Original Article

Formulation, development, and optimization of immediate release nateglinide tablets by factorial design

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ABSTRACT: In the present study, selection of superdisintegrants among sodium starch glycolate, cross povidone, Starch-1500 and cross carmellose sodium (CCS) was carried out for development of immediate release nateglinide tablets (NTG). A 3^2 full factorial design was used to investigate the influence of two independent variables, *i.e.*, amount of selected superdisintegrants and hardness of the tablets, on two dependent variables, *i.e.*, disintegration time and percentage of drug release at 30 min ($DR_{0.5h}$). The results revealed that CCS was the best superdisintegrant for the development of immediate release tablets of NTG. The sign of the coefficient of the polynomial equation signified that the disintegration time was decreased and $DR_{0.5h}$ was increased by decreasing the hardness of the tablets as well as by increasing the concentration of CCS in the tablets. A checkpoint batch of the tablets was prepared by changing the value of independent variables within the range used in the preparation of factorial batches of tablets to check the validity of the evolved optimized mathematical model. Stability studies of optimized formulations indicated that there was no significant change in the physical parameters, disintegration time, and percentage of drug release of tablets. The systematic formulation approach helped to understand the effect of formulation processing variables.

Keywords: Nateglinide, immediate release tablets, factorial design, superdisintegrants

1. Introduction

Despite increasing interest in controlled release drug delivery systems, more attention has been paid to formulate poorly soluble drugs as conventional tablets intended to be swallowed to disintegrate and release their medicaments rapidly in the gastro intestinal tract (GIT) for bioavailability. Bioavailability of a poorly soluble drug from a solid oral dosage form depends on the release of the drug substance from the dosage form, *i.e.*, disintegration of the solid oral dosage form which will increase the wettability of the drug by increasing the surface area of the drug particles (*1-3*). This highlights the importance of proper choice of disintegrant/superdisintegrants, *e.g.* cross carmellose sodium (CCS), sodium starch glycolate, cross povidone, Starch-1500, *etc.* and its consistency of performance which is of critical importance to increase the rate of dissolution and hence its bioavailability (*4,5*). There are various factors like hardness, concentration of binders, disintegrants, *etc*. which affect the disintegration time and rate of dissolution of the drug. Nateglinide (NTG) is a meglitinide derivative which is mainly used for the treatment of type-2 diabetes mellitus (*6,7*). The poor aqueous solubility of NTG gives rise to difficulties in the formulation of tablets with a desired dissolution rate. The present study aims to optimize the concentration of superdisintegrants and hardness of immediate release NTG tablets.

2. Materials and Methods

2.1. *Materials*

NTG was obtained as a gift sample from Glenmark Pharmaceuticals, Nashik, India. Lactose, CCS, sodium starch glycolate, cross povidone, Sarch-1500, polyvinylpyrolidine, Aerosil®, talc, and ethanol were procured from SD Fine Chemicals, Mumbai, India. All other chemicals used were of analytical grade.

2.2. *Differential scanning calorimetry (DSC)*

A differential scanning calorimeter (JADE DSC; PerkinElmer, Waltham, MA, USA) was used for thermal analysis of drug and drug-excipients mixtures (*8*). Excipients that were used in the development of

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formulation and their maximum ratio with drug used in a tablet were selected for the present study (Table 1). Individual samples (drug and excipients) as well as physical mixtures of drug and selected excipients (all passed through an 80-mesh sieve) were weighed directly in the DSC aluminum pan and scanned in the temperature range of 50-300°C under a nitrogen atmosphere. A heating rate of 20°C/min was used and thermograms obtained were observed for any interactions.

2.3. *Isothermal stress testing*

For isothermal stress testing (*8*,*9*), drug and different excipients were weighed directly in 4 mL-glass vials (n = 2) and mixed on a vortex mixer for 2 min. In each of the vials, 10% (w/w) water was added and the drug-excipients blend was further mixed using a glass capillary (both the ends of which were heat sealed). To prevent any loss of material, the capillary was broken and left inside the vial. Each vial was sealed using a teflon-lined screw cap and stored at 50°C in a hot air oven (Narang Scientific Industries, Haryana, India). These samples were periodically examined for any unusual color change. After 3 weeks storage under the above conditions, samples were quantitatively analyzed using a UV-visible spectrophotometer. Drug-excipients blends without added water, stored in a refrigerator, served as controls.

2.4. *Preparation of immediate release tablets*

NTG, half quantity of CCS, and lactose were passed through a No. 80 sieve and were mixed in a poly bag for 20 min. An alcoholic solution of povidone (10%, w/v) was added to the mixture in a quantity just sufficient to bind the mass. The wet mass was screened with a No. 20 sieve and dried in a tray dryer at 45°C. The dried granules were mixed with the remaining half of CCS and lubricated with 2% (w/w) talc and 2% (w/w) Aerosil. The granules ready for compression were converted into an 8 mm diameter size tablet using a single-punch tablet punching machine (Hardik, Ahmedabad, India). The composition of the preliminary and factorial design batches is shown in Tables 2 and 3, respectively.

2.5. *Evaluation of granules and tablets*

The prepared granules were evaluated for angle of repose, bulk density, tapped density, compressibility index, and Hausner ratio (*10*). The prepared tablets were evaluated for weight variation, friability, hardness, thickness, and disintegration time (*11*).

The *in vitro* drug release study was carried out in a United States Pharmacopoeia (USP) dissolution apparatus II (TDT-08L; Electrolab, Mumbai, India) using a rotating paddle at 50 rpm in 1,000 mL of 0.1 N HCl + 0.5% sodium lauryl sulphate as dissolution medium while maintaining the temperature at 37 ± 0.5°C (*12*). An

Table 1. Results of analysis of isothermal stress testing samples after 3 weeks of storage at stressed conditions

Samples	Ratios (drug-excipient)	% Drug remaining[a]	
		Control samples[b]	Stressed samples[c]
NTG	–	100.8 ± 0.7	99.7 ± 2.5
NTG + lactose	1:2	101.4 ± 1.3	100.3 ± 1.7
NTG + CCS	1:1	100.7 ± 1.8	99.8 ± 1.8
NTG + povidone	2:1	101.8 ± 1.5	99.3 ± 1.9
NTG + talc	3:1	100.6 ± 2.7	99.3 ± 2.0
NTG + Aerosil	3:1	100.5 ± 1.8	99.6 ± 1.2

[a] Values expressed as average ± standard deviation; [b] Drug excipient blends without added water and stored in refrigerator; [c] Drug excipient blends with 10% (w/w) added water and stored at 50°C for 3 weeks.

Table 2. Results of preliminary batches of immediate release NTG tablets

Items	Formulation[a]				
	T-1	T-2	T-3	T-4	T-5
Composition					
NTG (%, w/w)	40	40	40	40	40
Sodium starch glycolate (%, w/w)[b]	–	6	–	–	–
Starch-1500 (%, w/w)[b]	–	–	6	–	–
Cross povidone (%, w/w)[b]	–	–	–	6	–
CCS (%, w/w)[b]	–	–	–	–	6
Lactose	q.s.[c]	q.s.	q.s.	q.s.	q.s.
Disintegration time (sec)	> 3,600	> 3,600	> 3,600	136	20
$DR_{0.5h}$ (%, w/w)	3.2	37.7	7.2	62.6	100.6

[a] All batches contained 10% (w/w) polyvinylpyrrolidone in ethyl alcohol as a binder, 2% (w/w) talc, and 2% (w/w) Aerosil. Hardness of all batches was 5 kg/cm^2. [b] Intragranular 50% (w/w), extragranular 50% (w/w). [c] quantity sufficient for 100% (w/w).
Abbreviations: NTG, nateglinide; CCS, Cross carmellose sodium; $DR_{0.5h}$, Drug release at 0.5 h.

Table 3. 3^2 Full factorial design layout

Batch code[a]	Variable levels in coded form[b]		Dissolution at 0.5 h[c] (%, w/w)	Disintegration time[c] (sec)
	X_1 (%)	X_2 (kg/cm^2)		
F-1	–1	–1	67.8 ± 0.4	158.7 ± 3.1
F-2	–1	0	61.7 ± 1.0	179.3 ± 2.5
F-3	–1	+1	54.9 ± 0.8	193.3 ± 4.5
F-4	0	0	100.3 ± 0.8	33.0 ± 2.7
F-5	0	–1	94.3 ± 0.6	53.7 ± 3.1
F-6	0	+1	91.4 ± 1.2	39.7 ± 1.5
F-7	+1	–1	100.0 ± 0.4	17.3 ± 1.5
F-8	+1	0	100.5 ± 0.7	19.7 ± 1.5
F-9	+1	+1	100.1 ± 0.3	30.0 ± 1.0
Check point	–0.5	+0.5	77.8 ± 0.7	100.4 ± 3.3

[a] All batches contained 10% (w/w) polyvinylpyrrolidone in ethyl alcohol as a binder, 2% (w/w) talc, and 2% (w/w) Aerosil. [b] X_1, amount of cross carmelose sodium (%, w/w); X_2, hardness of the tablets (kg/cm^2). [c] Data are shown as mean ± S.D.

Table 4. Actual value of codes mentioned in Table 3

Codes values	Actual value	
	X_1*	X_2*
–1	2	3
0	4	5
+1	6	7
–0.5	3	4
+0.5	5	6

* X_1, amount of cross carmelose sodium (%, w/w); X_2, hardness of the tablets (kg/cm^2).

aliquot of 5 mL was withdrawn at different time intervals (5, 10, 20, 30, and 45 min) and an equal volume of fresh dissolution medium was added to maintain the sink condition. Samples were suitably diluted and analyzed in a UV-visible spectrophotometer at 210 nm to determine the amount of drug released (*13*). The fresh dissolution medium replacement was considered in the calculation of the amount of drug released.

2.6. *Full factorial design*

A 3^2 randomized full factorial design was used to optimize the variables in the present study. In this design 2 factors were evaluated, each at 3 levels, and experimental trials were performed at all 9 possible combinations (*14-16*). The percentage (2, 4, and 6%) of CCS (X_1), and hardness (3, 5, and 7 kg/cm^2) of tablets (X_2), were selected as independent variables. The disintegration time and percentage of drug released at 0.5 h ($DR_{0.5h}$) were selected as dependent variables.

2.7. *Stability study*

The factorial batches were subjected to accelerated stability (40 ± 2°C/75 ± 5% relative humidity) testing. After a specified period of time (1, 2, and 3 months), samples were withdrawn and subjected to assay, disintegration time and *in vitro* dissolution studies. The stability study was conducted as per the International Conference on Harmonization (ICH) guidelines (*17*).

3. Results and Discussion

A drug-excipient interaction study at an early stage of product development is an important exercise in the development of a stable dosage form. As shown in Figure 1, a sharp endothermic peak was observed at 143°C in the DSC thermogram of NTG. However, the endothermic peak of NTG was well preserved at 143 ± 2°C in the DSC thermogram of NTG-excipients mixtures (Figure 1). This result inferred that there was no interaction between drug and excipients (*8*).

In isothermal stress testing, it was observed that there was no physical change (color and appearance) as well as drug content after storage of drug-excipient blends under stressed conditions (Table 1), which supported previously reported results of a DSC study on drug-excipient compatibility testing (*9*).

The flow properties of granules can be judged from the angle of repose, compressibility index and Hausner ratio (*10,11*). An angle of repose (θ) < 30° indicates free flowing material and > 40° poor flow properties (*10*). A compressibility index < 10% indicates excellent flow properties and > 38% poor flow properties (*11*). A Hausner ratio of 1.00-1.11 indicates free flowing and > 1.60 poor flow properties (*11*). Values for angle of repose (θ), compressibility index (%), and Hausner ratio for all prepared granules were found to be in the range of 22.5 to 25.7°, 7.14 to 7.71%, and 1.07 to 1.08, respectively (Table 5), which showed that the granules were free flowing and can be used for tablet compression. A percentage of weight variation was observed within the limit of ± 7.5% (w/w) for all the prepared tablets (Table 5), which is acceptable for uncoated tablets as per United State Pharmacopoeia, National Formulary (USP-NFXXVI) (*18*). A friability test of the prepared tablets except batches of formulation-1 to formulation-3 (F-1 to F-3) passed (weight loss < 1%, w/w) (Table 5), which assumed that tablets of formulation-4 to formulation-9 (F-4 to F-9) prepared at higher hardnesses have sufficient mechanical integrity and strength.

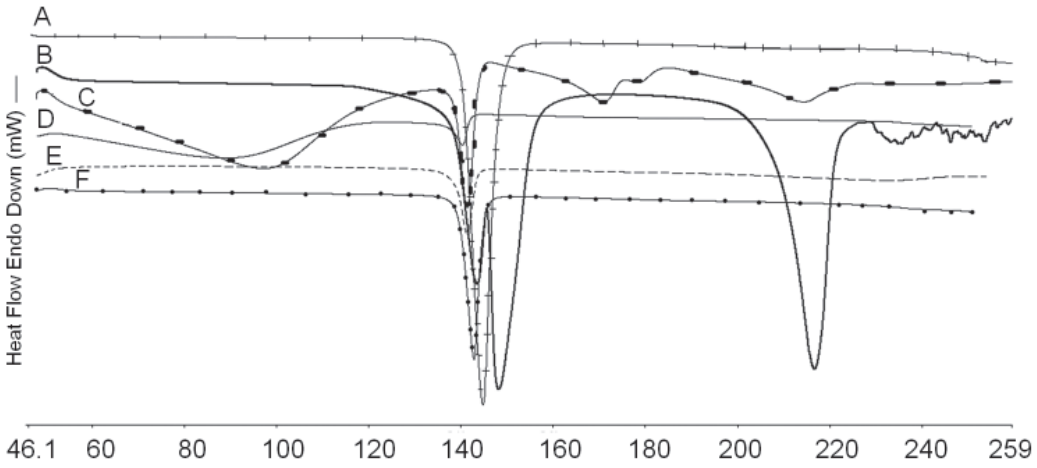

Figure 1. Comparision of DSC thermogram of NTG with drug-excipients mixture. A, NTG; B, Lactose + NTG; C, Cross carmelose sodium + NTG; D, Povidone + NTG; E, Talc + NTG; F, Aerosil + NTG.

Table 5. Evaluation of granules and tablets

Items	F-1	F-2	F-3	F-4	F-5	F-6	F-7	F-8	F-9
Evaluation of granules									
Angle of repose (θ)	22.9	24.8	23.3	22.5	24.2	25.7	23.8	25.3	24.2
Compressibility index (%)	7.63	7.14	7.37	7.63	7.36	7.48	7.52	7.26	7.36
Hausner ratio	1.07	1.07	1.08	1.07	1.08	1.07	1.07	1.08	1.08
Evaluation of tablets									
Weight variation (%, w/w)	3.2	4.8	3.1	2.8	5.2	4.6	3.7	3.7	4.2
Friability weight loss (%, w/w)	2.7	1.4	1.9	0.4	0.2	0.4	0.6	0.2	0.3

Lactose was selected as a soluble diluent for the water insoluble drug NTG by considering its advantages in terms of easy availability, cost-effectiveness, and relative moisture insensitivity. Povidone was used as a binder considering its widespread applicability in industry. The preliminary trial batches were conducted arbitrarily without addition of superdisintegrants (T-1) and in addition with 6% (w/w) of sodium starch glycolate (T-2), Starch-1500 (T-3), cross povidone (T-4), and CCS (T-5) at 5 kg/cm^2 hardness for the selection of superdisintegrants. As shown in Table 2, while the disintegration time of T-1, T-2 and T-3 were more than 3,600 sec and the disintegration time of T-4 was 136 sec, the batch containing CCS (T-5) showed a lower disintegration time and more $DR_{0.5h}$ than other superdisintegrant containing batches. Hence CCS was considered for further investigation. Hardness of the tablets has an impact on the disintegration time as well as the amount of drug released from tablets (*3*), therefore it needs to be optimized. In order to investigate the influence of concentration of CCS and hardness of the tablet systematically, a 3^2 factorial design was employed in this investigation.

The amount of CCS (X_1) in tablets and the hardness of the tablets (X_2) were chosen as independent variables in a 3^2 full factorial design. A statistical model incorporating interactive and polynomial terms was used to evaluate the responses.

$$Y = b_0 + b_1X_1 + b_2X_2 + b_{12}X_1X_2 + b_{11}X_1^2 + b_{22}X_2^2 \quad \text{--- Eq. 1}$$

where, Y is the dependent variable, b_0 is the arithmetic mean response of the 9 runs, and b_i is the estimated coefficient for the factor X_i. The main effects (X_1 and X_2) represent the average result of changing one factor at a time from its low to high value. The interaction terms (X_1X_2) showed how the response changes when two factors are simultaneously changed. The polynomial terms (X_1^2 and X_2^2) are included to investigate nonlinearity. As shown in Table 3, the disintegration time and $DR_{0.5h}$ for the 9 batches (F1 to F9) showed a wide variation (*i.e.*, 17.3 to 193.3 sec and 54.9 to 100.5% (w/w), respectively). The data indicates that the dependent variables such as disintegration time and $DR_{0.5h}$ are dependent on the selected independent variables such as concentration of CCS and hardness of the tablets. The fitted equations (full and reduced) relating the disintegration time responses and $DR_{0.5h}$ to the transformed factor are shown in Table 3. The polynomial equations can be used to draw conclusions after considering the magnitude of the coefficient and the mathematical sign it carries (*i.e.*, positive or negative). Table 6 shows the results of the analysis of variance (ANOVA), which was performed to identify insignificant factors (*15*). High values of the correlation coefficient for disintegration time and $DR_{0.5h}$ indicate a good fit (Table 6). The equations may be used to obtain estimates of the response because a small error of variance was noted in the replicates.

The significance test for regression coefficients was performed by applying the Students' *t* test. A coefficient

Table 6. Calculations for testing the model in portions

Items	DF	SS	MS	F	R^2
For % of dissolution at 0.5 h					
Regression					
FM	5	8.38×10^3	1.68×10^3	1.66×10^3	0.9969
RM	4	8.38×10^3	2.09×10^3	2.14×10^3	0.9970
Error					
FM	3	11.5	3.82		
RM	4	11.8	2.96		
For disintegration time					
Regression					
FM	5	1.304×10^5	2.61×10^4	2.76×10^3	0.9981
RM	4	1.304×10^5	3.26×10^4	3.40×10^3	0.9980
Error					
FM	3	77.6	25.9		
RM	4	90.1	22.5		

Abbreviations: DF, degrees of freedom; SS, sum of squares; MS, mean of squares; F, Fischer's ratio; R^2, regression coefficient; FM, full model; RM, reduced model. Details of calculations are shown in Mendenhall W and Sincich T (*19*).

Table 7. Summary of Regression Analysis Results

Items	b_0	b_1	b_2	b_{11}	b_{22}	b_{12}
For % of dissolution at 0.5 h						
Respons (% of dissolution at 0.5 h)						
FM	95.19	19.36	–3.62	–14.52	0.25	3.28
RM	95.36	19.36	–3.62	–14.52		3.8
For disintegration time						
Respons (disintegration time)						
FM	41.15	–77.39	11.33	57.61	1.44	–5.5
RM	42.11	–77.39	11.33	57.61		–5.5

Abbreviations: FM, full model; RM, reduced model.

is significant if the calculated t value is greater than the critical value of t. The significance level of coefficient b_{22} was found to be $p = 0.263$ and 0.549 in the full model for disintegration time and $DR_{0.5h}$, respectively (data not shown); therefore it was omitted from the full model for both cases to generate the reduced models. The results of statistical analysis are shown in Table 7. The coefficients b_1, b_2, b_{11}, and b_{12} were found to be significant at $p < 0.05$, hence they were both retained in the reduced model. The reduced model was tested in portions to determine whether the coefficient b_{22} contributes significant information for the prediction of disintegration time and $DR_{0.5h}$ or not (*16*). The results of multiple linear regression analyses (reduced model) reveal that, when increasing the concentration of CCS, a decrease in disintegration time is observed and the b_1 coefficients bear a negative sign. When the b_2 coefficient bears a positive sign it signifies an increase in disintegration time on decreasing the hardness (kg/cm^2) of the tablets. An increase in the value of $DR_{0.5h}$ is observed on increasing the concentration of CCS (b_1 is positive) and on decreasing the hardness (b_2 is negative) of the tablets.

The response surface plot of % of CCS (X_1) and hardness of the tablets (X_2) versus disintegration time and those versus % of $DR_{0.5h}$ are shown in Figures 2A and 2B, respectively. The response plot showed that there is a significant effect of both factors on selected response.

The model predicts the required disintegration

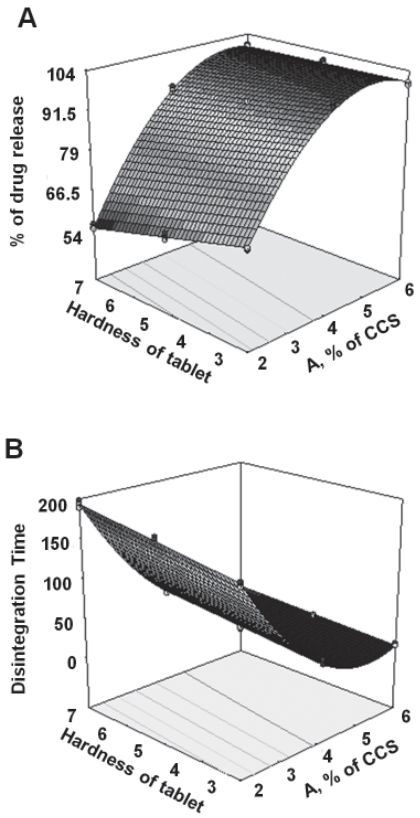

Figure 2. Response surface plot of % of drug release at 0.5 h (A) and disintegration time (B).

time (as per USP limits) and $DR_{0.5h}$ (100%) from point prediction, at 4% CCS and 5 kg/cm^2 hardness (F-5). Though the model also predicted the required response at a lower concentration of CCS and hardness; the tablets did not have a disintegration time within 3,600 sec at lower CCS concentrations and failed to pass the friability at lower hardness.

A checkpoint batch was prepared at $X_1 = -0.5$ level and $X_2 = +0.5$ (Table 3). From the reduced model, it is expected that the $DR_{0.5h}$ value of the checkpoint batch should be 77.8 and the value of disintegration time should be 100.4 seconds (Table 3). Thus, from the results of statistical optimization techniques it can be concluded that all of the models are mathematically significant.

Stability studies of factorial batches indicate no significant change in appearance of the tablets assay ($p < 0.05$), disintegration time ($p < 0.05$), and percentage drug release ($p < 0.05$).

4. Conclusion

In this study, the dissolution of poorly soluble drug NTG was significantly enhanced by using CCS compared to sodium starch glycolate, cross povidone and Starch-1500 as superdisintegrants in immediate release tablets. The results of a 3^2 full factorial design revealed that the amount of CCS and hardness of the tablets significantly affect the dependent variables, disintegration time, and percentage of $DR_{0.5h}$ from the tablets. It is concluded that by adopting a systematic formulation approach, an optimum point can be reached in the shortest time with a minimum effort. Tablets containing 4% (w/w) of CCS at 5 kg/cm^2 met the required disintegration time (as per USP limit) and $DR_{0.5h}$ (100%).

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