm^{-1}.

2.4. UV reference method

A validated UV spectrophotometry method was used as a reference method (see Table 1). The absorbance of sample was measured at 240 nm with an optical path length of 10 mm by using a HITACHI U-3010 (Tokyo, Japan).

Each tablet was weighed and transferred to a 20-ml volumetric flask. The active ingredient was dissolved with a phosphate buffer at pH 6.5 (prepared with 0.05 M KH_2PO_4 and 0.6 mM NaOH in 1000 ml water). Each sample was centrifuged at 4000 rpm during 10 min. The supernatant were then analyzed after dilution in the buffer.

2.5. Development of calibration models

A Partial least squares (PLS) calibration model was used for the calibration of the NIRS equipment. In PLS, the calibration involves correlating the data in the spectral matrix X with the data in the concentration matrix Y. The X and the Y matrices are reduced to only a few factors using all available information. This model was validated and the risk to use this model in routine was evaluated.

The calibration model was developed with 120 tablets randomly taken in the three concentrations batches. For this step, a leave-one-out cross validation was performed.

The NIRS method was then validated to prove that this analytical method is suitable for its intended use and consequently

Table 1
Validations results of reference UV-method and NIRS method

Parameter	API content (mg)	UV reference method	NIRS method
Trueness (recovery%)	7	97.8	101.3
	14	99.5	99.9
	21	98.9	99.3
Repeatability (RSD%)	7	2.3	2.7
	14	0.6	1.4
	21	1.9	1.3
Intermediate precision (RSD%)	7	2.3	2.7
	14	0.9	1.4
	21	1.9	1.3
Linearity of results	Intercept	−0.076	0.189
	Slope	0.9945	0.9831
	r^2	0.998	0.998
Limits of quantification (LOQ) mg	Lower LOQ	7.0	7.0
	Upper LOQ	21.6	21.6

to show the reliability of the results obtained within well defined limits. To perform this validation, three samples were assayed for each concentration level during 3 days [10,11].

2.6. Agreement between NIRS and reference method

The agreement between the two methods was evaluated by a statistical analysis described by Bland and Altman [12]. A plot of the differences between the two methods results against their average is used to compare conventional and NIRS techniques. This comparison was performed using nine tablets for each concentration level. This analysis is useful to conclude if the new technique is able to replace the conventional one.

2.7. Manufacturing process validation

The aim of this study was to demonstrate that the manufacturing process produces, with each batch, a finished product which complies with defined specifications. Three batches per concentration were produced during 3 days and some tablets were randomly selected from these batches and analyzed with the validated NIRS technique. Ten tablets per batch and per day were analyzed consisting in a total of 90 samples.

3. Results and discussion

3.1. Validation and agreement of the two methods

The NIR spectra used for the calibration after vector normalization are shown in Fig. 1. The region above 7000 cm^{-1} was selected because below this wavenumber the amount of light reaching the detector is low and the detector signal becomes noisy [13,14].

Fig. 2 shows the PLS regression with a determination coefficient value of 0.9979 and a Root Mean Square Error of Cross Validation (RMSECV) of 0.272 mg. The PLS model chosen required three factors and included a wavenumber range from 10,250 to 7000 cm^{-1} with vector normalization and first derivative as spectra pretreatment. These spectra pretreatments correct interferences such as the baseline drift caused by physical state differences of the analyzed samples or maximum absorbance variation.

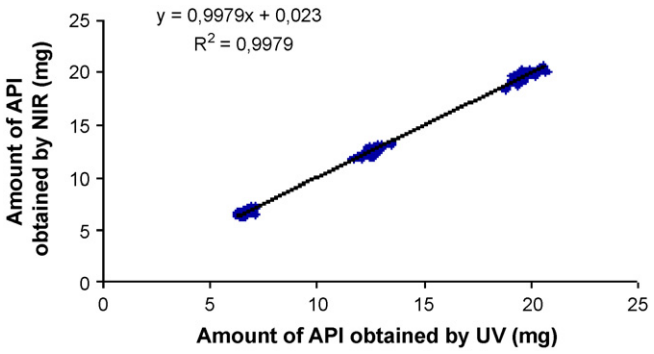

Fig. 2. PLS calibration: API amount obtained by UV vs. NIR spectrophotometry.

The reference method and the NIRS method were validated and the validation results are presented in Table 1. No day effect is observed; repeatability and intermediate precision have the same values.

The accuracy and risk profiles were evaluated for each method [15–17]. The acceptance limits were set at ±10% because each assay was considered as an individual unit assay. Accuracy refers to the closeness of agreement between the test result and the accepted reference value [18,19]. Accuracy profiles are illustrated in Fig. 3 for both analytical methods. The plain line is the relative bias, the dashed lines are the β-expectation tolerance limits and the dotted curves represent the acceptance limits (±10%). As the β-expectation tolerance limits for both analytical methods are included in the acceptance limits, it can be concluded that these methods will provide during routine use results with adequate accuracy.

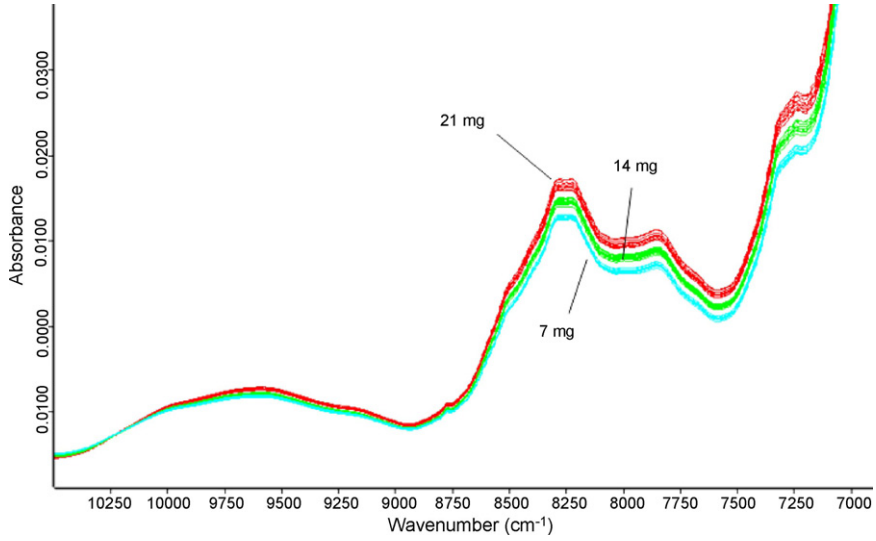

Fig. 1. NIR spectra used for the calibration of the NIR spectrometer.

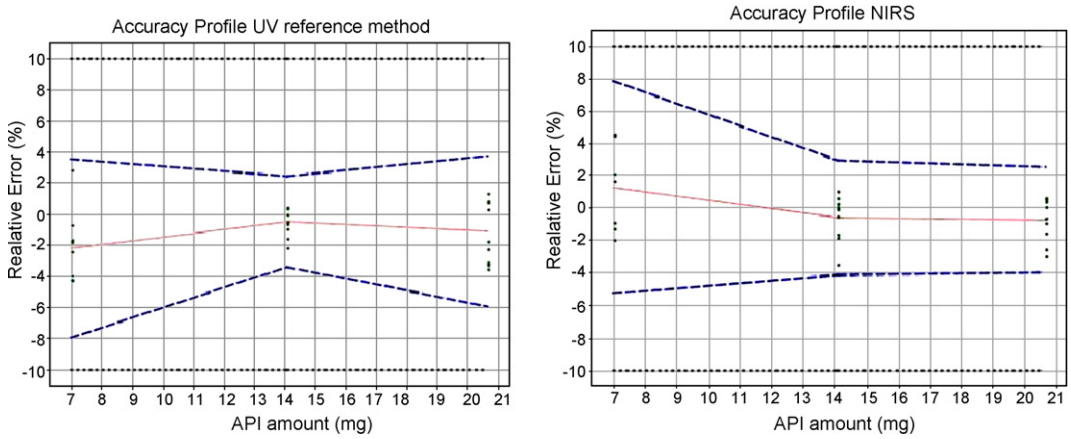

Fig. 3. Accuracy profiles obtained during validation of UV Reference method and NIRS method. The plain line is the relative bias, the dashed lines are the β-expectation tolerance limits and the dotted curves represent the ±10% acceptance limits. The dots represent the individual results.

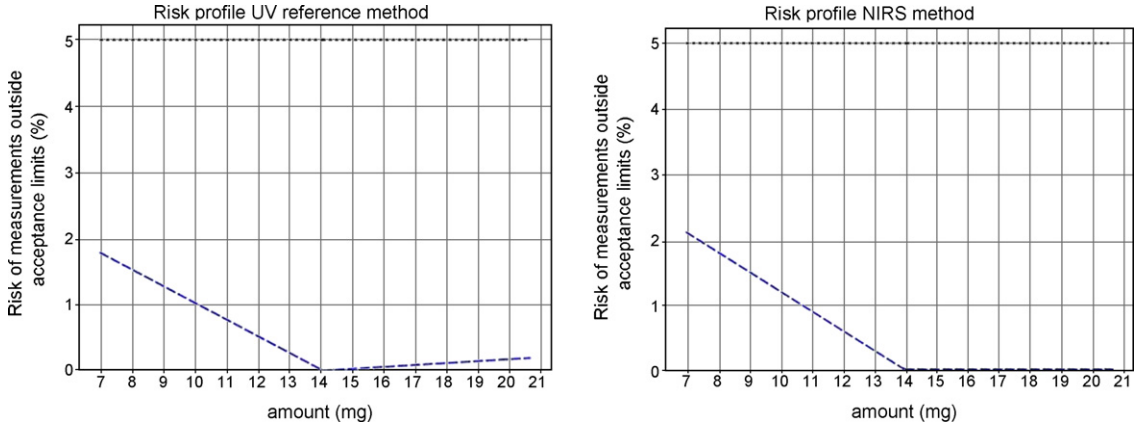

Fig. 4. Risk profiles to have a sample outside the specifications for UV reference method and NIRS method. The dotted line represents the maximum tolerated risk settled at 5%. The dashed line represents the effective risk of having future results falling outside the ±10% acceptance limits.

Based on those accuracy profiles, the risk of having future measurements falling outside the acceptance limits was estimated for each technique. Fig. 4 shows their corresponding risk profiles. For each concentration level, the risk to find future results outside the ±10% acceptance limit is below 5%, the maximum risk level chosen. This risk is smaller for the two highest levels than for tablet containing 7 mg of active principal ingredient (API).

Once validated, the agreement between the two techniques was evaluated to know if the NIR method could replace the reference method. The Bland and Altman plot is showed in Fig. 5. This plot represents the difference between the methods results against their average and displays their agreement. The tolerance interval limits (TI) delimits the area containing 95% of the difference values obtained. As these limits are confined inside the ±10% acceptance limits, the two methods agree sufficiently for the NIRS to replace safely the conventional UV–vis spectrophotometry. For the manufacturing process validation, the NIRS was therefore used during routine analysis to determine if the produced batches give a finished product meeting the quality criteria.

3.2. Manufacturing process validation

For the manufacturing process validation, three tablet batches containing three levels of API amounts were manufactured each

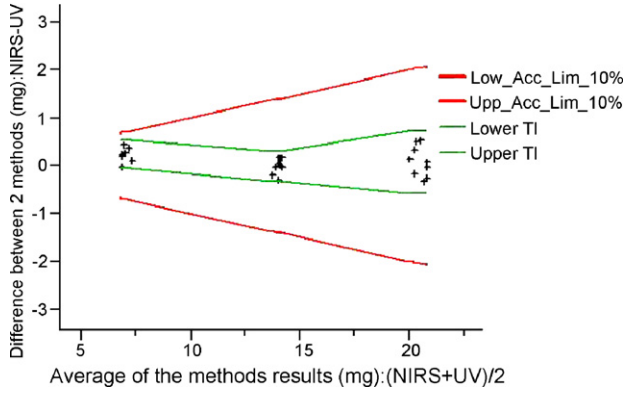

Fig. 5. Bland and Altman's plot: differences between the 2 methods against mean for each concentration level. The continuous lines are the ±10% acceptance limits. The dashed lines are the 95% agreements limits.

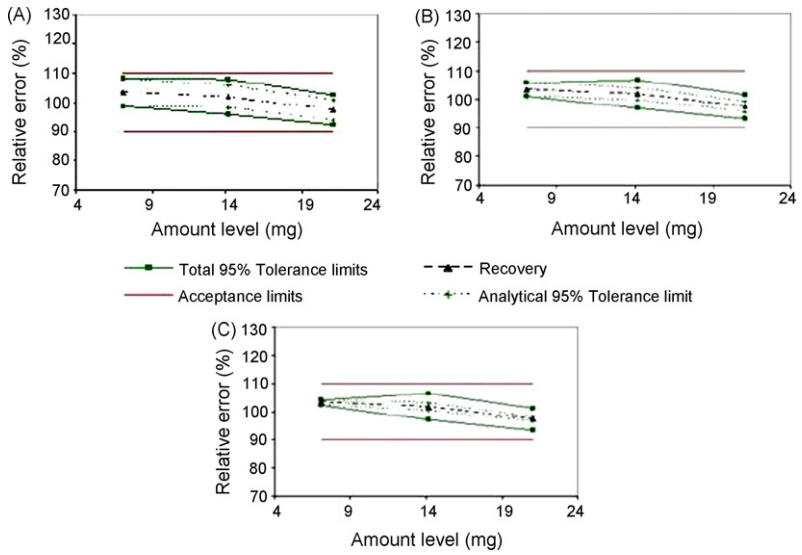

Fig. 6. Accuracy profiles obtained after batches analysis considering the final result of an assay directly (A) or as the mean of 2 (B) or 5 measurements (C).

during 3 different days, resulting in a total of nine batches. Ten tablets were randomly selected from these batches and assayed during 3 days with the validated NIRS technique. To validate all the manufacturing process, it is useful to evaluate the influence of this process and not only the influence of the analytical method as it is often performed. Fig. 6A shows the accuracy profile obtained with the nine batches. As can be seen, the total 95% tolerance intervals are all included in the ±10% acceptance limits demonstrating the ability of the whole process to provide products of adequate quality. In addition, for the three amount levels, we have evaluated the batch-to-batch variation representing the production variability together with the day-to-day and repeatability variation representing the analytical method source of variability. Similarly to the analytical method validation results, no day effect for the three levels was observed. However a batch effect for tablets containing 14 and 21 mg of API (amount levels 2 and 3) was present. As it can be seen on the accuracy profile of Fig. 6A for these two amounts levels, the total 95% tolerance intervals are wider than the analytical 95% tolerance limits due to this batch effect. This means that although the manufacturing process has an influence on the amount of API included in the tablets, overall the drug products produced will be of acceptable quality.

Table 2 contains details about the tolerance limits for each API amount. The total risk to have a future sample outside the specification limits has been also evaluated. This risk including both the risk of the analytical method and the risk of the manufacturing process to obtain a result outside the specification is smaller than 5‰ for the three types of tablets. Therefore it guarantees to the laboratories as well as the regulatory bodies that in the future less than 5 tablet out of 1000 produced will have a quantity of API at more than 10% of the targeted dose.

Then, in order to improve the results, the final result of an assay was considered as the mean of two or five measurements. Fig. 6B and C shows, respectively, the improved accuracy profiles. The batch effect is always present, indeed the only variability that is improved is the repeatability variance of the analytical procedure which is reduced by a factor of $\sqrt{2}$ or $\sqrt{5}$, respectively. In the first situation the risk to have future measurement outside the specifications is reduced to maximum 1‰, whereas in the second one the risk amounts is reduced to maximum 0.4‰ of the API amount. Due to the batch effect observed, a profile with narrower acceptance limits cannot be envisaged in the case of the two highest quantity levels of Diltiazem HCl tablets.

This batch effect could be due to an artifact during the manufacturing process. During the manufacturing process investigation, the only problem met is API agglomerates which appear with the highest API concentrations. Normally, these agglomerates are broken during the manufacturing process but maybe not enough for certain batches. These agglomerates could modify the mixing homogeneity and these homogeneity alterations could be therefore responsible of variations in API tablet amounts.

Table 2
Results of batches analysis and manufacturing process validation

Amount (mg)	Recovery (%)	Total relative upper tolerance limit (%)	Total relative lower tolerance limit (%)	Analytical relative upper tolerance limit (%)	Analytical relative lower tolerance limit (%)	Total risk (%)
7.1	103.3	108.0	98.6	108.0	98.6	0.3
14.2	101.8	107.7	95.9	105.7	97.9	0.4
21.6	97.4	102.5	92.3	100.8	94.0	0.2

4. Conclusions

During this study, we showed that NIRS is an interesting tool as a real time release system. The validation of the manufacturing process has shown that there is not only an influence of the analytical technique but the manufacturing process should also be considered.

We have demonstrated that with our model we have a batch effect and no day effect. It means that the manufacturing process influences the finished product analysis and this should be taken into consideration during the finished product analyses. The risk to obtain future tablets exceeding the specification limits was also estimated. One way to reduce effectively this risk is to consider a result as an average of several independent results. This is a cost effective solution when considering NIRS as quality control technique for batch releases due to the rapidity of the analysis.

A validation of the manufacturing process has therefore been performed which fits with PAT concept and allowed to manage the risk of obtaining out of specifications products.

Acknowledgements

The Ministry of the French Community of Belgium financially supported this work. Thanks are due to the Walloon Region and the European Social Fund for a research grant to E.R. (First Europe Objective 3 project No. 215269). Thanks to Arlenda® for the validation software.

References

[1] E.T.S. Skibsted, Ph.D. Thesis, PAT and Beyond, 2005.
[2] American Food Drug Administration (FDA), Guidance for Industry PAT—A Framework for Innovative Pharmaceutical Manufacturing and Quality Assurance, FDA, 2004.
[3] L. Rodrigues, T. Alves, J. Cardoso, J. Menezes, IDrugs 9 (2006) 44–48.
[4] M. Blanco, M. Alcala, J.M. Gonzalez, E. Torras, J. Pharm. Sci. 95 (2006) 2137–2144.
[5] E.W. Ciurczak, J.K. Drennen, in: D.A. Burns, E.W. Ciurczak (Eds.), Handbook of Near-infrared Analysis, 2001, pp. 609–632.
[6] Y. Chen, S.S. Thosar, R.A. Forbess, M.A. Kemper, R. Rubinovitz, A.J. Shukla, Drug Dev. Ind. Pharm. 27 (2001) 623–631.
[7] J.D. Kirsch, J.K. Drennen, J. Pharm. Biomed. Anal. 19 (1999) 351–362.
[8] M. Laasonen, T. Harmia-Pulkkinen, C. Simard, M. Rasanen, H. Vuorela, Anal. Chem. 75 (2003) 754–760.
[9] T. Li, A.D. Donner, C. Choi, J. Pharm. Sci. 92 (2003) 1526–1530.
[10] B. Boulanger, W. Dewé, P. Chiap, J. Crommen, Ph. Hubert, J. Pharm. Biomed. Anal. 32 (2003) 753–765.
[11] Ph. Hubert, J.-J. Nguyen-Huu, B. Boulanger, E. Chapuzet, P. Chiap, N. Cohen, P.-A. Compagnon, W. Dewé, M. Feinberg, M. Lallier, M. Laurentie, N. Mercier, G. Muzard, C. Nivet, L. Valat, J. Pharm. Biomed. Anal. 36 (2004) 579–586.
[12] J.M. Bland, D.G. Altman, Lancet 1 (1986) 307–310.
[13] J.L. Ramirez, M.K. Bellamy, R.J. Romanach, AAPS PharmSciTech. 2 (2001) 1–10.
[14] J. Gottfries, H. Depui, M. Fransson, M. Jongeneelen, M. Josefson, F.W. Langkilde, D.T. Witte, J. Pharm. Biomed. Anal. 14 (1996) 1495–1503.
[15] R.D. Marini, A.-C. Servais, E. Rozet, P. Chiap, B. Boulanger, S. Rudaz, J. Crommen, P. Hubert, M. Fillet, J. Chromatogr. A 1120 (2006) 102–111.
[16] E. Rozet, R. Morello, F. Lecomte, G.B. Martin, P. Chiap, J. Crommen, K.S. Boos, P. Hubert, J. Chromatogr. B 844 (2006) 251–260.
[17] B. Boulanger, W. Dewe, A. Gilbert, B. Govaerts, M. Maumy-Bertrand, Chemometr. Intell. Lab. Syst. 86 (2007) 198–207.
[18] International Conference on Harmonization (ICH) of Technical Requirements for registration of Pharmaceuticals for Human Use, Topic Q2 (R1): Validation of Analytical Procedures: Text and Methodology, Geneva, 2005.
[19] E. Rozet, A. Ceccato, C. Hubert, E. Ziemons, R. Oprean, S. Rudaz, B. Boulanger, P. Hubert, J. Chromatogr. A 1158 (2007) 111–125.