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A study of physicochemical and biopharmaceutical properties of Amoxicillin tablets using full factorial design and PCA biplot

Kerly F.M. Pasqualoto *, Reinaldo F. Teófilo, Marcos Guterres, Flávia S. Pereira, Márcia M.C. Ferreira

Laboratory for Theoretical and Applied Chemometrics, Department of Physical Chemistry, Institute of Chemistry, The State University of Campinas–UNICAMP, P.O. Box 6154, Campinas SP 13084-971, Brazil

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Abstract

The variables that influence the tablets obtained by direct compression method deserve to be studied to minimize formulations costs and to improve the physicochemical and biopharmaceutical properties of the compacts. Here, we explore the adjuvants effects on amoxicillin tablet formulations considering multiple responses, and indicate the most suitable formulation composition. A 2^3 full factorial design was built to different amoxicillin formulations, each one containing three replicate batches, and eight responses (physicochemical and biopharmaceutical parameters) were obtained. The microcrystalline cellulose (MCC) type Avicel® PH-102 (low) or PH-200 (high), the absence (low) or presence (high) of spray-dried lactose (LAC), and the absence (low) or presence (high) of disintegrant (DIS) were the levels investigated. The more relevant responses to the distinct formulations from the experimental design were hardness, friability, and the amount of amoxicillin dissolved during the first hour. PCA biplot indicated high values of amount of drug dissolved in 60 min as advantageous responses for the investigated amoxicillin tablet formulations and high values of friability as not desirable. Considering the individual response evaluation, the most suitable amoxicillin tablet formulation should present in its composition the MCC type Avicel® PH-102 (low level) and the superdisintegrant agent (DIS high level), croscarmellose sodium, but no LAC (low level).

Keywords: Factorial design; PCA biplot; Direct compression; Physicochemical and biopharmaceutical properties; Amoxicillin tablets

1. Introduction

Tablet manufacturing by direct compression (DC) has increased steadily over the years. It offers advantages over other manufacturing processes, such as wet granulation, and provides high efficiency [1]. The tablets are compressed directly from powder blends of the active ingredient and suitable excipients. Each excipient is selected to meet the needs of processibility and product use. The major types of excipients or adjuvants used are fillers or diluents, binders, disintegranting agents, and lubricants—which are present in nearly all tablet formulations [2].

DC is the most efficient process because it is fastest and simplest for the tablet manufacturing and protects the drug from heat and moisture [3]. Moreover, the tablet characteristics such as stability, dissolution, and bioavailability of the active drugs also may be improved using the DC method [4].

Although DC technique seems quite simple, the selection of appropriate excipients and their levels in the formulation is crucial for a successful tablet formulation. The DC fillers binders must fulfill certain requirements: good binding functionality and powder flow are essential; a well-designed particle size distribution provides favorable mixing conditions; compatibility with other excipients or drugs is also essential, as is the ability to carry high amounts of active ingredient [1,3,5]. Microcrystalline cellulose (MCC) and spray-dried lactose (LAC) are examples of DC fillers binders [2].

MCC (Avicel®) shows a relatively free flow, good compressibility, and high dilution potential, being also physiologically inert and nontoxic. Its filler-binder-disintegrant properties lead to easy and prompt tablet disintegration, allowing the drug dissolution [6–9].

* Corresponding author. Tel.: +55 19 3521 3102; fax: +55 19 3788 3023.
E-mail address: kerly@netpoint.com.br (K.F.M. Pasqualoto).

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LAC can be used in association with microcrystalline cellulose to reduce formulation costs [6,9]. According to specifications of the manufacturer, the disintegration time of lactose is dependent on formulations, and some of these formulations need the addition of a disintegrant agent [6].

Croscarmellose sodium is a superdisintegrant agent (DIS), which improves appreciably the disintegration time of tablet formulations. It presents an advantage in DC, since it can be used in low concentrations, providing suitable dissolution of the formulations [6,8,10].

The variables that influence the tablets obtained by DC method deserve to be studied to minimize formulations costs and to improve the physicochemical and biopharmaceutical properties of the resulting compacts.

In several process development and manufacturing applications, potentially influential variables are numerous. Screening reduces the number of variables by identifying the key variables (the "vital few") that affect product quality. Therefore, it may also suggest the "best" or optimal settings for these factors. An example of methodology extensively used in industry for screening is factorial designs [11–16]. Here, the focus is on the screening or process characterization [11,13–15].

Amoxicillin 500 mg tablets formulations were previously developed using DC technique [6]. MCC, LAC, and DIS were the experimental adjuvants, and the compacts were evaluated based on their physicochemical and biopharmaceutical properties, such as weight variability, thickness and diameter, hardness, friability, drug content (amoxicillin concentration), disintegration time, and dissolution profile.

In this study, a full factorial design is employed to explore the adjuvants (independent variables) effects amoxicillin 500 mg tablet formulations [6] considering those responses (the dependent variables) that are considered to be more relevant, as well as to identify the most suitable formulation composition. Additionally, a PCA biplot [17–20] was applied to explore and visualize the investigated tablet formulations (three replicate batches and its respective average for each tablet formulation) and the most relevant responses.

2. Experimental

A 2^3 full factorial design [11–15] was built to eight different amoxicillin formulations, each one containing three replicate batches, and eight responses were obtained [6]. Each independent variable was investigated at two levels as presented in Table 1.

Table 1
Independent variables and respective levels investigated

Variables	Levels	
	−	+
MCC type	Avicel® PH-102	Avicel® PH-200
LAC	Absence	Presence
DIS	Absence	PRESENCE

The resulting compacts from each formulation were evaluated based on the following physicochemical and biopharmaceutical parameters: weight variability, thickness and diameter, hardness, friability, drug content (iodometric assay), disintegration time, and dissolution profile (120 min). The detailed methodology was reported in Ref. [6]. All investigated formulations generated tablets presenting those parameters within the specified values, according to the official methodology. The responses of each formulation are presented in Table 2.

The dissolution profile data were considered in two different approaches: (a) the percentage of amoxicillin dissolved during 90 min (official methodology) and (b) the amount of amoxicillin dissolved at the first hour of experimentation (Hixson–Crowell law: $W_0^{1/3} - W^{1/3} = K(t - t_1)$ [21], where W_0 is the initial theoretical amount of drug in the tablets submitted to dissolution ($W_0 = 500$ mg), t_1 is the disintegration time (it was considered as 1 min), W is the amount of drug not dissolved in t, and K is the rate dissolution constant; t varied from 10 to 60 min) [6].

The thickness, diameter, and disintegration time properties barely changed for all investigated formulations (see Table 2). The weight variability measures had their values changing due to the DIS level, but they still remained within the accepted limits (691.60–764.40 mg for the formulations without DIS, and 698.25–771.75 mg for those containing DIS).

The iodometric assay was used to determine the drug content, which is related to the amoxicillin concentration in the tablet formulation observing the labeled drug amount. The values presented for that response (Table 2) did not change significantly regarding all formulations. The considered labeled drug amount was 500 mg (100%), and the accepted limits were 450–600 mg (90–120%).

According to the methodology used for the dissolution profiles [6], the amount of drug dissolved during a period of 90 min should not be less than 80% of the amount labeled (500 mg). All formulations presented much more than 80% of amoxicillin dissolved in 90 min (Table 2, dissolution), suggesting that the DC method improved the drug availability.

The amount of amoxicillin dissolved during the first hour of experimentation (dissolution) is related to the dissolution release rate, as already mentioned. That response can be used to verify if the manufacturing method (DC) is, or is not, improving the drug availability.

A suitable tablet formulation must have low friability to avoid dust as well as must be hard enough to prevent crushing, but flexible enough not to be easily broken. The friability measure is based on the loss of powder (weight loss) and it must be less than 1.5% for all amoxicillin tablet formulations. The tablet hardness indicates the force used to crush the compact, using a manual apparatus with an air pump and, according to the official methodology [6], the lowest value acceptable is 45 N. Friability and hardness are physical parameters that need to be considered for optimization process.

Regarding all comments above, the manufacturing method (DC), and the previously reported statistical analysis [6], the responses more pertinent to the distinct formulations from the experimental design were hardness, friability, and the amount of amoxicillin dissolved during the first hour of experimentation

Table 2
Physicochemical and biopharmaceutical properties of the resulting amoxicillin 500 mg tablets in the original design order

Weight variability (mg)	Thickness (cm)	Diameter (cm)	Hardness (N)	Friability (%)	$t_{disintegration}$ (min)	Iodometric assay (%)	Dissolution[a] 90 min (%)	Dissolution[b] ($W_0^{1/3} - W^{1/3}$) ($mg^{1/3}$)
730.13 ± 3.78	0.46 ± 0.01	1.36	117.8 ± 18.5	0.17 ± 0.03	<1	95.50 ± 3.05	98.23 ± 1.95	3.48 ± 0.87
730.72 ± 3.06	0.46 ± 0.01	1.36	106.7 ± 20.2	0.19 ± 0.03	<1	94.92 ± 0.64	99.17 ± 6.12	3.77 ± 0.91
724.23 ± 8.08	0.44 ± 0.00	1.36	118.3 ± 7.6	0.20 ± 0.05	<1	92.86 ± 2.79	100.83 ± 2.01	3.57 ± 1.04
730.51 ± 0.66	0.44 ± 0.00	1.36	110.2 ± 3.4	0.24 ± 0.04	<1	93.54 ± 2.29	96.90 ± 0.46	3.36 ± 0.86
732.03 ± 13.49	0.46 ± 0.01	1.36	115.2 ± 4.5	0.11 ± 0.01	<1	92.01 ± 0.54	101.80 ± 2.95	4.77 ± 1.55
737.86 ± 12.89	0.47 ± 0.01	1.36	100.5 ± 9.1	0.13 ± 0.03	<1	98.02 ± 1.37	95.93 ± 4.40	3.71 ± 0.87
740.95 ± 4.11	0.45 ± 0.01	1.36	103.0 ± 8.3	0.20 ± 0.09	<1	95.84 ± 1.70	92.83 ± 3.55	3.44 ± 0.72
741.50 ± 7.15	0.45 ± 0.00	1.36	85.3 ± 14.8	0.37 ± 0.05	<1	96.43 ± 1.76	89.20 ± 1.21	2.85 ± 0.54

$t_{disintegration}$ = disintegration time.
[a] Dissolution corresponds to the percent of amoxicillin dissolved during 90 min.
[b] Dissolution corresponds to the amount of amoxicillin dissolved at the first hour [Hixson–Crowell law: $W_0^{1/3} - W^{1/3} = K(t - t_1)$] [6,21].

(dissolution release rate), and a statistical evaluation considering each response was individually performed.

Moreover, an exploratory data analysis [principal component analysis (PCA)] [17,18] was carried out to visualize the investigated tablet formulations (three replicate batches and its respective average for each tablet formulation) and the most relevant responses. The results were displayed as a biplot graph.

The biplot diagram is commonly used for graphing row and column elements using a single display [19]. The method has been used to display objects and variables on the same graph in principal components analysis, row and column factors in correspondence analysis of two-way contingency tables, and to detect interaction in two-way analysis of variance tables [20]. Similarities between species or sites may be gleaned from these types of plots. Also it is common to interpret the axes in the biplot and treat the coordinates as scores on these axes.

3. Results and discussion

Regarding each response individually, the MCC and DIS levels were significant and negative for hardness, as presented in Table 3. The MCC type Avicel® PH-102 as well as the absence of DIS increased the hardness of the tablet formulations.

The lower values of hardness in formulations containing the MCC type Avicel® PH-200 could be explained by its larger mean particle size (200 μm). The larger mean particle size is responsible for providing a smaller superficial area and, consequently, the particle contact area for bonding also becomes smaller [9,22].

The larger mean particle size of Avicel® PH-200 would be impairing the interactions or bonds between particles of the same material [cohesion], such as MCC; or, between particles of different components on formulation [adhesion] during the compaction procedure, such as mixture of MCC, LAC, and DIS particles [6].

Moreover, formulations containing DIS plus two insoluble direct compression systems (diluent/drug) can present a decrease in breaking force (hardness), as observed by Khan and Rhodes (1973) [10].

The variables LAC and DIS presented an interaction for the friability response (see Table 3). The presence of both LAC and DIS increased the friability, as shown in Fig. 1.

LAC is included under the brittle materials category by the Wiederkehr–von Vincenz classification [9], which is based on the compaction properties of materials. That is, the lactose generally gives a lower mechanical strength and decreases the resistance of tablets to fragmentation [23].

The MCC type Avicel® PH-200 also contributed to increase the friability property (Table 3). Once again, the smaller superficial area of the MCC type Avicel® PH-200 would be impairing the interactions between the particles [cohesion and adhesion] due to the smaller contact area for bonding during the compaction step, producing tablets less resistant to crushing, and crumbling [6]. Doelker [9] emphasized the importance of the particle size in the interparticular bonds of diluents with the

Table 3
Results from the individual response evaluation

	Hardness		Friability		Amount of amoxicillin dissolved in 60 min	
	Coefficient	p	Coefficient	p	Coefficient	p
Mean	107.13*	0.000	0.20*	0.000	3.62*	0.000
MCC	−6.46*	0.021	0.03*	0.004	−0.20*	0.032
LAC	−2.92	0.263	0.05*	0.000	−0.31*	0.002
DIS	−6.13*	0.027	0.001	0.895	0.07	0.394
MCC × LAC	0.00	1.000	0.02	0.058	0.00	0.973
MCC × DIS	−1.63	0.527	0.02	0.081	−0.22*	0.020
LAC × DIS	−3.92	0.139	0.03*	0.006	−0.23*	0.014

* Significant coefficients with $\alpha = 0.05$ and 16 degree of freedom.

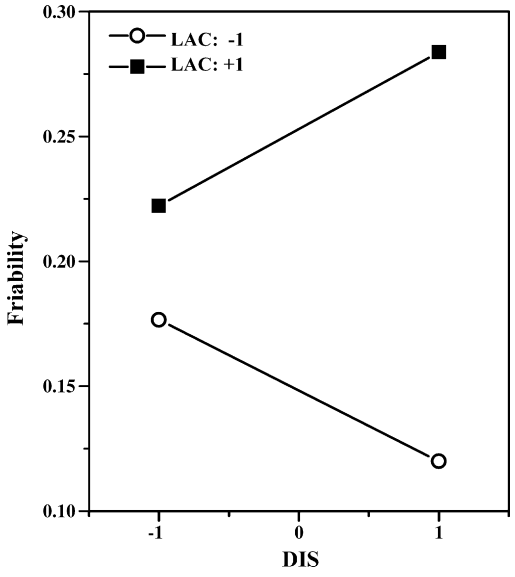

Fig. 1. Plot of marginal means analysis–interaction LAC × DIS for the friability response.

other components of the formulation, indicating that in some cases the dilution potential of Avicel® PH-200 may not be as good as that found using Avicel® PH-102.

The amount of drug dissolved response presented two interactions, one between the MCC and DIS levels, and another between the LAC and DIS levels (see Table 3). The combination of MCC low level (Avicel® PH-102), LAC low level (absence) and DIS high level (presence) simultaneously increases the amount of drug dissolved considering the plot of marginal means analysis (Fig. 2).

The improved performance of DIS in the formulations presenting Avicel® PH-102 could be explained due to the larger interparticular contact area for bonding of this MCC type, since it has a smaller mean particle size (100 μm) [9,22].

Regarding the biopharmaceutical property (amount of drug dissolved at the first hour—dissolution profile) (Table 3), the

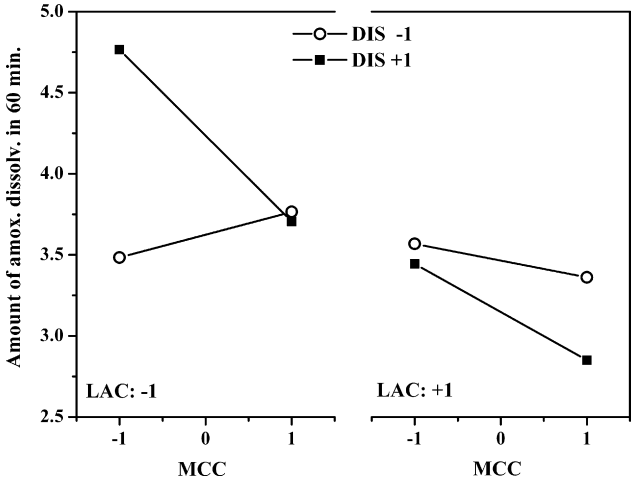

Fig. 2. Plot of marginal means analysis–interaction MCC × DIS and LAC × DIS for the amount of drug dissolved in 60 min.

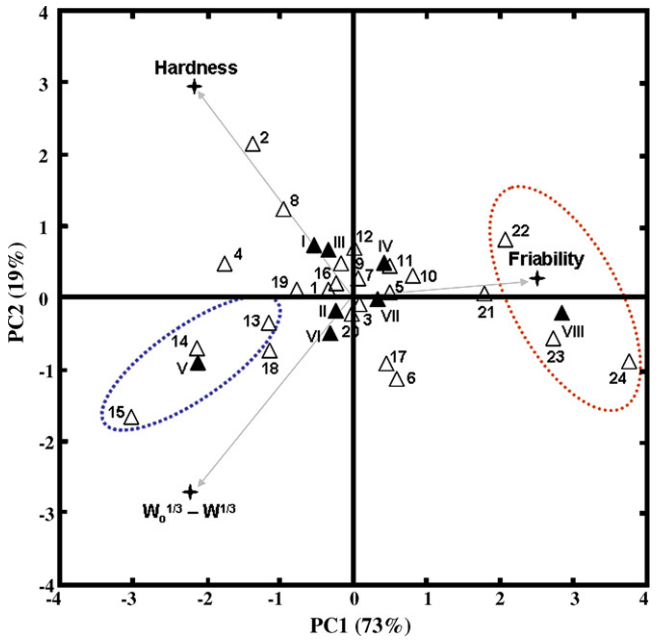

Fig. 3. The PCA biplot. The relevant responses are symbolized by stars and they are superimposed to the individuals display. The three replicate batches of each tablet formulation are represented as arabic numbers (white triangles) and their respective averages are indicated as roman numbers (black triangles).

individual response evaluation indicated as the most suitable amoxicillin tablet formulation that one presenting low level of MCC (Avicel® PH-102), low level of LAC (absence), and high level of DIS (presence of croscarmellose sodium) in its composition.

As already mentioned, a preliminary PCA [18,19] was employed to explore and visualize the data considering the most relevant responses (hardness, friability, and the amount of amoxicillin dissolved in 60 min) found for the investigated tablet formulations. A biplot display was used, and it is presented in Fig. 3.

PC1 or factor 1 describing 73 % of the original information is related to the LAC level. The averages III (batches 7–9), IV (batches 10–12), VII (batches 19–21), and VIII (batches 22–24), which correspond to the formulations containing lactose (LAC high level), are located at the positive side of PC1 (Fig. 3), except to average III (possibly due to batch 8). Those formulations have higher friability values (Table 2).

PC2 or factor 2 describes 19 % of data information and expresses the DIS level. Averages I (batches 1–3), II (batches 4–6), III (batches 7–9), and IV (batches 10–12) represent formulations containing DIS low level. All those, except to average II (probably due to batch 6), are located at the positive side of PC2 (Fig. 3) and correspond to the formulations presenting higher hardness values (Table 2).

Moreover, averages V (batches 13–15), VI (16–18), VII (19–21), and VIII (22–24) are at the negative side of PC2 (Fig. 3) and are related to the formulations containing DIS high level. Regarding the dissolution property, the presence of a disintegrant agent decreases the tablet's disintegration time (t_1, Hixson–Crowell model [21]) and, consequently, increases the

dissolution release rate, which reflects directly in the amount of drug dissolved ($W_0^{1/3} - W^{1/3}$).

The batches 2 and 8 of averages I and III, respectively, were probably responsible for the increase of the hardness values of those tablet formulations (I → MCC low level, LAC low level and DIS low level; III → MCC low level, LAC high level and DIS low level).

Formulation containing MCC high level, LAC high level and DIS high level presented the highest friability value (batches 22–24; average VIII). Although all friability values were according to the official specifications, high friability values are not desirable for any tablet formulation.

The suitable composition can usually be defined regarding the dissolution profile of a tablet formulation, which is a biopharmaceutical property. Formulation containing MCC low level, LAC low level, and DIS high level presented the highest value of the amoxicillin amount dissolved in 60 min (batches 13–15; average V) (see Fig. 3).

4. Conclusions

The full factorial design using multiple and individual response evaluation was a useful tool to explore the influence of the adjuvants (MCC, LAC, and DIS) on the relevant physicochemical and biopharmaceutical properties (hardness, friability and dissolution profile) of the investigated amoxicillin tablet formulations.

The individual response analysis indicates the composition MCC low level, LAC low level, and DIS high level as the most suitable amoxicillin tablet formulation.

Considering the PCA biplot, high values of amount of drug dissolved in 60 min are favorable responses for the investigated amoxicillin tablet formulations, whereas high values of friability are not desirable.

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