https://doi.org/10.31925/farmacia.2021.3.11 **ORIGINAL ARTICLE**

APPLYING THE PRINCIPLES OF QUALITY BY DESIGN (QbD) COUPLED WITH MULTIVARIATE DATA ANALYSIS (MVDA) IN ESTABLISHING THE IMPACT OF RAW MATERIAL VARIABILITY FOR EXTENDED RELEASE TABLETS

KINGA ILYÉS [1], TIBOR CASIAN [1*], DANA HALES [1], GHEORGHE BORODI [2], LUCIA RUS [3], RAREȘ ȘTIUFIUC [4,5], IOAN TOMUȚĂ [1]

[1]*Department of Pharmaceutical Technology and Biopharmacy, Faculty of Pharmacy, "Iuliu Haţieganu" University of Medicine and Pharmacy, Cluj-Napoca, Romania*
[2]*National Institute for Research and Development of Isotopic and Molecular Technologies, Cluj-Napoca, Romania*
[3]*Department of Drug Analysis, Faculty of Pharmacy, "Iuliu Haţieganu" University of Medicine and Pharmacy, Cluj-Napoca, Romania*
[4]*Department of Pharmaceutical Biophysics, Faculty of Pharmacy, "Iuliu Haţieganu" University of Medicine and Pharmacy, Cluj-Napoca, Romania*
[5]*MedFuture Research Centre for Advanced Medicine, "Iuliu Haţieganu" University of Medicine and Pharmacy, Cluj-Napoca, Romania*

**corresponding author: casian.tibor@umfcluj.ro*

Manuscript received: June 2020

Abstract

The raw material is acknowledged to be a source of variability, however, the conventional, empirical development approach does not offer much information regarding its critical attributes and how processes can be modulated to remain in the constant quality region of the product. The study aimed to develop extended release hydrophilic matrix tablets with indapamide based on the Quality by Design (QbD) concept, evaluating the impact of interchanging different types and suppliers of raw material on the finished product quality profile. Results showed significant *in vitro* release test variability, with 16 - 71% failure rates when compared to four different EMA and FDA dissolution specification recommendations. Design of experiments based impact assessment concluded that the active pharmaceutical ingredient, hydroxypropyl methylcellulose and compression force was accountable for the variation, while orthogonal partial least squares (O2PLS) based root cause analysis extension redefined results in term of critical material attributes. Findings suggest that a risk-based, multivariate analysis assisted control strategy for the incoming raw materials could prevent quality concerns within routine manufacturing.

Rezumat

Materia primă este recunoscută a fi o sursă de variabilitate, totuși abordarea de dezvoltare empirică, convențională nu oferă multe informații cu privire la atributele critice și modul în care procesele pot fi modulate pentru a rămâne în regiunea de calitate constantă a produsului. Studiul propune dezvoltarea de comprimate cu indapamidă de tip matrice hidrofilă cu eliberare prelungită pe baza conceptului de calitate prin design (*Quality by Design* – QbD), cu evaluare concomitentă a impactului materiilor prime asupra calității finale. Rezultatele au evidențiat o variabilitate semnificativă în cadrul testului de eliberare *in vitro*, cu rate de eșec între 16 - 71% în comparație cu patru recomandări diferite privind specificațiile de dizolvare EMA și FDA. Evaluarea impactului pe baza metodei planurilor experimentale a concluzionat că substanța activă, hidroxipropil metilceluloza și forța de comprimare ar fi responsabile pentru variație, în timp ce identificarea cauzei prin metoda celor mai mici pătrate ortogonale (O2PLS) a redefinit rezultele în termenii atributelor critice ale materialelor. Concluziile sugerează că o strategie de control, bazată pe analiza multivariată, ar putea preveni problemele de calitate din cadrul unei producții de rutină.

Keywords: Quality by Design**,** multivariate analysis, controlled release, indapamide

Introduction

In the pharmaceutical domain, the notion of quality is considered a reflection of safety and efficiency of a certain product. Due to several quality assurance system failures, translated in the form of drug recalls and shortages, US FDA (United States Food and Drug Administration) started promoting new notions within the pharmaceutical professional community, like PAT (Process Analytical Technology), QbD (Quality by Design), emerging technologies (continuous manufacturing, 3D-printing), that are anticipated to enable real time control, giving a systematic understanding of products/ processes, simplifying production etc. in order to

improve access to high-quality products [12, 16, 18, 27, 36, 38].

The notion of QbD implies a systematic, risk based R&D (research and development) process, having as pillars the ICH guidelines: Q8 (pharmaceutical development) [22], Q9 (quality risk management) [23] and Q10 (pharmaceutical quality system) [24]. On the regulatory scale, the notion exists only as a recommendation, still the FDA and the European Medicines Agency (EMA) even launched a joint pilot program to facilitate the implementation and harmonize regulatory decisions with regards to QbD applications [10].

This concept relies on the premises that "quality cannot be tested into the product, it should be built in", using statistical quality risk management tools to improve product/process knowledge and control. Experiments are conducted in a systematical manner, based on a Design of Experiments (DoE). Statistical models enable the identification of the critical material attributes (CMA) and process parameters (CPP), defining their individual/additive relation to final product quality. Based on the global process equation and the predefined quality target profile (QTP), a design space (DS) is determined, that serves as a scientific fundament to establish process limits (batch production record) and control strategies (test methods, specifications raw/in-process/finished product). It serves preventive, but also corrective purposes, the QbD R&D helping manufacturing investigations and setting appropriate corrective and preventive actions (CAPA). Although it implies a more extensive development process, in the integral Chemistry Manufacturing Control (CMC) perspective, the QbD R&D is presumed to improve cost-effectiveness [50] (Figure 1).

A 9-year retrospective study, conducted by the FDA Center for Drug Evaluation and Research (CDER) on 370 field alert reports on dissolution failures, showed that one third was still pending investigation as no root cause could be determined. Also, it was established that modified release (MR) products posed more problems than immediate release (IR) formulations [46].

In a common root cause analysis, the "8M" sources of error are examined as a part of the preliminary risk assessment: material, methods, measurement, machines, milieu, man power, management, money. The impact of raw material (RM) variability is acknowledged, but still the control strategy mostly relies on a simplified approach covering the following tests: description, identification, assay, impurities.

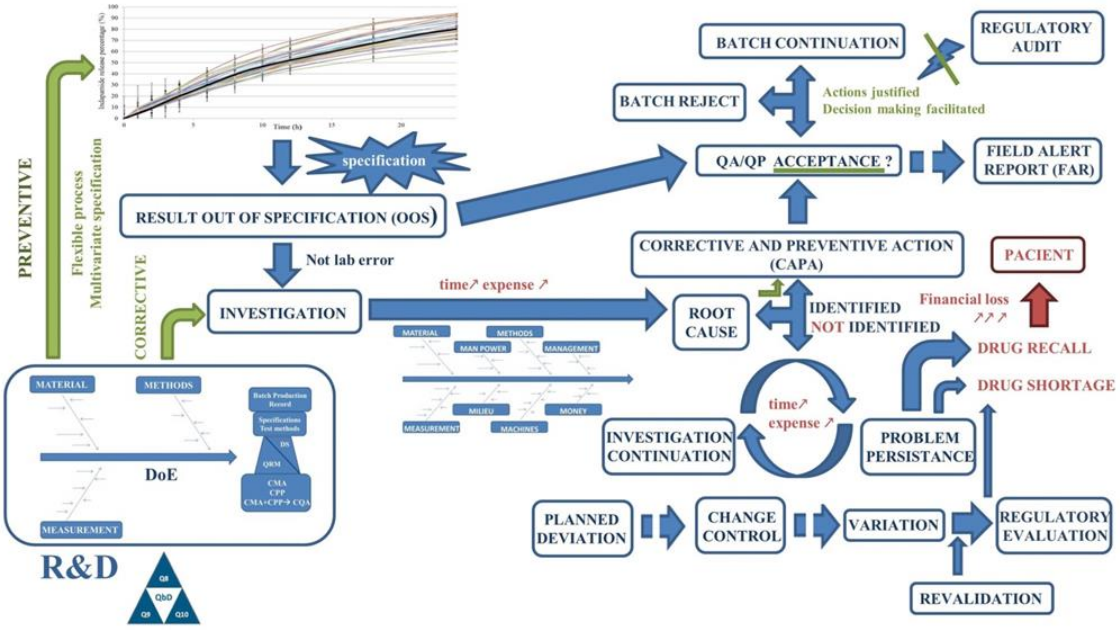

Figure 1.

Schematic representation of quality problem management in routine manufacturing, with possible negative outcomes (marked in red) and the QbD-based R&D areas of improvement (marked with green)

CMA – critical material attributes, CPP – critical process parameters, CQA – critical quality attribute (product), QRM – quality risk management, DS – design space, QA – quality assurance, QP – qualified person, CAPA – corrective and preventive actions, OOS – out of specification (result), FAR – field alert report, R&D – research and development, QbD – Quality by Design, DoE – Design of Experiments

The study constituted a Design of Experiments (DoE) mediated robustness testing on extended release tablets, evaluating the impact of RM variability, simulated by interchanging different suppliers of the same material. As opposed to the common approach, where in the Manufacturing Authorization (MA) specific suppliers

are mentioned, for this study the focus was shifted on the raw material characteristics. For the materials that clearly differed in typology, the sort was mentioned, but for others, delimitation was made according to the supplier codification. Typology was taken into consideration in the context of process flexibility, evaluating if it is necessary to restrict production only to a certain sort of excipient.

Initial impact assessment findings were extended by applying multivariate data analysis (MVDA), enabling the redefinition of factors in terms of physicochemical descriptors, and also providing efficient quality control opportunities. The importance of the solid state characterization [5] and the potential of applying historical data in terms of retrospective QbD [45] were also evaluated within this step.

The study comes in completion to earlier investigations related to active pharmaceutical ingredient (API) variability on product quality [13, 34] and excipient evaluation in context of second supplier qualification [20]. DoE coupled with MVDA enables the possibility of simultaneous assessment of API and excipient effect on quality outcomes, with the potential to define the areas of limitation and flexibility within the material control and process modulation.

Materials and Methods

Materials

Indapamide was used as a model API, being able to compare five different material quality profiles from three different suppliers (codified as S.A, S.B, S.C). Supplier A and B, each provided two different batches, the ones from B being wittingly micro (S.B1) and macro (S.B2) scaled.

As a filler, different types of monohydrate lactose were taken into consideration: FlowLac100 (FL 100) and Tablettose 80 (TB 80) from Meggle (Wasserburg, Germany), SuperTab 11SD (ST 11SD) and SuperTab 14SD (ST 14SD) from DFE Pharma (Goch, Germany). In case of TB 80, two different batches were evaluated (TB80 b1, TB80 b2).

Seven different, QbD grade, batches of hydroxypropyl methylcellulose (HPMC) Methocel Premium CR K15M (Ph. Eur. 2208) were kindly donated by Dow Chemical Company (Midland, USA). Six batches presented the upper and lower values with regards to viscosity, hydroxypropoxyl content (%HP), particle size (LV – low viscosity, HV – high viscosity, LHP – low hydroxypropoxyl content, HHP – high hydroxypropoxyl content, LPS – low particle size, HPS – high particle size) and one served as a centre point between all these values.

For polyvinylpyrrolidone (PVP) three different suppliers and two different typologies were used: K25 (S.D, S.E) and K30.

Magnesium stearate (Mg-St) was provided by other three suppliers, which were codified S.F, S.G and S.H.

Colloidal silicon dioxide was not varied between the formulations; all experimental runs were prepared using Aerosil 200 from Evonik (Essen, Germany).

All other materials used within the study were of analytical grade.

Technological process

The powder blends were prepared manually by geometrical mixing, followed by compression with an EK-0 eccentric tableting machine (Korsch, Germany) equipped with 7 mm punches.

The indapamide tablets (1.5 mg/tablet, average weight 210 mg) were prepared based on the same quantitative formula, but with qualitative variations regarding the raw materials used. The general formula included: indapamide (0.71%, w/w), monohydrate lactose (61.60%, w/w), HPMC (35.01%, w/w), PVP (1.43%, w/w), colloidal silicon dioxide (0.50%, w/w), Mg-St (0.75%, w/w).

Besides formulation parameters, the effect of three processing conditions was also evaluated. Compression force was modulated in the 150 to 300 N range. Also, the relevance of two operational "habits" used within routine production was tested: two different initial blending methods and the addition of magnesium stearate within the process. The two different blending methods involved the preliminary mixing of the API in a proportion of 1:2 either with PVP (method A) or monohydrate lactose (method B) then adding the rest of the excipients in a 1:1 rate to the existing mass according to a geometrical mixing methodology. The hypotheses that magnesium stearate added at the end assures better flow properties to the blend was also evaluated, adding it half way into the mixing process (during) or at the final phase (end).

The rational on which different types/batches of raw material and processing conditions were chosen for a specific formulation was based on a D-optimal experimental design (Table I). The experimental runs were prepared and analysed in a controlled, randomized order, to reduce external, milieu variability.

Table I

D-optimal screening experimental design

Exp. Name	X1	X2	X3	X4	X5	X6	X7	X8
N1	S.B1	FL 100	LV	K30	S.F	A	during	150
N2	S.B2	TB 80 b1	LPS	K30	S.F	A	during	300
N3	S.A2	TB 80 b2	HPS	K25 S.E	S.F	A	during	150
N4	S.A1	FL 100	LHP	K25 S.D	S.F	A	during	300
N5	S.C	TB 80 b1	centre	K30	S.G	A	during	150
N6	S.B2	ST 11SD	HV	K25 S.E	S.G	A	during	150

Exp. Name	X1	X2	X3	X4	X5	X6	X7	X8
N7	S.B1	ST 14SD	HHP	K25 S.E	S.G	A	during	300
N8	S.A1	TB 80 b2	centre	K30	S.H	A	during	300
N9	S.A2	ST 14SD	HV	K30	S.F	B	during	300
N10	S.A1	ST 14SD	LPS	K25 S.D	S.G	B	during	150
N11	S.C	FL 100	HPS	K25 S.D	S.G	B	during	300
N12	S.B1	ST 11SD	LHP	K25 S.E	S.H	B	during	300
N13	S.A2	TB 80 b1	LV	K25 S.D	S.H	B	during	300
N14	S.B2	TB 80 b2	HHP	K25 S.D	S.H	B	during	150
N15	S.C	ST 11SD	LV	K25 S.D	S.F	A	end	300
N16	S.A2	ST 11SD	HHP	K30	S.G	A	end	300
N17	S.B1	TB 80 b1	HV	K25 S.D	S.G	A	end	300
N18	S.C	ST 14SD	LHP	K25 S.E	S.H	A	end	150
N19	S.A2	FL 100	LPS	K25 S.E	S.H	A	end	150
N20	S.B2	ST 14SD	HPS	K25 S.D	S.H	A	end	300
N21	S.A1	TB 80 b1	HHP	K25 S.E	S.F	B	end	150
N22	S.C	TB 80 b2	LPS	K25 S.E	S.F	B	end	300
N23	S.B2	FL 100	centre	K25 S.E	S.F	B	end	300
N24	S.B1	ST 11SD	centre	K25 S.D	S.F	B	end	150
N25	S.B2	TB 80 b2	LHP	K30	S.G	B	end	150
N26	S.A1	TB 80 b2	LV	K25 S.E	S.G	B	end	300
N27	S.C	FL 100	HV	K30	S.H	B	end	150
N28	S.A1	ST 11SD	HPS	K30	S.H	B	end	150
N29	S.C	FL 100	centre	K25 S.E	S.H	B	end	225
N30	S.C	FL 100	centre	K25 S.E	S.H	B	end	225
N31	S.C	FL 100	centre	K25 S.E	S.H	B	end	225

X1 – indapamide supplier, X2 – lactose monohydrate type, X3 – hydroxypropyl methylcellulose K15M batch, X4 – polyvinylpyrrolidone type/supplier (K25), X5 – magnesium stearate supplier, X6 – blending method, X7 – magnesium stearate addition, X8 – compression force, FL 100 – FlowLac 100, TB 80 – Tablettose 80 (different batches b1, b2), ST 11SD – SuperTab 11SD, ST 14SD – SuperTab 14SD, LV – low viscosity, HV – high viscosity, LHP – low hydroxypropoxyl content, HHP – high hydroxypropoxyl content, LPS – low particle size, HPS – high particle size, S. – supplier

Pharmaceutical characterization (intermediate and final product quality)

Pharmaco-technical analysis was done on the powder blends and tablets. The in-process controls were related to the flow properties, loss on drying and the final product quality was evaluated based on the hardness and release characteristics at multiple time points.

For the result accuracy, the tests were conducted in triplicate for each experimental run.

Flow characteristics. Flow characteristics (Y1-Y4) were evaluated according to European Pharmacopoeia (Ph. Eur. 9.0) methods from monographs 2.9.16 (flowability), 2.9.34 (bulk density and tapped density of powders), 2.9.36 (powder flow). Apparatus included BEP2 (Copley Scientific, UK) and SVM 121 (Erweka, Germany).

Loss on drying (LOD). Loss on drying (Y5) was tested with moisture analyser MB45 (Ohaus, USA), the drying procedure consisting in the exposure of 1 g powder samples to a temperature of 80°C, the moisture percentage being recorded at the moment when the mass loss did not exceed 1 mg in 90 s.

Hardness. The hardness (Y6) was tested on Dr. Schleuniger 6D (Pharmatron, Switzerland).

In vitro drug release. Drug release experiments were carried out with PTWS 100 (Pharmatest, Germany) USP type I (basket) dissolution apparatus in 900 mL phosphate buffer pH 6.8 at 37 ± 0.5°C and 100 rpm stirring rate. 2 mL aliquots of the dissolution medium were withdrawn in the course of 24 h, evaluating the dissolution percentage at 10 time points (Y7 - Y16): 1, 2, 3, 4, 6, 8, 10, 12, 18 and 24 h. Each withdrawn sample was replenished using the same amount of dissolution medium. The samples were collected, filtered and assayed using HPLC 1100 (Agilent, USA) with UV-detection. Data was processed and recorded *via* Chemstation software.

The assay constituted an adaptation of a previously referenced method [40], before use validated according to ICH Q2 [25]. The quantification was done according to the linear regression of standard indapamide in pharmaceutical reconstructed formula in the range of 0.13 - 2 µg/mL.

The chromatographic separation was carried out at 35°C on a Zorbax SB-C18 analytical column 100 mm x 3 mm, 3.5 µm, using as the mobile phase a mixture of phosphoric acid solution 0.1% and aceto-nitrile (50:50, v/v) at a flow rate of 0.8 mL/min. In the given conditions, indapamide had a retention time of 1.26 min. 20 µL of each collected dissolution sample was injected in the system, separated *via* RP-HPLC and quantified at 240 nm.

Indapamide solid state characterization

Laser diffraction (LD). Particle size measurements were conducted with laser diffraction analyser Mastersizer 3000 equipped with wet dispersion unit Hydro MV (Malvern Instruments Ltd, UK).

Water (refractive index = 1.33) was chosen as the dispersion medium, indapamide being characterized as a practically insoluble compound with a water solubility of 75 mg/L [30]. 5 mg indapamide (refractive index = 1.693, absorption index = 0.01) was initially mixed with 3 drops of polysorbate 80 (Merck, Darmstadt, Germany) solution of 1% concentration, then suspended in 10 mL distilled water and finally sonicated for 30 s.

Each measurement was carried out 5 times, the time of one analysis constituting 10 s.

The measurement range was kept between 0.01 and 3500 μm. The particle size was calculated based on the Mie theory of light scattering, which is also appropriate for samples that may have particles under 50 μm [33]. Particle size distribution volume based percentiles D10, D50, D90 and span were recorded. The span was calculated, based on equation 1:

$$\text{Span} = \frac{\text{D90 - D10}}{\text{D50}}, \quad (1).$$

Differential calorimetry (DSC). DSC studies were carried out on a DSC 822e/700 (Mettler Toledo, USA), using 40 μL aluminium crucibles with 2.5 - 3 mg of sample. Working conditions covered the 25 - 400°C range with a heating rate of 10°C/min, under dynamic N_2 atmosphere with a flow rate of 50 mL/min. Thermograms were processed in STARe v12.10 (Mettler Toledo, USA).

The onset and peak temperatures, mean normalized enthalpy (MNE) corresponding to the endothermic peak of indapamide were recorded.

X-ray powder diffraction (XRPD). The X-ray diffraction patterns of the samples were obtained by using a Bruker D8 Advanced X-ray diffractometer (Bruker AXS, Germany) equipped with a Ge (1 1 1) monochromator in the incident beam (in order to obtain only Cu Kα1 radiation) and LynxEye super speed detector. Measurements were conducted in the angular 2θ range of 3 - 40 degrees.

ConQuest was used to search and compare the crystal structure of the evaluated batches of indapamide to the ones available in the Cambridge Structural Database (CSD). Sample grinding was avoided not to disrupt the solid-state information.

Raman spectroscopy. Spectral acquisition was done with a NT-MDT NTEGRA Spectra AFM inVia spectrometer (Renishaw, UK) in static scan configuration over the 503 cm^{-1} - 1800 cm^{-1} spectral range using an excitation wavelength of 785 nm and an objective of 20x. An exposure time of 10 seconds and 20 accumulations were used to obtain qualitative spectra. The spectral resolution was set to 1 cm^{-1}.

Data analysis

Initial raw material impact assessment was based on the D-optimal experimental design, by fitting for each response (Y1 - Y16) a polynomial equation, that described through its coefficients the magnitude and direction of effects exerted by the input variables. Data was fitted using projections to latent structures by partial least squares (PLS) method. Model performance was evaluated considering R^2 (goodness of fit), Q^2 (goodness of prediction), model validity and reproducibility under identical experimental conditions. The reference values considered for a good and stable model were: $Q^2 > 0.5$, $R^2 - Q^2 < 0.3$, reproducibility > 0.5, validity > 0.25) [9].

ANOVA was applied for model significance and lack of fit testing. For model significance the size of modelled variability was compared with residuals, whereas for the lack of fit test, the replicate error and model error were taken into account. Interpretations were made based on F-test probability values (p): for model significance $p < 0.05$ and for lack of fit $p > 0.05$. Model interpretation was done by generating scaled and cantered coefficient plots. DoE data was analysed through Modde Pro 11 (Sartorius Stedim, Sweden).

The root cause investigation was extended using multivariate data analysis methods. For this step, the main factors affecting drug release were considered, as demonstrated by DoE analysis results. Each experimental run was redefined by adding the correspondent raw material physicochemical descriptor. Therefore, the input matrix (X) consisted of: indapamide (D10, D50, D90, span, onset and peak temperatures, MNE), HPMC (viscosity, hydroxypropoxyl content, particle size) physicochemical descriptors and compression force value. For indapamide, solid state characterization performed as presented under section 2.4 provided additional data, while the HPMC descriptors were taken from the Batch Certificate of Analysis (CoA) documents supplied by the vendor. Using the dissolution data as the response matrix (Y), an O2PLS (two-way orthogonal PLS) model was employed to determine the CMA responsible for the outcome variability, providing an improved overview of the dissolution profile variability sources. Prior to model development, input variables (X) were scaled to unit variance and response variables (Y) were cantered. Model interpretation was done by generating loading coefficient plots that describe unique and joint sources of variability between X and Y. Simca14 (Sartorius Stedim, Sweden) was used for multivariate data analysis.

Results and Discussion

Intermediate and end product characteristics

Ensuring an appropriate release profile to suit API characteristics and pharmacological destination of product is the most important quality attribute for a modified release dosage form. Indapamide, a thiazide-like diuretic, is used as a first line pharmacological treatment in case of hypertension [49]. Characterized by a water solubility of 75 mg/L and a n-octanol-water partition coefficient (logP) of 2.2, according to the biopharmaceutical classification system (BCS), it

is considered a class II compound with low solubility and high permeability [30, 47]. The biological half-life is long (approx. 16 h), still a MR formulation is preferred to the IR, because it delivers a smoother pharmacokinetic profile, avoiding increased plasma peak concentrations, which may determine unwanted side effects [44].

The statistical differences between MR products and the IR counterparts, with regards to the quality concerns, may be partly attributed to the complicated formulation designs and partly to the extensive processing conditions [46]. In the present study, the technological process was kept simple to gain a clear perspective with regards to the impact of raw material variability. Involving only two preparation phases, the probability of introducing systematical errors with each step was reduced.

Preliminary analysis of the results (Table II) showed that powder blends presented comparable flow properties, adequate LOD% and, as expected, variable hardness.

Table II

Pharmaceutical characterization results

Batch	Flow time (sec)	Angle of repose (degrees)	Hausner ratio	Carr index	Loss on drying (%)	Hardness (N)
N1	4.28 ± 0.71	42.25 ± 0.92	1.32 ± 1.57E-16	24.32 ± 1.15E-14	1.44 ± 0.15	142.33 ± 11.59
N2	3.81 ± 0.24	43.26 ± 0.49	1.25 ± 0.03	20.11 ± 1.68	1.53 ± 0.05	250.33 ± 12.06
N3	3.59 ± 0.15	43.96 ± 0.81	1.30 ± 0.01	23.18 ± 0.51	1.52 ± 0.07	147.67 ± 14.05
N4	3.46 ± 0.45	44.47 ± 1.50	1.30 ± 1.57E-16	22.97 ± 7.54E-15	1.48 ± 0.04	308.50 ± 17.86
N5	3.97 ± 0.59	43.32 ± 0.60	1.32 ± 0.01	24.04 ± 0.84	1.39 ± 0.09	150.33 ± 5.03
N6	3.66 ± 0.13	45.55 ± 0.84	1.30 ± 1.57E-16	22.98 ± 7.53E-15	1.54 ± 0.07	145.00 ± 1.00
N7	4.22 ± 0.49	43.08 ± 0.70	1.30 ± 0.01	23.35 ± 0.59	1.29 ± 0.09	193.00 ± 2.65
N8	3.74 ± 0.44	45.07 ± 1.16	1.27 ± 0.01	21.13 ± 0.54	1.40 ± 0.10	192.75 ± 9.74
N9	3.81 ± 0.12	42.95 ± 1.13	1.25 ± 0.02	20.08 ± 1.25	1.47 ± 0.15	268.00 ± 11.36
N10	3.62 ± 0.35	43.57 ± 0.41	1.31 ± 0.01	23.59 ± 0.7	1.45 ± 0.10	179.00 ± 2.65
N11	4.26 ± 0.19	41.86 ± 0.34	1.32 ± 0.02	24.50 ± 1.02	1.59 ± 0.11	214.67 ± 11.59
N12	3.14 ± 0.27	46.41 ± 0.52	1.32 ± 0.01	24.43 ± 0.40	1.38 ± 0.09	223.67 ± 4.51
N13	4.31 ± 0.25	40.69 ± 0.38	1.30 ± 0.001	22.86 ± 0.09	1.40 ± 0.10	218.33 ± 15.57
N14	3.55 ± 0.19	43.49 ± 0.56	1.31 ± 0.01	23.40 ± 0.38	1.49 ± 0.03	165.33 ± 1.15
N15	3.26 ± 0.36	42.27 ± 1.49	1.28 ± 0.001	21.87 ± 0.09	1.48 ± 0.17	309.00 ± 10.00
N16	3.70 ± 0.30	45.31 ± 1.02	1.28 ± 0.02	21.67 ± 1.21	1.36 ± 0.05	201.33 ± 7.64
N17	4.02 ± 0.12	43.48 ± 0.72	1.29 ± 0.01	22.43 ± 0.39	1.35 ± 0.14	196.67 ± 2.08
N18	5.12 ± 0.79	40.74 ± 1.68	1.33 ± 0.02	24.65 ± 1.03	1.67 ± 0.08	148.33 ± 8.08
N19	3.8 ± 0.10	43.06 ± 0.31	1.26 ± 0	20.67 ± 6.2E-15	1.42 ± 0.05	141.33 ± 8.02
N20	4.29 ± 0.25	41.61 ± 1.03	1.30 ± 0.01	22.79 ± 0.35	1.68 ± 0.11	255.33 ± 10.12
N21	3.51 ± 0.72	44.06 ± 0.97	1.24 ± 0.01	19.20 ± 0.93	1.32 ± 0.06	151.67 ± 15.01
N22	3.44 ± 0.35	45.14 ± 0.62	1.32 ± 0.01	24.18 ± 0.50	1.21 ± 0.08	269.67 ± 3.21
N23	4.41 ± 0.21	42.73 ± 0.82	1.27 ± 0.01	21.37 ± 0.33	1.46 ± 0.04	307.00 ± 5.00
N24	3.49 ± 0.40	46.45 ± 0.72	1.30 ± 0.01	22.97 ± 0.36	1.51 ± 0.14	154.33 ± 15.95
N25	4.40 ± 0.67	46.68 ± 2.40	1.34 ± 0.01	25.32 ± 0.56	1.55 ± 0.10	162.67 ± 3.06
N26	3.63 ± 0.37	45.08 ± 1.49	1.30 ± 0.01	22.95 ± 0.65	1.27 ± 0.13	241.75 ± 12.26
N27	3.91 ± 0.10	43.08 ± 0.66	1.28 ± 0.03	21.60 ± 1.77	1.57 ± 0.10	164 ± 10.44
N28	3.66 ± 0.35	43.94 ± 0.75	1.28 ± 1.57E-16	21.71 ± 8.7E-15	1.85 ± 0.16	147 ± 13.89
N29	4.30 ± 0.38	44.32 ± 0.82	1.25 ± 1.57E-16	19.74 ± 9.06E-15	2.08 ± 0.04	190.5 ± 7.05
N30	4.11 ± 0.19	43.13 ± 0.80	1.25 ± 0.001	20.22 ± 0.08	2.05 ± 0.13	203.00 ± 16.70
N31	4.13 ± 0.34	43.32 ± 0.40	1.24 ± 0	19.48 ± 4.35E-15	1.97 ± 0.20	205.67 ± 4.16

*Values represent the mean ± standard deviation of triplicate tests

According to the Ph. Eur. 9.0 based classification of powder flow properties, flowability of blends was defined mostly as passable. The experimental design runs presented a 40.69 - 46.68 angle of repose, 1.24 - 1.34 Hausner ratio and 19.2 - 25.32 Carr index. Maximum determined LOD was 2.08%, which indicates a relatively acceptable amount of volatile matter (including water) that can impact product quality (e.g. compressibility, degradation rate).

Applying compression force on two levels in tablet preparation phase generated variability in tablet crushing strength. Tablet hardness ranged from 141 (N19) to 309 (N15) N.

Dissolution variability in the 24 h *in vitro* release experiments was considerable and presented increasing tendency towards later sampling points (Figure 2). At the first time point (1 h) release percentages ranged from 3.67% (N14) to 9.46% (N21), whereas at the last time point (24 h) the differences in released indapamide increased significantly, ranging from 60.50% (N9) to 94.30% (N8).

To assess the dissolution variability in terms of quality compliance, the FDA and EMA guidelines on modified release products were considered [11, 14]. According to the guideline recommendations, the dissolution specification for modified release oral dosage forms is established in minimum 3 time points:

an early time point to exclude dose dumping and/or to characterize initial dose (typically 20 to 30% dissolved), at least one point to ensure compliance with the shape of the dissolution profile (around 50% dissolved) and one to ensure that the majority of the active substance has been released (Q = 80%). Based on the dissolution profile of the evaluated batches, the 4, 12 and 24 h time points would constitute the optimal, minimum number of sampling points for the integral process monitoring.

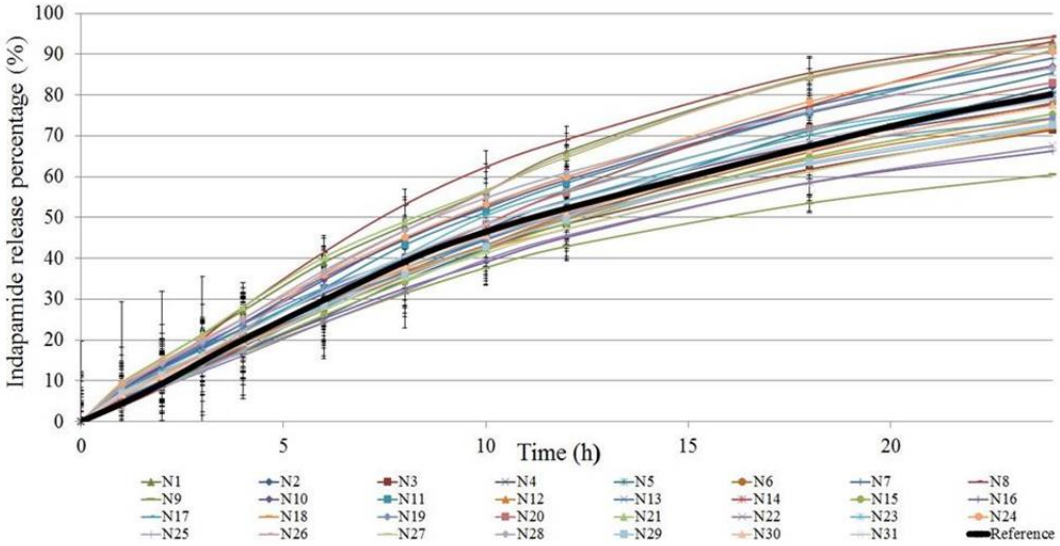

Figure 2.
Dissolution profiles of experimental run formulations opposed to the reference product (Tertensif® SR) – bolded line, highlighting the degree of deviations from the target dissolution profile

To simulate the likelihood of failure of the evaluated 31 experimental runs in the routine manufacturing context (Table III), the dissolution data was tested against 4 different specification approaches: (1) – based on previously the mentioned EMA recommendations, with broad intervals, simulating the condition when reference product profile is not taken into account: 4 h (10 - 30%), 12 h (40 - 60%), 24 h (min 80%); (2) – based on the FDA recommendations, comparison with reference product - Tertensif® SR (Les Laboratoires Servier, France), recommended range for any dissolution time point is ± 10% to the mean dissolution profile of reference product; (3) – based on the FDA recommendation extension, maximum ± 25% compared to the mean dissolution values of reference product can be still considered acceptable; (4) – based on fit factors, f1 (difference factor) and f2 (similarity factor): for comparable dissolution profiles f1 should be maximum 15 and f2 should range in the interval from 50 to 100 [6].

$$f1=\frac{\sum_{i=1}^{n}|R_i-T_i|}{\sum_{i=1}^{n}R_i}\cdot 100, \qquad (2),$$

$$f2=50\cdot\log\left\{\left[1+\frac{1}{n}\cdot\sum_{i=1}^{n}\left(R_i-T_i\right)^2\right]^{-0.5}\cdot 100\right\}, \quad (3).$$

Fit factors are calculated based on equations (Eq. 2) and (Eq. 3), where n represents the number of time points, while R_i and T_i the release percentages for reference/test product at i time point. For the f1/f2 based comparisons, all sampling points were considered.

Table III
Dissolution profile quality compliance to FDA and EMA guideline recommendations

	Method-1			**Method-2**			**Method-3**			**Method-4**	
Criteria of acceptance	4 h: 10 - 30% 12 h:40 - 60% 24 h: min 80%			± 10% mean dissolution profile reference product			± 25% mean dissolution profile reference product			f1 max 15 f2 50 - 100	
Sample	4 h	12 h	24 h	4 h	12 h	24 h	4 h	12 h	24 h	f1	f2
N1	P	F	P	F	F	F	P	P	P	25.24	50.77
N2	P	P	P	F	P	P	P	P	P	6.57	76.42
N3	P	P	F	P	P	F	P	P	P	10.22	67.57
N4	P	P	F	F	P	P	P	P	P	6.32	78.01
N5	P	P	P	P	P	P	P	P	P	4.50	80.83
N6	P	P	F	P	P	P	P	P	P	5.19	80.77
N7	P	P	P	F	F	F	P	P	P	15.36	61.16
N8	P	F	P	F	F	F	F	F	F	30.57	45.73

	Method-1			Method-2			Method-3			Method-4	
Criteria of acceptance	4 h: 10 - 30% 12 h:40 - 60% 24 h: min 80%			± 10% mean dissolution profile reference product			± 25% mean dissolution profile reference product			f1 max 15 f2 50 - 100	
Sample	4 h	12 h	24 h	4 h	12 h	24 h	4 h	12 h	24 h	f1	f2
N9	P	P	F	F	F	F	P	P	P	19.70	51.10
N10	P	P	P	F	F	P	P	P	P	14.70	62.83
N11	P	P	P	P	F	F	P	P	P	12.39	63.08
N12	P	P	F	P	P	P	P	P	P	7.96	73.63
N13	P	P	F	P	P	P	P	P	P	3.43	88.20
N14	P	P	P	P	P	F	P	P	P	9.38	63.45
N15	P	P	F	F	P	P	P	P	P	8.38	71.56
N16	P	P	F	F	F	F	P	P	P	15.40	57.27
N17	P	P	P	F	P	P	P	P	P	5.74	79.94
N18	P	P	F	P	P	F	P	P	P	9.36	68.18
N19	P	P	F	F	P	P	P	P	P	7.92	71.36
N20	P	P	P	P	P	P	P	P	P	3.97	82.48
N21	P	F	P	F	F	F	F	P	P	25.16	51.20
N22	P	P	F	F	F	F	P	P	P	15.21	58.26
N23	P	P	F	P	P	P	P	P	P	4.61	79.60
N24	P	P	P	F	F	F	F	P	P	18.34	58.03
N25	P	P	F	F	P	P	P	P	P	6.48	72.81
N26	P	F	P	F	F	F	P	F	P	19.23	53.86
N27	P	P	F	P	P	P	P	P	P	10.82	66.72
N28	P	F	P	F	F	P	F	P	P	17.86	58.61
N29	P	P	F	P	P	P	P	P	P	8.62	70.67
N30	P	P	F	P	P	P	P	P	P	4.03	85.31
N31	P	P	F	P	P	P	P	P	P	5.27	82.83
Failure %	71			71			16			32	

P – pass; F – fail.

The four evaluation methods determined different failure rates. According to method-1, 71% failed to comply, the batches mostly failing at final time point.

When comparing dissolution profiles to the reference product based on method-2, most of the previously failed batches proved to show larger than ± 10% differences at 4 h and 12 h time points also, resulting in similar failure rates. The FDA ± 10% limit (method-2) proved to be quite restrictive, while the ± 25% limit (method-3) was found insufficiently discriminative. All failed batches according to method-3 showed different dissolution profiles, but certain method-4 rejects did not turn up in method-3, suggesting that the ± 25% is too large in the context of dissolution profile similarity.

The fit factor method is presumed to supplement the guideline recommendations in establishing appropriate dissolution specifications. In this case, setting limits of ± 17%, based on the minimum number of required time points (3: 4 h, 12 h and 24 h), is able to scan out all questionable batches.

Considering the resulted product characteristics and dissolution profile quality compliance results, it is noticeable that the process parameters and RM variability, simulated by interchanging different suppliers, influenced the performance of the hydrophilic matrix tablets. The dissolution variability determined that several experimental runs, when compared to reference product, presented dissimilar release profiles. In the CMC context, the established failure rates would imply time and resource consuming repercussions, in the attempt to ensure consistent quality (Figure 1).

To evaluate the effect of RM and process factor induced variability, DoE was applied.

Raw material variability impact assessment via DoE

Considering the high number of multi-level qualitative factors, the D-optimal design was an appropriate choice for the generation of experimental runs. The D-optimal is a fractioned design consisted of the best subset of experiments that span the largest volume in the experimental region. The experimental run selection is directed towards maximizing the determinant of the X'X matrix. 31 runs proved to be sufficient for the simultaneous evaluation of the 8 selected factors, through a linear model with screening objective. For data fitting purposes, PLS was chosen over multiple linear regression (MLR) due to its ability to deal with many responses simultaneously, suiting even designs with lower sphericity [9].

Considering the performance parameter values of a good model, mentioned under section 2.2.4, hardness (Y6) and release rate parameters (Y7 - Y16) lead to appropriate models ($p < 0.05$) and no lack of fit ($p > 0.05$). Responses such as LOD (Y5) and flow characteristics (Y1 - Y4) were poorly fitted. Moreover, Y3, Y4 and Y5 models were found to be not significant ($p > 0.05$) (Table IV). The poor modelling of these responses can be attributed to the small response

variability obtained between the experimental run formulations (Table II).

Table IV

Regression model evaluation

Response	Summary of fit				ANOVA	
	R2	Q2	Model Validity	Reproducibility	Model Significance	Lack of fit
Y1 - flow time	0.39	0.21	0.335	0.945	0.01	0.07
Y2 - angle of repose	0.47	0.33	0.635	0.825	0.002	0.233
Y3 - Hausner ratio	0.18	0.06	0.195	0.958	**0.268**	0.04
Y4 - Carr index	0.17	0.05	0.23	0.951	**0.299**	0.046
Y5 - loss on drying	0.27	0.1	0.245	0.954	**0.076**	0.049
Y6 - hardness	0.89	0.82	0.536	0.976	2.77E-11	0.157
Y7 - % released after 1 h	0.8	0.63	0.354	0.978	8.74E-07	0.076
Y8 - % released after 2 h	0.77	0.66	0.53	0.951	2.41E-07	0.153
Y9 - % released after 3 h	0.75	0.64	0.506	0.951	8.08E-07	0.139
Y10 - released after 4 h	0.72	0.59	0.624	0.908	3.60E-06	0.223
Y11 - released after 6 h	0.67	0.51	0.709	0.841	2.17E-05	0.313
Y12 - released after 8 h	0.76	0.47	0.756	0.818	2.12E-04	0.378
Y13 - % released after 10 h	0.8	0.53	0.821	0.78	5.38E-05	0.49
Y14 - % released after 12 h	0.82	0.54	0.714	0.885	2.03E-05	0.32
Y15 - % released after 18 h	0.81	0.53	0.698	0.891	2.50E-05	0.299
Y16 - % released after 24 h	0.75	0.42	0.663	0.876	3.58E-04	0.261

For the model interpretation, the corresponding coefficient plots were generated enhancing the identification of significant model terms, together with an estimation of the direction and magnitude of impact. In this graphical representation a coefficient is classified as significant if its 95% confidence interval (error bar) does not extend through y = 0. The calculated confidence interval gives an estimation of experimental noise reflecting the coefficient uncertainty.

As expected, direct correlation was determined between applied compression force and hardness. In addition, hardness was also influenced by a sort of Mg-St (S.F), factor that also showed a significant interaction with compression force (Figure 3). Formulations with Mg-St S.F were more sensitive to changes in compression force. Applying a low compression force yielded tablets with similar hardness despite the type of Mg-St supplier, whereas a high compression force highlighted differences in tablet hardness. This effect could be attributed to lubrication performance differences in report to the blending process that in return may affect the compactibility/compressibility of powder blends [37].

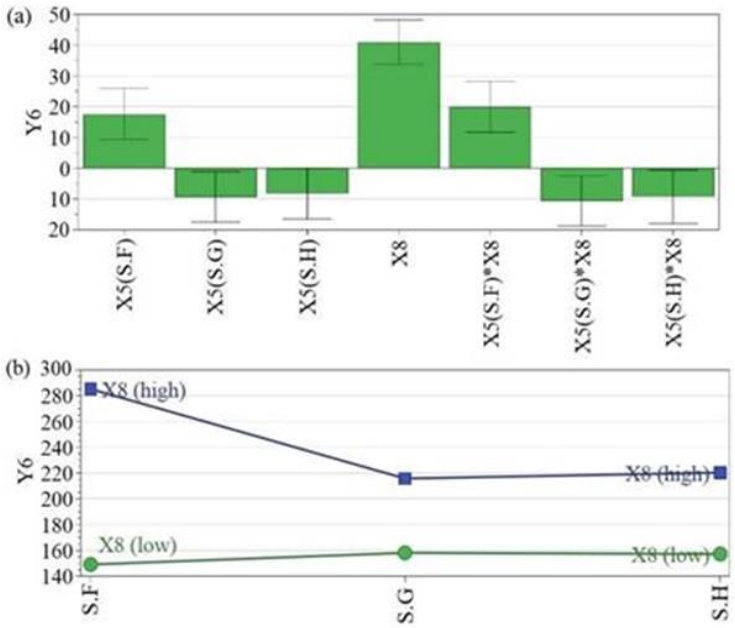

Figure 3.

a – Coefficient plot for hardness (Y6); b – X5*X8 interaction plot
X5 – magnesium stearate supplier; X8 – compression force

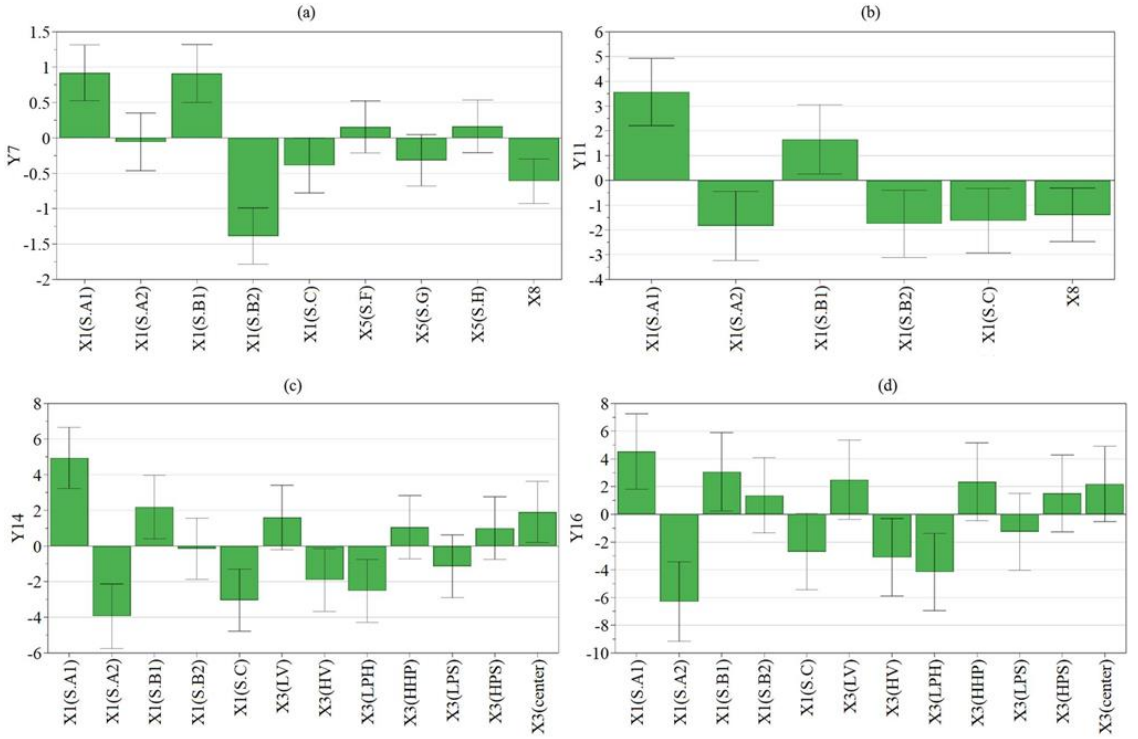

Figure 4.
Coefficient plots for dissolution data
a – 1, 2, 3 and 4 h; b – 6 h; c - 8, 10, 12 and 18 h; d – 24 h; X1 – indapamide supplier; X3 – hydroxypropyl methylcellulose K15M batch; X5 – magnesium stearate supplier; X8 – compression force; LV – low viscosity; HV – high viscosity; LHP – low hydroxypropoxyl content; HHP – high hydroxypropoxyl content; LPS – low particle size; HPS – high particle size; S. – supplier

The significant factors impacting dissolution were the API supplier; HPMC batch and compression force (Figure 4). Considering similar factor-effect patterns for different dissolution time points, the following coefficient plots can be used to interpret multiple responses: the coefficient plot of Y7 (Figure 4a) – 1, 2, 3 and 4 h; coefficient plot of Y11 (Figure 4b) – 6 h; coefficient plot of Y12 (Figure 4c) – 8, 10, 12 and 18 h; coefficient plot of Y16 (Figure 4d) – 24 h.

The most significant effect was attributed to the API variability, the change impacting all ten dissolution time points. Not only inter-supplier, but also inter-batch variability was detected. The supplier B batches predictably exerted a positive (S.B1 micro) and negative (S.B2 macro) effect, in accordance to their size, dissolution being directly correlated with the particle surface area [28]. S.B1 had a positive effect along all the dissolution process, starting off with the same magnitude of impact as S.A1, but diminished from 6 h onwards. S.B2 acted in the opposite direction, delaying indapamide release in first four hours, but lost its significance from hour 8.

The S.A and S.C batches influenced dissolution in different manners. Batches S.A2 and S.C determined dissolution decreasing effects. The impact of S.A2 debuted from hour 6 and increased towards later time points, inhibiting to a great extent the dissolution process. Still, from the perspective of integral dissolution profile, the greatest influence was attributed to batch S.A1, which improved the dissolution status.

The effect of compression force and HPMC acted alternatively. While the first 6 h were governed by the API and compression force, from 8 h onwards API and HPMC limited the dissolution. From the HPMC QbD batches the HV, LHP and centre proved to impact the process, presumably changing the characteristics of the rate controlling gel layer. Compression force was modulated in the 150 - 300 N range to offer flexibility to the process. RM variability caused deviations along the initial sampling points can be overcome by adjusting the compression force in the appropriate direction, within the previously mentioned range.

The lactose, PVP variability proved to have no/low significance on quality outcomes, information of aid in assuring flexibility to the process, not limiting manufacturing to a certain type/supplier of excipient. It can be presumed that the evaluated sorts can be interchanged with no impact on dissolution; however, the best approach would be to establish a multivariate domain of flexibility based on the preliminary quality control results.

The blending method and addition time of Mg-St also proved to be insignificant; however, these results are conclusive for the lab-scale. These observations should be re-evaluated also during the scale-up.

Based on the established PLS models it is possible to make predictions regarding the optimal material and process combinations that needed to obtain the desired quality profile. However, this approach is limited to the respective batches of API/excipient. In order to control and overcome variability of new batches, the investigated qualitative factors should be redefined in terms of physicochemical descriptors, also known as CMA.

Extended Root cause analysis via O2PLS

It is known that when dealing with an extended release, hydrophilic matrix formulations with poorly soluble API, the release kinetics are governed by the rate-controlling polymer and the API itself [41]. However, based on the previously presented findings, it is not sufficient to limit production to a certain sort or supplier of API/excipient.

The Noyes-Whitney principle expresses that the solubility rate is directly correlated with the specific surface area of drug particle in contact with dissolution media, the diffusion coefficient of API, the difference between the saturation solubility of the drug and concentration in the media after time t and inversely proportional to the thickness of the layer of diffusion [29]. In the present case, the HPMC hydrates, swells and forms a hydrogel layer that controls directly the diffusion and indirectly the dissolution of indapamide. The system is not purely diffusional, concurrent erosion arises after polymer chain disentanglement [39]. Chemically HPMC is a semi-synthetic polymer, with methoxyl and hydroxypropoxyl substituents attached to the cellulosic backbone. Although, kept between Ph. Eur. and more restrictive supplier specification limits it is essential to define the CMAs simultaneous effect in relation to the quality outcomes. Dow's QbD samples are based on viscosity, %HP and particle size. Viscosity is a reflection of molecular weight and molecular weight distribution, while the substitution affects the hydrophobic/hydrophilic status of the material. The %HP attributes hydrophilicity, improving hydration/gel formation, while viscosity influences the gel strength in relation to diffusion [1, 2, 4, 48]. The effect of particle size is debatable, but some studies suggest that it may affect the gel quality [19].

With regards to poorly soluble APIs, like indapamide, particle size effect on solubility is acknowledged [21, 35]. The smaller particle size, the higher the surface area is and the faster the dissolution process. Dissolution is also modulated through crystallinity. Amorphous materials possess higher internal energy and specific volume, with improved thermodynamic (solubility, motility) properties. Among the crystalline group, crystal structure differences (polymorphs) can also determine different physicochemical characteristics that further propagate to deviations in bioavailability and stability [5, 31]. For indapamide, polymorphs and pseudo-polymorphic forms have been identified [17]. Based on thermodynamic stability equilibriums, in time, amorphous materials can convert to crystalline, while polymorphs can stabilize in lower energy crystal structures, so commercial batch API solid forms should be screened and controlled [29].

Therefore, investigation was extended by employing thermal and spectroscopic methods for the solid-state characterization of indapamide, whereas for HPMC the quality descriptors were taken from the CoA supplied by the vendor (Table V).

Table V

Quantitative physicochemical descriptors for indapamide and HPMC

Indapamide (IND)								**Hydroxypropyl methylcellulose (HPMC)**			
Batch	D10 (µm)	D50 (µm)	D90 (µm)	Size Span	MNE (J/g)	Onset T (°C)	Peak T (°C)	Batch	Viscosity (mPa * s)	%HP	Size (%)
S. A1	1.4	3.04	6.36	1.632	-71.38	158	163	LV	13413	9.6	55
S. A2	6.82	21.1	54	2.236	-69.53	161.9	169.1	HV	24755	9.1	55.8
S. B1	1.7	4.6	10.6	1.935	-66.61	156.7	162.8	LPS	20156	9.4	52.6
S. B2	21.3	52.2	109	1.68	-70.04	163.9	171.2	HPS	17380	9.5	64.2
S. C	8.2	28.5	64.5	1.975	-68.39	161.3	169.3	LHP	16833	8.4	56.2
								HHP	16698	10.5	56.2
								centre	19036	9.4	57.5

*Particle size for HPMC is expressed in % to pass through a 230 US standard sieve; %HP – hydroxypropoxyl substitution percentage; MNE – mean normalized enthalpy; T – temperature

Laser diffraction particle size results indicated that all batches of indapamide could be classified as very fine (D50 ≤ 125 µm) according to Ph. Eur. 9.0 monograph 2.9.35 Powder fineness. Volume based distribution values Dx (D10, D50 and D90) were taken into consideration, expressing the dimensions under which x *per* cent of the cumulative undersize are situated. The classification in terms of particle size, was the following: S.A1 < S.B1 < S.A2 < S.C < S.B2. Particle size span, representing the distribution width, was also reported, comparable results being obtained. Batch S.A2 presented the widest range.

Indapamide batches were also checked for polymorphism and crystallinity. Based on the XRPD diffractograms (Figure 5a) and Raman spectra (Figure 6) no inter-batch variability could be detected.

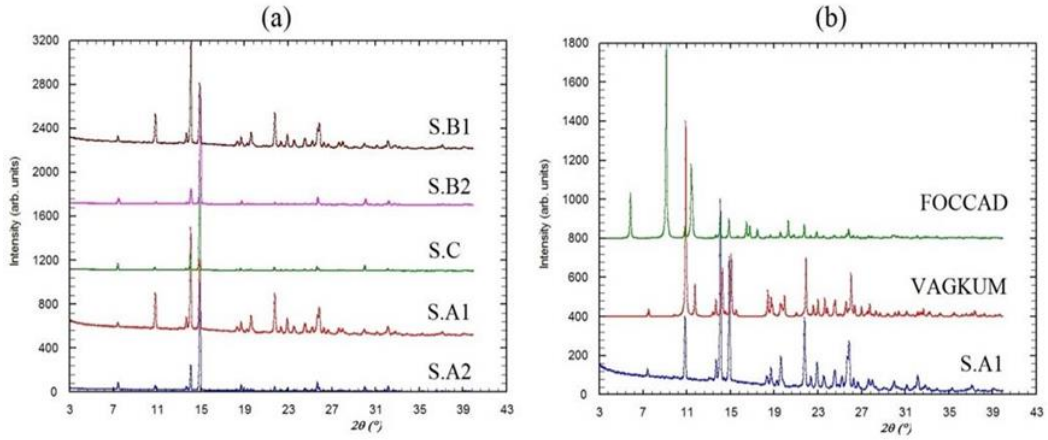

Figure 5.
X-ray powder diffraction patterns of the indapamide batches
a – inter-batch comparison; b – comparison to the indexed crystal structures from the CSD database

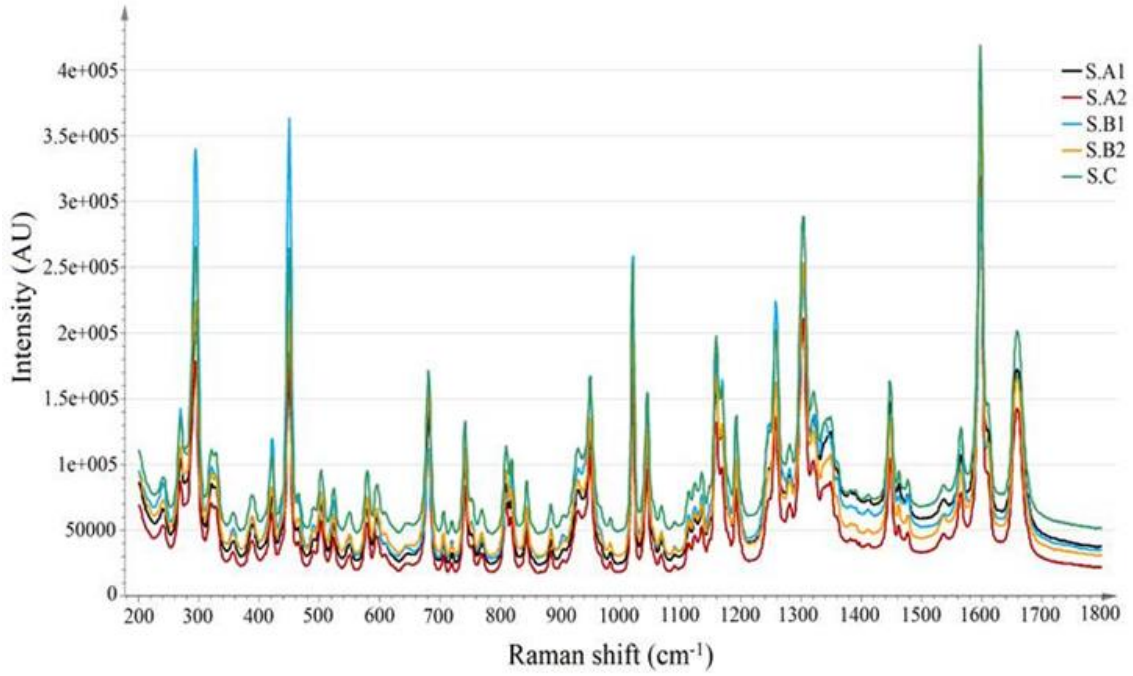

Figure 6.
Raman spectra of the indapamide batches

The XRPD diffraction patterns for the evaluated batches were similar, characterized by the same main peaks, the peak intensity differences were presumably caused by the particle size variability and/or preferential orientation [52]. With no evidence of broad, amorphous "halo" peaks it was suggested that all batches were predominantly crystalline. When compared against the existing crystal structures from the CSD database, codified as FOCCAD and VAGKUM, differences could be noted (Figure 5b).

Melt behaviour parameters (Table V) withhold information related to polymorphism, crystallinity and purity [31, 42], but were mostly taken into consideration based on their quantifiable nature. The onset and peak temperatures represent the start and end of melt, while the MNE gives an indication related to the energy of the phenomena. Compared to the cited melt temperature (Tm) of indapamide (160 - 162°C [43]), slight shifts were noted in the parameters of the evaluated batches. The significance of the thermal data was evaluated in the following section.

The quantitative physicochemical descriptors of indapamide and HPMC (Table V) along with compression force (the three critical parameters accountable for dissolution variability) were processed through MVDA tools. O2PLS method was employed to redefine results by means of CMAs. The method represents an extension of PLS, integrating notions of OPLS (orthogonal PLS) and PCA (principal component analysis). Rather than a sole regression, O2PLS provides an overview of

the global process, being applied for multi-block data situations [8].

For a two-block X/Y dataset O2PLS integrates data in joint and unique variabilities. In the present case, X represented the physicochemical descriptors (Table V) along with compression force and Y the dissolution data along the 10 time points.

Fitting the data generated a 2+0+1 O2PLS model (Table VI), with 2 predictive components accounting for joint XY variability and 1 orthogonal component for the Y block. Considering the R2 of 0.653 and Q2 of 0.505, a good model was obtained.

Table VI

Overview for O2PLS 2+0+1 model

Component	R2X	R2X (cum)	R2	R2 (cum)	Q2	Q2 (cum)	R2Y	R2Y (cum)
Model		0.563		0.653		0.505		0.995
Predictive		0.563		0.653		0.505		0.977
P1	0.405	0.46	0.614	0.613	0.372	0.372	0.907	0.907
P2	0.158	0.563	0.0387	0.653	0.133	0.505	0.0702	0.977
Orthogonal in Y (PCA)								0.0175
O1							0.0175	0.0175

Table VI shows that joint variation prevail over unique information. 56.3% of the variation in the CMA+CPP block was responsible for 97.7% of the variability found in the dissolution data, while the orthogonal in Y component captured only 1.75% as unique variability. The high percentage of explained dissolution variability confirmed the quality of experimental work and the absence of major systematic errors.

The considerable information overlap between the two data blocks was captured by two predictive components. The first predictive component was accountable for the majority of the dissolution variability (90.7%), and could be explained by variability in API particle size volume-based distributions (Dv10, Dv50 and Dv90) and melt temperature characteristics (Onset, Peak) (Figure 7a). Indapamide batches that presented a reduced particle size and low melt temperatures (Figure 7a) yielded a higher amount of dissolved active ingredient, characteristic especially for the later time points (Figure 7b).

The remaining predictive dissolution variability of 7.02% (second component), given by indapamide particle size span, MNE and HPMC viscosity (Figure 7c), manifested effect especially at the last two sampling points (Figure 7d).

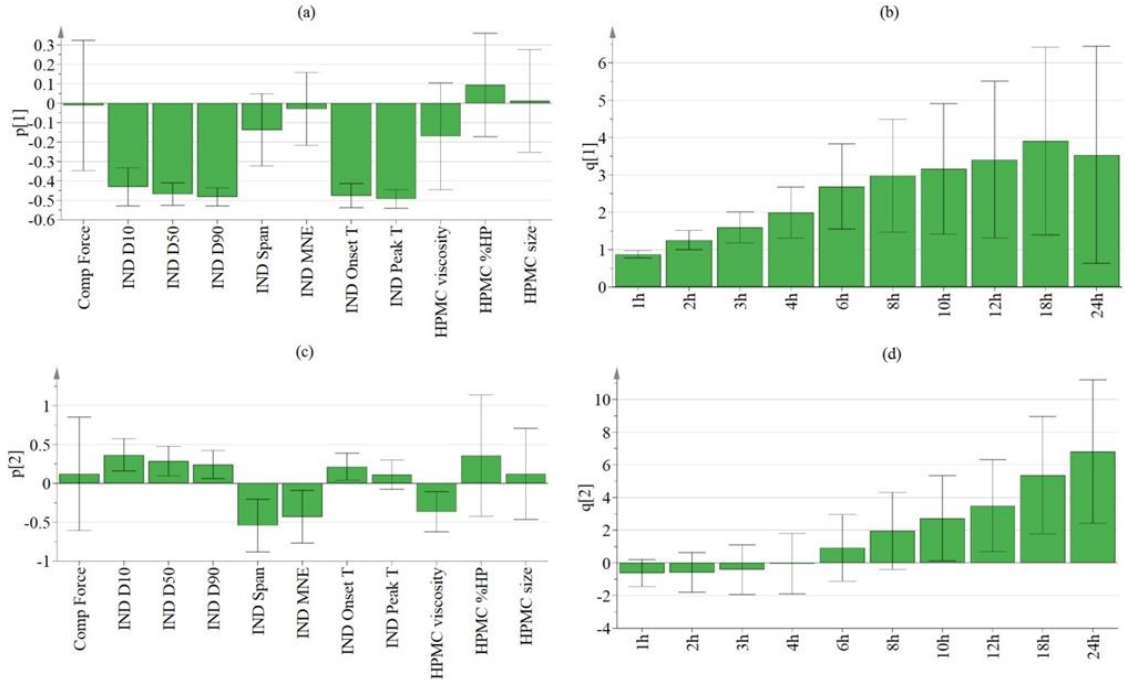

Figure 7.

a – Descriptor loadings for the first predictive component (p1); b – Dissolution loadings for the first predictive component (q1); c – Descriptor loadings for the second predictive component (p2); d – Dissolution loadings for the second predictive component (q2)

Evaluating the variable importance on projection (VIP) parameter for the present model, the impacting formulation/process factors were ranked in the following order: IND Peak T > IND D90 > IND D50 > IND Onset T > IND D10 > IND Span > HPMC viscosity > IND MNE, the others proving to be insignificant. Although according to DoE results, compression force was a significant factor for the first sampling points, O2PLS results suggested otherwise. The reason for this discrepancy in factor influence is given by the fact that in DoE data analysis, each sampling point was modelled separately, whereas for O2PLS the entire dissolution profile represented one response, enhancing its overview potential. Due to the considerably smaller differences in released indapamide at initial sampling points, where compression force manifested an effect, compared to later time points, the factor appears as not being influential. However, DoE generated models are still applicable for the modulation of initial segment of active ingredient dissolution, enabling adjustments in factor settings to compensate for raw material variability. The purpose of O2PLS was resumed to define a region of factor (physicochemical descriptor) combination in multivariate space, which would ensure a low risk of failure and a consistent quality profile.

The importance of the thermal data and particle size characteristics was recognized and reconfirmed throughout the data analysis workflow. Besides the particle size volume-based distribution span is also important, as it defines the relationship between them.

To define the CMA combinations that lead to a low risk of failure the score plot of the two predictive components was generated along with the corresponding loading plot for interpretation purposes (Figure 8).

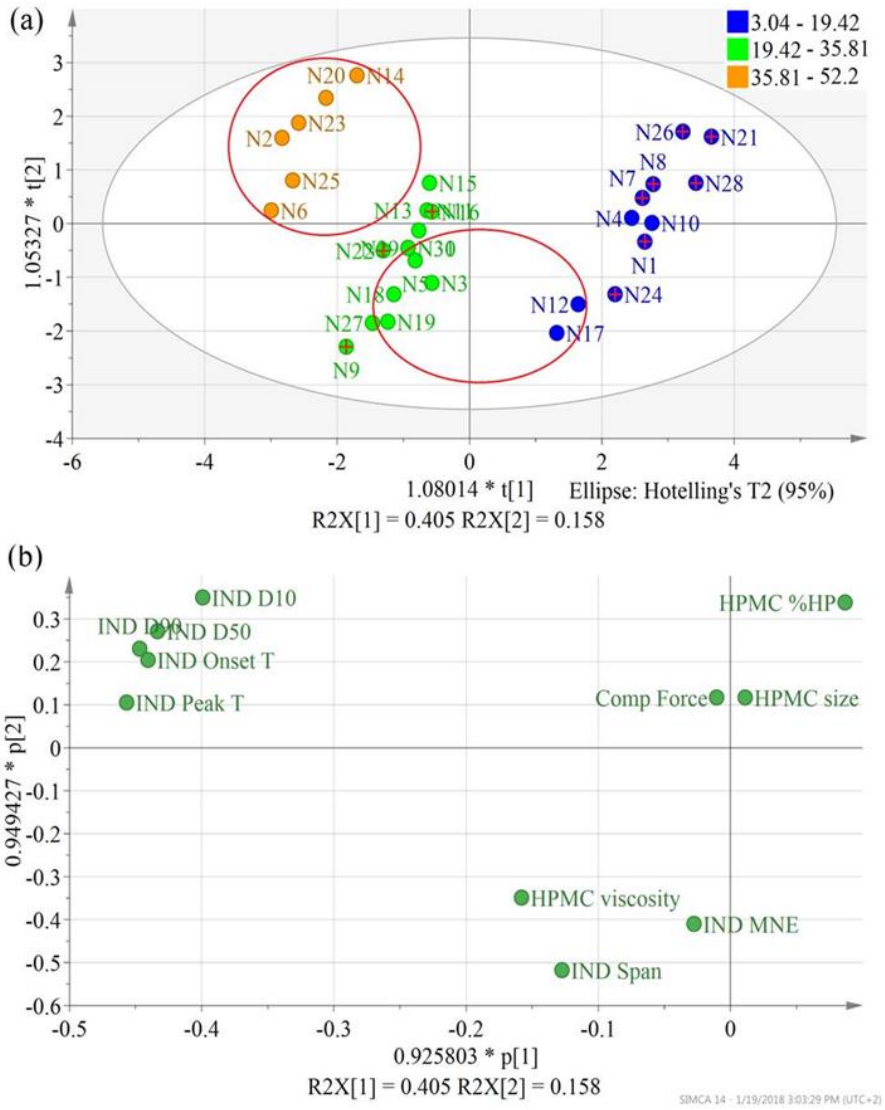

Figure 8.

a – Score scatter plot of first (t1) *versus* second predictive component (t2) with colour coding based on indapamide D50 value; b – Loading scatter plot (p1) *vs*. (p2); Observations that pose dissolution similarity issues according to f1 criteria are marked with a red cross; low failure risk regions are encircled

In the colour-coded region of the t1/t2 scatter plot three clusters could be identified in the direction of the first predictive vector, reflecting different indapamide particle size classes (Figure 8a). As suggested by the loading plot (Figure 8b), observations with higher particle size are found in the upper left quadrant (orange cluster), and the decrease in particle size occurs towards the right quadrant (blue cluster). The disposition of observations according to the second predictive component was determined by differences in indapamide size span, MNE and polymer viscosity.

Considering the disposition of failed experimental runs (f1 criteria) two regions with reduced risk of failure were highlighted. In order to ensure similar dissolution profiles with the reference product the indapamide particle size characteristics and thermal behaviour are of main interest considering that the first predictive component was responsible for approx. 90% of dissolution variability. The first confidence region is represented by the delimited area that includes the orange cluster, characterized by a D50 value between 35.81 µm and 52.2 µm. Experimental runs containing indapamide with the following characteristics (S.B2 batches) presented similar dissolution profiles with the reference product.

In return the blue cluster, characterized by low particle size contained mostly failing batches (S.A1, S.B1), resulting in profiles that differed to a high extent from the reference product ($f1 > 15$). Lower particle size based experimental runs failed due to the faster dissolution of the active ingredient.

However, if indapamide suppliers with lower particle size and higher span are used, the viscosity of HPMC is the decisional factor that can ensure the desired release profile. To ensure similar dissolution profiles in this second confidence region with smaller particle size, the release of active ingredient has to be hampered by increasing the viscosity of the polymeric gel, impacting the diffusion phenomenon. Although the second predictive component explains only 7.02% dissolution variability and interpretations should be made with caution, the presence of a confidence area offers the possibility of adjusting HPMC sort dependent on the API batch.

Based on these findings, particle size measurements and/or DSC tests are of key interest in ensuring a consistent quality profile for the extended release indapamide tablets. The criticality of particle size has been highlighted by several different authors also [13, 15, 51]. Moreover, the score plot can be used as a multivariate control method that could predict the risk of failure associated with future formulations that differ in terms of active ingredient or HPMC sort. For a low risk of failure the physicochemical descriptors of future formulations should generate projections within the delimited confidence regions. The procedure of pattern recognition on datasets has also been used previously to differentiate raw material clusters based on their flowing properties [3].

The need of applying data analytics in pharmaceutical development, establishing multivariate specifications for the raw material has already been signalled before [7, 32], but due to the lack of specific case studies it's benefits are yet to be recognized within the professional community.

According to ICH Q6A guideline [26], specifications are chosen to confirm the quality of the drug substance and drug product rather than to establish full characterization. The risk-based control strategy agrees with this quotation, however, in R&D full characterization is recommended to fundament CMA based multivariate raw material specifications and to determine the process flexibility potential.

Conclusions

On a micro scale, the present study proved that small changes within the physicochemical characteristics of the input raw material can compromise the quality of the final product. The degree of impact was significant. According to EMA and FDA guidelines, dissolution recommendations of the most evaluated batches presented out of specification (OOS) potential, which in the CMC environment would require time and resource consuming actions and investigations.

On a macro scale, the study highlighted the need to integrate the QbD based approach within R&D. Inter-supplier and inter-batch determined outcome variability, proved that production should not be limited to specific suppliers, but rather to predefined CMA intervals that are set to assure quality. Also, the common approach related to RM specification establishment should be redefined. A risk-based, multivariate analysis assisted control strategy of the incoming raw material is set to give reliability to the manufacturing process.

Acknowledgement

The authors would like to acknowledge the support of Richter Gedeon (Târgu-Mureș, Romania), Magista C&C (Constanța, Romania) and Colorcon Limited (Budapest, Hungary) with Dow Chemical Company (Midland, USA) in providing excipient samples for evaluation. The authors would also like to thank the implication in the solid-state characterization of indapamide the Rompharm Company (Bucharest, Romania), the National Institute for Research and Development of Isotopic and Molecular Technologies and the MedFuture Research Centre for Advanced Medicine (Cluj-Napoca, Romania). This project was supported by the Collegium Talentum 2019 Programme of Hungary and by the "Iuliu Hațieganu" University of Medicine and Pharmacy, Cluj-Napoca, Romania, internal doctoral research grant 3066/26/01.02.2018.

Conflict of interest

The authors declare no conflict of interest.

References

1. Alecu C, Tomuţă I, Dudas D, Leucuţa S, Mirciou C, Optimization of metoprolol tartrate modified release matrix tablets formulation using surelease as binder for granulation. *Farmacia*, 2013; 61(6): 1117-1130.
2. Antonoaea P, Bîrsan M, Todoran N, Rédai E, Vlad RA, Muntean DL, Rusu A, Ciurba A, Comparative analysis of the release performance of indomethacin from transdermal therapeutic systems. *Farmacia*, 2020; 68(6): 1029-1035.
3. Benedetti A, Khoo J, Sharma S, Facco P, Barolo M, Zomer S, Data analytics on raw material properties to accelerate pharmaceutical drug development. *Int J Pharm.*, 2019; 536: 122-134.
4. Caccavo D, Lamberti G, Barba AA, Abrahmsén-Alami S, Viridén A, Larsson A, Effects of HPMC substituent pattern on water up-take, polymer and drug release: An experimental and modelling study. *Int J Pharm.*, 2017; 528(1-2): 705-713.
5. Calvo NL, Maggio RM, Kaufman TS, Chemometrics-assisted solid-state characterization of pharmaceutically relevant materials. Polymorphic substances. *J Pharm Biomed Anal.*, 2018; 147: 518-534.
6. Cascone S, Modeling and comparison of release profiles: Effect of the dissolution method. *Eur J Pharm Sci.*, 2017; 106: 352-361.
7. Duchesne C, MacGregor JF, Establishing Multivariate Specification Regions for Incoming Materials. *J Qual Technol.*, 2004; 36(1): 78-94.
8. Eriksson L, Byrne T, Johansson E, Trygg J, Vikström C, Multi- and Megavariate Data Analysis Basic Principles and Applications. MKS Umetrics AB: Malmö, Sweden, 2013.
9. Eriksson L, Johansson E, Kettaneh-Wold N, Wikström C, Wold S, Design of experiments: principles and applications. Umetrics Academy: Umeå, Sweden, 2008.
10. European Medicines Agency, Report from the EMA-FDA QbD pilot program (EMA/213746/2017). 2017, www.ema.europa.eu/docs/en_GB/document_library/Other/2017/04/WC500225533.pdf.
11. European Medicines Agency, Guideline on quality of oral modified release products (EMA/CHMP/QWP/428693/2013). 2014, www.ema.europa.eu/docs/en_GB/document_library/Scientific_guideline/2014/07/WC500170465.pdf.
12. Fisher AC, Lee SL, Harris DP, Buhse L, Kozlowski S, Yu L, Kopcha M, Advancing pharmaceutical quality: An overview of science and research in the U.S. FDA's Office of Pharmaceutical Quality. *Int J Pharm.*, 2016; 515(1-2): 390-402.
13. Fonteyne M, Wickström H, Peeters E, Vercruysse J, Ehlers H, Peters BH, Remon JP, Vervaet C, Ketolainen J, Sandler N, Rantanen J, Naelapää K, De Beer T, Influence of raw material properties upon critical quality attributes of continuously produced granules and tablets. *Eur J Pharm Biopharm.*, 2014; 87(2): 252-263.
14. Food and Drug Administration, Guidance for Industry Guidance for Industry Extended Release Oral Dosage Forms: Development, Evaluation, and Application of *In Vitro*/*In Vivo* Correlations. 1997, www.fda.gov/downloads/drugs/guidances/ucm070239.pdf.
15. Gavan A, Iurian S, Casian T, Porfire A, Porav S, Voina I, Oprea A, Tomuță I, Fluidised bed granulation of two APIs: QbD approach and development of a NIR in-line monitoring method. *Asian J Pharm Sci.*, 2020; 15(4): 506-517.
16. Gavan A, Sylvester B, Casian T, Tomuță I, In-Line Fluid Bed Granulation Monitoring By Nir Spectroscopy. Method Development and Validation. *Farmacia*, 2019; 67(2): 248-257.
17. Ghugare P, Dongre V, Karmuse P, Rana R, Singh D, Kumar A, Filmwala Z, Solid state investigation and characterization of the polymorphic and pseudo-polymorphic forms of indapamide. *J Pharm Biomed Anal.*, 2010; 51(3): 532-540.
18. Hales D, Vlase L, Porav SA, Bodoki A, Barbu-Tudoran L, Achim M, Tomuță I, A quality by design (QbD) study on enoxaparin sodium loaded polymeric microspheres for colon-specific delivery. *Eur J Pharm.*, 2017; 100: 249-261.
19. Heng PWS, Chan LW, Easterbrook MG, Li X, Investigation of the influence of mean HPMC particle size and number of polymer particles on the release of aspirin from swellable hydrophilic matrix tablets. *J Control Release.*, 2001; 76(1-2): 39-49.
20. Hertrampf A, Müller H, Menezes JC, Herdling T, Advanced qualification of pharmaceutical excipient suppliers by multiple analytics and multivariate analysis combined. *Int J Pharm.*, 2015; 495(1): 447-458.
21. Hintz RJ, Johnson KC, The effect of particle size distribution on dissolution rate and oral absorption. *Int J Pharm.*, 1989; 51(1): 9-17.
22. ICH Expert Working Group, ICH Harmonised Tripartite Guideline - Pharmaceutical Development, Q8(R2). 2009, https://database.ich.org/sites/default/files/Q8%28R2%29 Guideline.pdf.
23. ICH Expert Working Group, ICH Harmonised Tripartite Guideline - Quality Risk Management, Q9. 2005, www.ich.org/fileadmin/Public_Web_Site/ICH_Products/Guidelines/Quality/Q9/Step4/Q9_Guideline.pdf.
24. ICH Expert Working Group, ICH Harmonised Tripartite Guideline - Pharmaceutical Quality System, Q10. 2008, www.ich.org/fileadmin/Public_Web_Site/ICH_Products/Guidelines/Quality/Q10/Step4/Q10_Guideline.pdf.
25. ICH Expert Working Group, ICH Harmonised Tripartite Guideline - Validation of Analytical Procedures: Text and Methodology, ICH Q2(R1). 2005, www.ich.org/fileadmin/Public_Web_Site/ICH_Products/Guidelines/Quality/Q2_R1/Step4/Q2_R1__Guideline.pdf.
26. ICH Expert Working Group, ICH Harmonised Tripartite Guideline - Specifications: Test procedures and acceptance criteria for new drug substances and new drug products: chemical substances, ICH Q6A. 1999, www.ich.org/fileadmin/Public_Web_Site/ICH_Products/Guidelines/Quality/Q6A/Step4/Q6Astep4.pdf.
27. Ilyés K, Balogh A, Casian T, Igricz T, Borbás E, Démuth B, Vass P, Menyhárt L, Kovács NK, Marosi G, Tomuță I, Nagy ZK, 3D floating tablets: Appropriate 3D design from the perspective of different *in vitro* dissolution testing methodologies. *Int J Pharm.*, 2019; 567: 118433: 1-11.

28. Jambhekar SS, Breen PJ, Drug dissolution: significance of physicochemical properties and physiological conditions. *Drug Discov Today*, 2013; 18(23-24): 1173-1184.
29. Kanaujia P, Poovizhi P, Ng WK, Tan RBH, Amorphous formulations for dissolution and bioavailability enhancement of poorly soluble APIs. *Powder Technol.*, 2015; 285: 2-15.
30. Kim S, Thiessen PA, Bolton EE, Chen J, Fu G, Gindulyte A, Han L, He J, He S, Shoemaker BA, Wang J, Yu B, Zhang J, PubChem Substance and Compound databases. *Nucleic Acids Res.*, 2016; 44(D1): D1202-1213.
31. Law D, Zhou D, Solid-State Characterization and Techniques. In: Developing Solid Oral Dosage Forms: Pharmaceutical Theory and Practice, 2nd Edition. Elsevier, 2017; 59-84.
32. MacGregor JF, Liu Z, Bruwer MJ, Polsky B, Visscher G, Setting simultaneous specifications on multiple raw materials to ensure product quality and minimize risk. *Chemom Intell Lab Syst.*, 2016; 157: 96-103.
33. Malvern Instruments Ltd, Laser Diffraction: Particle size distribution from nanometers to millimeters. 2017, www.malvernpanalytical.com/en/products/technology/light-scattering/laser-diffraction.
34. Matji A, Carvajal L, Conde F, Peña MA, Donato N, Serrano DR, Torrado JJ, Effect of the characteristics of raw material ibuprofen on roller compaction and dissolution. *J Drug Deliv Sci Technol.*, 2017; 42: 237-244.
35. Mosharraf M, Nyström C, The effect of particle size and shape on the surface specific dissolution rate of microsized practically insoluble drugs. *Int J Pharm.*, 1995; 122(1-2): 35-47.
36. Muntean AC, Negoi OI, Rus LL, Vonica AL, Tomuță I, Formulation of orodispersible tablets containing paracetamol and their *in vitro* characterization - a QbD approach. *Farmacia*, 2020; 68(3): 436-446.
37. Nakagawa H, Kano M, Hasebe S, Suzuki T, Wakiyama N, Real-time monitoring of lubrication properties of magnesium stearate using NIR spectrometer and thermal effusivity sensor. *Int J Pharm.*, 2013; 441(1-2): 402-413.
38. Pallagi E, Ambrus R, Szabó-Révész P, Csóka I, Adaptation of the quality by design concept in early pharmaceutical development of an intranasal nanosized formulation. *Int J Pharm.*, 2015; 491(1-2): 384-392.
39. Pani NR, Nath LK, Development of controlled release tablet by optimizing HPMC: Consideration of theoretical release and RSM. *Carbohydr Polym.*, 2014; 104: 238-245.
40. Porfire A, Rus L, Vonica AL, Tomuță I, High-throughput NIR-chemometric methods for determination of drug content and pharmaceutical properties of indapamide powder blends for tabletting. *J Pharm Biomed Anal.*, 2012; 70: 301-309.
41. Qui Y, Rational Design of Oral Modified-Release Drug Delivery Systems. In: Developing Solid Oral Dosage Forms: Pharmaceutical Theory and Practice, 2nd Edition. Elsevier, 2017; 519-554.
42. Rus LM, Iurian S, Kacso I, Borodi G, Porav S, Hegheș SC, Iuga CA, Tomuță I, Development of meloxicam oral lyophilisates: role of thermal analysis and complementary techniques. *Farmacia*, 2019; 67(1): 56-67.
43. Rus LM, Tomuță I, Iuga C, Maier C, Kacso I, Borodi G, Bratu I, Bojiță M, Compatibility studies of Indapamide/pharmaceutical excipients used in tablet preformulation. *Farmacia*, 2012; 60(1): 92-101.
44. Sassard J, Bataillard A, McIntyre H, An overview of the pharmacology and clinical efficacy of indapamide sustained release. *Fundam Clin Pharmacol.*, 2005; 19(6): 637-645.
45. Silva BMA, Vicente S, Cunha S, Coelho JFJ, Silva C, Reis MS, Simões S, Retrospective Quality by Design (rQbD) applied to the optimization of orodispersible films. *Int J Pharm.*, 2017; 528(1-2): 655-663.
46. Sun D, Hu M, Browning M, Friedman RL, Jiang W, Zhao L, Wen H, Dissolution Failure of Solid Oral Drug Products in Field Alert Reports. *J Pharm Sci.*, 2017; 106(5): 1302-1309.
47. Takagi T, Ramachandran C, Bermejo M, Yamashita S, Yu LX, Amidon GL, A Provisional Biopharmaceutical Classification of the Top 200 Oral Drug Products in the United States, Great Britain, Spain, and Japan. *Mol Pharm.*, 2006; 3(6): 631-643.
48. Viridén A, Wittgren B, Larsson A, Investigation of critical polymer properties for polymer release and swelling of HPMC matrix tablets. *Eur J Pharm Sci.*, 2009; 36(2–3): 297-309.
49. Whelton PK, Committee W, Carey RM, Chair V, Aronow WS, Committee Member W, Casey DE, Collins KJ, Dennison Himmelfarb C, DePalma SM, Gidding S, Jamerson KA, Jones DW, MacLaughlin EJ, Muntner P, Ovbiagele B, Smith SC, Spencer CC, Stafford RS, Taler SJ, Thomas RJ, Williams KA, Williamson JD, Wright JT, 2017 ACC/AHA/AAPA/ABC/ACPM/AGS/APhA/ASH/ASPC/NMA/PCNA Guideline for the Prevention, Detection, Evaluation, and Management of High Blood Pressure in Adults. *J Am Coll Cardiol.*, 2017; 71(19): e127-e248.
50. Yu LX, Kopcha M, The future of pharmaceutical quality and the path to get there. *Int J Pharm.*, 2017; 528(1-2): 354-359.
51. Zhao C, Jin C, Gao H, Wang L, Liu H, He Z, Effect of raw material variability of glipizide on the *in vitro* dissolution rate and *in vivo* bioavailability performance: The importance of particle size. *Asian J Pharm Sci.*, 2019; 14(2): 165-173.
52. Zolotoyabko E, Basic concepts of X-ray diffraction. Wiley-VCH, 2014.