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OPTIMISATION METHOD USED FOR ALUMINIUM – BASED ANTACID PREPARATION

The aim of this work was to optimize the rheological properties of $AlPO_4$ suspension, the main ingredient of the antacid preparation – Alfogel. The exact rheological characterisation of the system is very important due to its stability and transportation through the pipe system into the reaction vessel. Box–Behnken design was used to estimate the effect of the concentrations of cations and the content of active pharmaceutical ingredient on the rheological properties of the system. The relationship between one or more response functions and a set of independent variables can be examined by using response surface methods, such as Box–Behnken designs. The concentration of Na^+ and Mg^{2+} ions as well as the content of $AlPO_4$, ranged from the optimum set of conditions to their minimum and maximum values. The viscosity, yield stress point and thixotropy of the colloidal system were used to assess the rheological properties of the $AlPO_4$ suspension. The system was characterised using a second order polynomial mathematical model. The process was optimised and the proposed model was confirmed in laboratory scale experiments.

Key words: Box–Behnken, experimental design, optimisation, aluminium phosphate, rheological properties, viscosity, thixotropy, yield stress point.

Aluminium compounds, such as hydroxide, oxide, phosphate, magaldrate, hydrotalcite and almagate, are widely used as antacid preparations in the treatment of mild gastro–oesophageal reflux disease and heartburn. Aluminium hydroxide is most commonly used, due to its great acid neutralizing capacity however, it has been reported to be less effective compared to other antacid compounds, such as magnesium hydroxide, calcium carbonate and sodium bicarbonate, or the "lattice–layer" aluminium–magnesium hydroxide complexes [1]. Antacids containing both aluminium and magnesium salts are an ideal combination, as one ingredient complements the other. Aluminium salts, which dissolve slowly in the stomach, start to work gradually, but provide long–lasting relief. They can also cause constipation. Magnesium salts act fast and neutralize acids effectively. In addition, they can act as a laxative. Hence, antacids containing both ingredients should provide quick, long–lasting relief, with less risk of diarrhoea or constipation.

Galenika's pharmaceutical preparation Alfogel is an aluminium phosphate – based antacid, with added organic polymer as well as inorganic salts. This mixture has excellent adhesive properties to gastric mucosa, especially to damaged gastric and duodenal mucosa. Aluminium phosphate – based antacids do not cause the rebound effect, otherwise seen in formulations made with aluminium or magnesium hydroxide or calcium carbonate. Antacid preparations are made by mixing an inorganic phase made of some neutralising compound and an organic phase, usually made of organic polymer. The purpose of using organic polymer is that they increase the adhesion of the antacid preparation to the damaged intestinal mucosa. It is very important to find the proper ratio of the inorganic and organic phase of the product. The rheological behaviour of the product is also very important (from the manufacturer's point of view) and greatly influenced by this ratio.

The pre–formulation experiments are most commonly performed by changing, in an unsystematic manner, the level of each variable (factor), with only one variable being changed at a time, keeping all other variables unchanged in order to study the effects of one specific variable on the selected response. In this way it is very difficult to find the optimal condition of all of the variables of the entire system. Such methodology requires an enormous number of experiments and relies greatly on the analyst's experience [2].

This traditional approach is not an efficient and economic strategy, as it cannot provide information of the optimum of the entire system and can lead only to a determination of the local optimum of the system. The one–at–a–time factor optimisation neglects the interaction between the factors and requires unnecessary numerous runs. The relationship between one or more response parameters and a set of independent (input) variables can be examined well by using response surface methods, such as Box–Behnken designs [3]. Response surface methodology is applicable once the preliminary screening has been performed on a set of data, by applying primary factor screening using, for example, factorial designs, to determine which factors significantly affect the response.

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We have previously determined, using a two–level, full factorial design, that there are two major groups of parameters that can influence the rheological behaviour of the final antacid preparation, namely:

1) organic substances that are mixed together and added into the inorganic phase of the product (agar–agar, pectin, Na–CMC, sucrose);

2) inorganic substances, (aluminium phosphate, sodium chloride, magnesium chloride).

The initial step of the manufacturing process is actually mixing inorganic substances to be used as a vehicle for inorganic substances which are incorporated. The rheological behaviour of this inorganic phrase is very important for the manufacturing process and for the rheology of the final product.

The objective of this study was to evaluate and determine the way rheological parameters are affected by changing the ratio of inorganic phase substances. To estimate the factor of the effects in this multi–component system, we used Response Surface Modelling (RSM) Design – Box–Behnken Design. This particular design is appropriate for fitting second order response models. They are rotatable or nearly rotatable, depending on the number of factors. This design is "corner free", no runs are done at the design corners, which means that there are no experiments where at least one of the factors is not in its midpoint [4]. The application of this methodology was previously used in optimisation for the food manufacturing technology process, microbiological studies and pharmaceutical formulation development [5].

2. MATERIAL AND METHODS

2.1. Instrumentation

A RheoStress 1 rheometer with a thermocontroller unit was used in all of our rheological measurements. Homogenisation of the samples was performed using an IKA – Werke EuroStar Digital Stirrer and T25 Turbo Homogeniser, whereas the pH of the specimens was measured using a MeterLab PHM220 Lab pH–meter.

The electrophoretic mobility and, subsequently, the point of zero charge were determined in cooperation with the Institute of Nuclear Sciences – Vinča.

Chemicals and reagents

All the reagents used were of analytical grade, except for aluminium phosphate, which was synthesized by using a semi – continuous model of the process [6].

Procedure

Synthesis of aluminium phosphate

Aluminium phosphate was synthesized in a way to obtain a thixotropic system of the required pH value, point of zero charge (PZC) viscosity, and neutralising capacity.

The aluminium phosphate that we prepared was chemically amorphous aluminium hydroxyphosphate [7]. It is not a stoichiometric compound and therefore has no fixed hydroxyl to phosphate ratio. It is likely that the conditions during precipitation affect the hydroxyl to phosphate ratio of the precipitate and, consequently, the charge of the particle surface. We precipitated $AlPO_4$ at different pH values and measured the point of zero charge, as described by Burrell and al. [8] and subsequently decided to keep the appropriate hydroxyl to phosphate ratio so as to obtain a thixotropic system at pH 5.5–6.0. Our findings suggest that in this pH range, the surface charge of $AlPO_4$ particles was negative. These data are in the correlation with the findings presented by Burell et al (2001). The hydroxyl to phosphate ratio at the time of precipitation was held constant by pumping a solution of phosphate and aluminium ions (solution one) and a solution of sodium hydroxide (solution two) into a reaction vessel at rates which maintained a constant pH in the reaction vessel.

Preliminary experiments and factor selection

Preliminary experiments were performed to determine the effect factors as well as their design space. We estimated their effect using two–level six factorial design (data not shown). The conclusion was that all of the inorganic phase substances and some of organic phase substances are involved in interactions that result in the rheological properties of the final product. Precise rheological characterisation of the inorganic phase is very important due to its stability and transportation through the pipe system into the reaction vessel.

Experimental design

A three–factor three–level Box–Behnken experimental design was used to evaluate the effects of selected independent variables on the response and to optimise the procedure. The levels were selected based on knowledge of the system, acquired from the initial experiment trials. The following parameters, listed in Table 1 ranged from the optimum set of conditions to their minimum and maximum values. The design consisted of replicated centre points (run 9–17) and a set of points lying at the midpoints of each edge of the multidimensional cube that defines the region of interest.

Table 1. Factors and factor levels used for Box–Behnken experimental design

factor		level		
		–1	0	+1
X_1	C Na^+ (mol/L)	0.005	0.0175	0.03
X_2	C Mg^{2+} (mol/L)	0.005	0.0175	0.03
X_3	% $AlPO_4$	10	15	20

The stochastic mathematical model had the following form:

$$y = b_0 + b_1x_1 + b_2x_2 + b_3x_3 + b_4x_1x_2 + \\ + b_5x_2x_3 + b_6x_1x_3 + b_7x_1^2b_8x_2^2 + b_9x_3^2 + E. \quad (1)$$

In this equation, y is the selected response function, b_0 – b_9 are the regression coefficients, x_1 – x_3 are the factors involved in interaction, and E is the error. The inclusion of centre points provided a more precise estimate of the experimental error and provided a measure for the adequacy of the model (lack of fit). It also enabled the determination of the significance of the main, interaction and quadratic effects.

Three response functions were utilized for the determination of the optimum conditions. The first response was the viscosity of the system. After the process of synthesis, the $AlPO_4$ suspension was rigorously mixed by stirring for twenty minutes, using a high speed stirrer. The stable and homogeneous suspension was then allowed to age and stabilize at room temperature for 24 hours. Before the measurements, a predesigned amount of electrolyte was added and the suspension was vigorously stirred for fifteen minutes. The sample was then allowed to rest for five minutes, after which the viscosity of the system was measured for ten minutes, at one minute intervals. The extrapolation shown in Figure 1 was used to obtain a single value from these measurements.

The second response tested was the yield stress. It is defined as the minimum required force to initiate flow. A curve analysis was performed to look for the bending points of the deformation, where the stress curve was presented in a double logarithmic scale (Figure 2).

Table 2. Coded values for experiments 1–17 used for Box–Behnken experimental design

Experimental run	factor and factor level		
	X_1	X_2	X_3
1	–1	–1	0
2	+1	–1	0
3	–1	+1	0
4	+1	+1	0
5	–1	0	–1
6	+1	0	–1
7	–1	0	+1
8	+1	0	+1
9	0	–1	–1
10	0	+1	–1
11	0	–1	+1
12	0	+1	+1
13	0	0	0
14	0	0	0
15	0	0	0
16	0	0	0
17	0	0	0

Table 3. Responses selected for Box–Behnken experimental design

Response	
y_1	viscosity
y_2	yield stress point
y_3	thixotropy

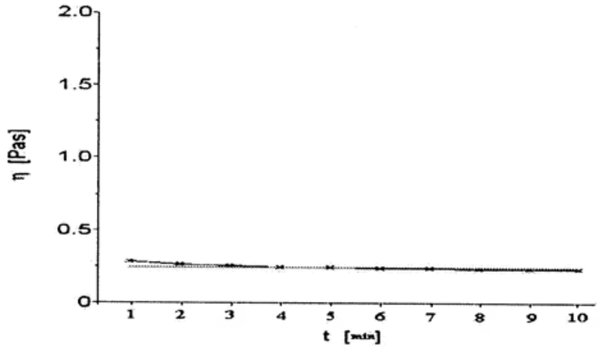

Figure 1. Determination of the viscosity of the system

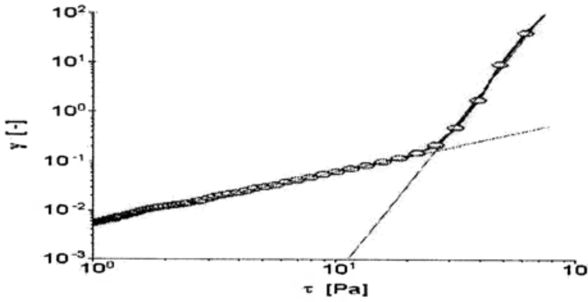

Figure 2. Determination of the yield stress of the system

The third response tested was the thixotropy of the system. The thixotropic loop is a test procedure to determine the effect of time related to the flow properties. It consists basically of an upward and downward sheer ramp. The test is supposed to be isothermal, so the temperature was kept at 25°C. The measurement data are usually presented in a linear diagram of the shear stress as a function of the shear rate. When ramping up, the material is exposed to shear forces which will destroy its internal structure. The downward ramp will show lower stress values at the corresponding shear rate than the upward curve if the material cannot rebuild its structure rapidly enough. This behaviour creates an area between the up and down curve (Figure 3). By calculating this area we obtained a value to characterize the thixotropy of the material.

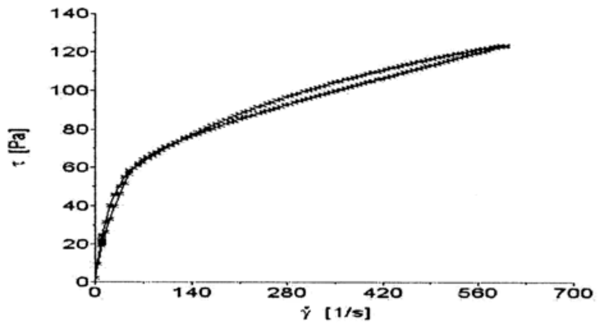

Figure 3. Determination of the thixotropy of the system

The prediction plot, showing the effects of the investigated parameters on the rheological properties of $AlPO_4$ is shown in Figure 4.

RESULTS AND DISCUSSION

Experimental considerations

Thixotropy is a complex rheological phenomenon, and up to now, the mechanism of thixotropic phenomena has not been thoroughly understood. Earlier study of the thixotropy of some Mg–Al–compounds proposed a mechanism of dispersion particle–steric network–dense floc units [9].

High–speed stirring may destroy some of these structures and thereby enhance the dispersity of the system. During the settling or under low shear rate, these destroyed structures may be recovered, because the particles come close each other to form continuous network structures and the suspension will show an increasing of the thixotropy value. We found that $AlPO_4$ particles have negative charge at pH values below 6, if they are precipitated at pH 5.5–6.0. It was supposed that

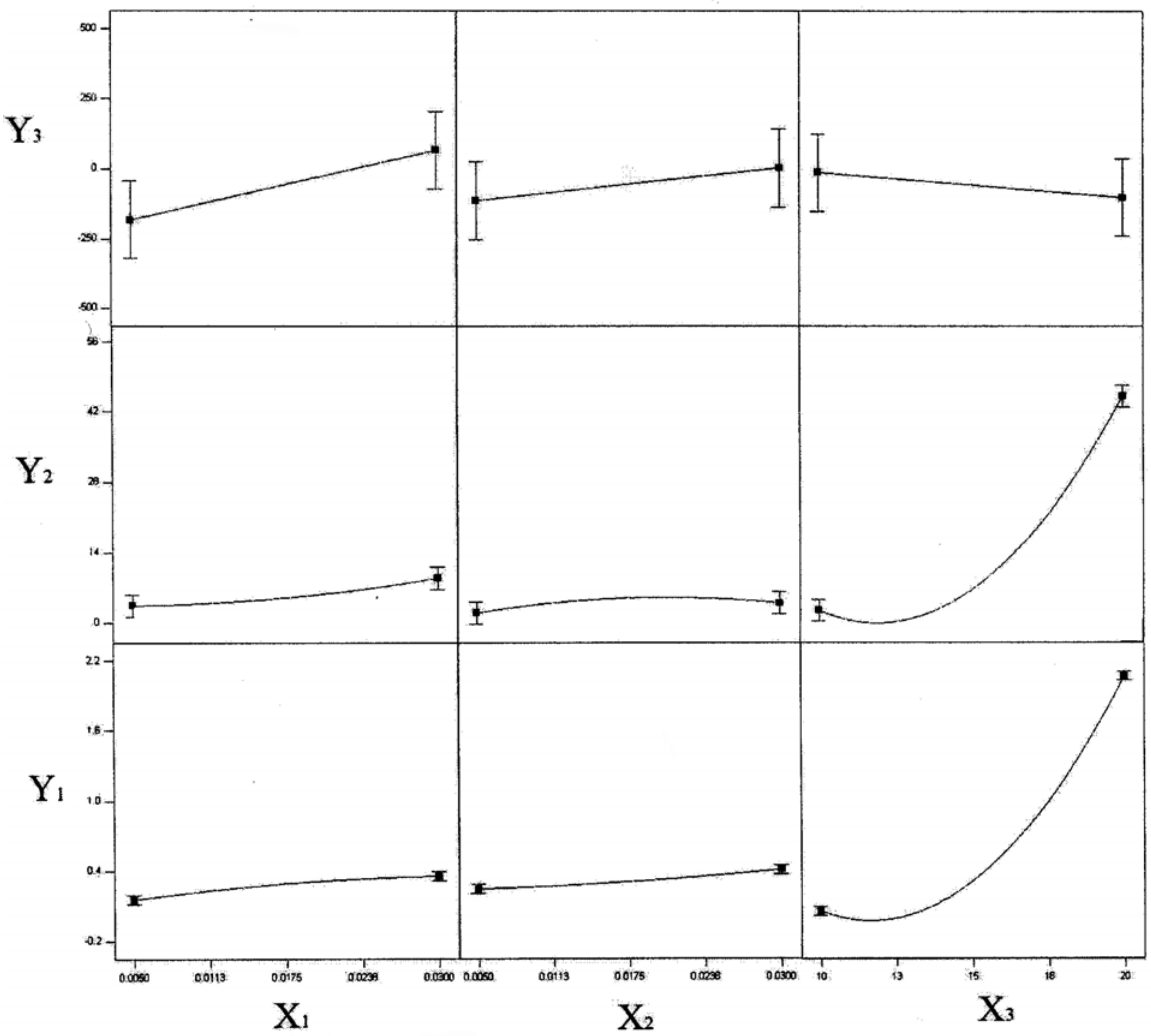

Figure 4. Prediction of the effects of the analyzed parameters

particles precipitated under such conditions affect each other by electrostatic interactions (primary, repulsive forces) and the addition of ions can affect the mode of particl interaction because of the compression of the electrical double layer which was the cause of the thickening of the investigated suspension system (homogeneous coagulation). We supposed that Na^+ and Mg^{2+} cations would adhere to the negatively charged surface of these particles and compressed the electrical double layer, causing the particles to pack more tightly, which would make it possible to record changes in the values of thixotropy and viscosity as well as the flow ability of the suspension system.

Second order model and analysis of variance

In the investigated design space, the estimated coefficients in equation (1), were obtained by using the statistical software Design–Expert v6.0.0.1 The least squares regression method was performed. The resulting equations are as follows:

for viscosity:

$$Y_1 = [1/(1.85 - 0.49\ X_1 - 0.43\ X_2 - 2.18X_3 + 0.22X_1^2 + 0.047X_2^2 + 0.90\ X_3^2 + 0.28\ X_1X_2 + 0.49\ X_1X_3 + 0.44\ X_2X_3)]^2 \quad (2)$$

for yield stress:

$$Y_2 = (2.23 + 0.37\ X_1 + 0.27\ X_2 + 3.05\ X_3 - 0.12\ X_{1}2 - 0.21\ X_{2}2 + 1.55\ X_{3}2 - 0.28\ X_1X_2 + 0.34\ X_1X_3 + 2.554 \text{ x } 10^3\ X_2X_3)^2 \quad (3)$$

Table 4. Applied transformations for the response data

y_1	inverse square root
y_2	square root
y_3	none

for thixotropy:

$$Y_3 = -58.44 + 124.11\ X_1 + 58.58\ X_2 - 44.46\ X_3 + 12.38\ X_1X_2 + 258.90\ X_1X_3 + 180.58\ X_2X_3 \quad (4)$$

A positive value for the coefficient in equations 2–4 indicates a synergistic effect, while a negative one indicates an antagonistic effect upon the response. A larger coefficient (by its absolute value) means that the causal factor has more potential effect on the response. Table 7 presents the p–value estimation for all of the coded factor effects and for all the three responses. A factor has a statistically significant effect if p–value is less than 0.05. It can be seen from Table 7 that the response y_1 is significantly affected by X_1 (concentration of Na^+ ions), X_2 (concentration of Mg^{2+} ions) and X_3 (content of $AlPO_4$), as well as by the quadratic term of X_1 and X_3, and by the interaction effects of X_1X_2, X_1X_3 and X_2X_3.

Y_2 is significantly affected by X_1, X_3 and the quadratic term of X_3. A significant factor for the response Y_3 was only the interaction effect of factors X_1 and X_2.

The response data was transformed during data analysis of the Y_1 and Y_2 functions. Transformation is needed if the error (residuals) is a function of the magnitude of the response (predicted values), and in this case, transformation can be useful to stabilize the variance of the model. However, if the ratio of the maximum response to the minimum response is large, transforming the response will not make much difference. The transformation for each response is listed in Table 4.

An appropriate model was selected according to the F Value and the Prob>F value (p value). Values of Prob>F (p value) less than 0.05 indicate that the model

Table 5. Sequential Model Sum of SquaresSource

Source	Response								
	Y_1			Y_2			Y_3		
	Sum of Squares	F Value	Prob>F	Sum of Squares	F Value	Prob>F	Sum of Squares	F Value	Prob>F
Linear	41.33	30.57	<0.0001	76.24	28.15	<0.0001	1.665×10^5	1.08	0.3929
2FI	2.05	1.79	0.2125	0.77	0.23	0.8713	3.992×10^5	4.92	**0.0237**
Quadratic	3.74	115.83	**<0.0001**	10.18	29.94	**0.0002**	36024.43	0.36	0.7850

Table 6. Model Summary Statistic

Source	Response								
	Y_1			Y_2			Y_3		
	R–Squared	Adjusted R–Squared	Predicted R–Squared	R–Squared	Adjusted R–Squared	Predicted R–Squared	R–Squared	Adjusted R–Squared	Predicted R–Squared
Linear	0.8758	0.8472	0.7708	0.8666	0.8358	0.7388	0.1991	0.0143	–0.6715
2FI	0.9192	0.8707	0.7125	0.8753	0.8005	0.4345	0.6765	0.4824	–0.6415
Quadratic	0.9984	0.9964	0.9751	0.9910	0.9794	0.8568	0.7196	0.3590	–3.4359

Table 7. ANOVA for the RSM Model

Source	Response					
	Y_1		Y_2		Y_3	
	F Value	Prob>F	F Value	Prob>F	F Value	Prob>F
Model	486.79	**<0.0001**	85.49	**<0.0001**	3.49	**0.0398**
X_1	180.06	**<0.0001**	9.71	**0.0169**	4.56	0.0586
X_2	134.65	**<0.0001**	5.12	0.0581	1.02	0.3375
X_3	3528.56	**<0.0001**	658.04	**<0.0001**	0.58	0.4622
X_1^2	18.98	**0.0033**	0.50	0.5005	0.023	/
X_2^2	0.85	0.3872	1.70	0.2334	9.91	/
X_3^2	314.76	**<0.0001**	89.09	**<0.0001**	4.82	/
$X_1 X_2$	30.08	**0.0009**	2.73	0.1424	/	0.8833
$X_1 X_3$	88.40	**<0.0001**	4.04	0.0845	/	**0.0104**
$X_2 X_3$	71.89	**<0.0001**	$2.302\text{x}10^4$	0.9883	/	0.0528
Adeq Precisio	**69.442**		**27.807**		**7.258**	

Table 8. Limits used to define the acceptable design space

Response	Nominal value
Y_1	$0.8 < Y_1 < 2.282$
Y_2	$30 < Y_2 < 35$
Y_3	$-100 < Y_3 < 100$ Pa/s

is statistically significant. It was obvious that adding quadratic terms to the terms already in the model was significant for Y_1 and Y_2, whereas only linear terms addition was possible for Y_3.

A measure of the amount of variation around the mean explained by the model, R squared value showed that 99.84%, 99.10% and 67.65% of the variability of the data of Y_1, Y_2 and Y_3, respectively, could be explained by the model. Such behavior implies the the model function, used for the interpretation of the Y_3 response was less adequate than Y_1 and Y_2.

Lack of fit is a measure of the range for the predicted response relative to its associated error, actually a signal to noise ratio. Its desired value is 4 or more, and values of 69.442, 27.807 and 7.258 for Y_1, Y_2 and Y_3 respectively mean that the selected model can be used to navigate the design space.

The predicted R squared is a measure of the adequacy of the model predicting a response value. Values of 0.9751 and 0.8568 means that there is a good correlation between the polynomial equations and the Y_1 and Y_2 responses. However, the negative value of –0.6415 suggests that the used polynomial function for the description of the system is not adequate for all of the design points, especially the points at the edge of the design space.

Response surface plots

The relationship between the dependent and independent variables was further estimated by using three-dimensional (3D) plots for all of the measured responses. They were formed based on the model polynomial functions proposed. Since each model had more than two factors, one factor was held constant for each diagram, therefore, a total of 9 response surface diagrams were produced (3 for each response). The response surface plots are presented in Figure 5.

Optimisation of the rheological properties of the system

After generating the polynomial equations coorelating the dependent and independent variables, the process was optimised. Graphical and numerical optimisation was performed. In graphical optimisation the goal is to find a region (parts of the design space) where the requirements simultaneously meet the critical properties, the so called "sweet spot". By superimposing or overlaying the critical response contours on a contour plot it is possible to visually search for the best compromise. Graphical optimization displays the area of

Table 9. Observed and predicted responses and residual values at optimal values of factors investigated

Variable	Nominal value	Calculated values	Response	Nominal value	Expected values	Observed values	Residuals
X_1	in range	0,0094 mol/L	Y_1	max	0,889831 Pas	0,9183	–0.028469
X_2	in range	0,0300 mol/L	Y_2	10,00 Pa	11,9116 Pa	10,735	1.1766
X_3	>17%	17%	Y_3	$-100<Y_3$	–99.9802 Pa/s	–58,976	–41.0042

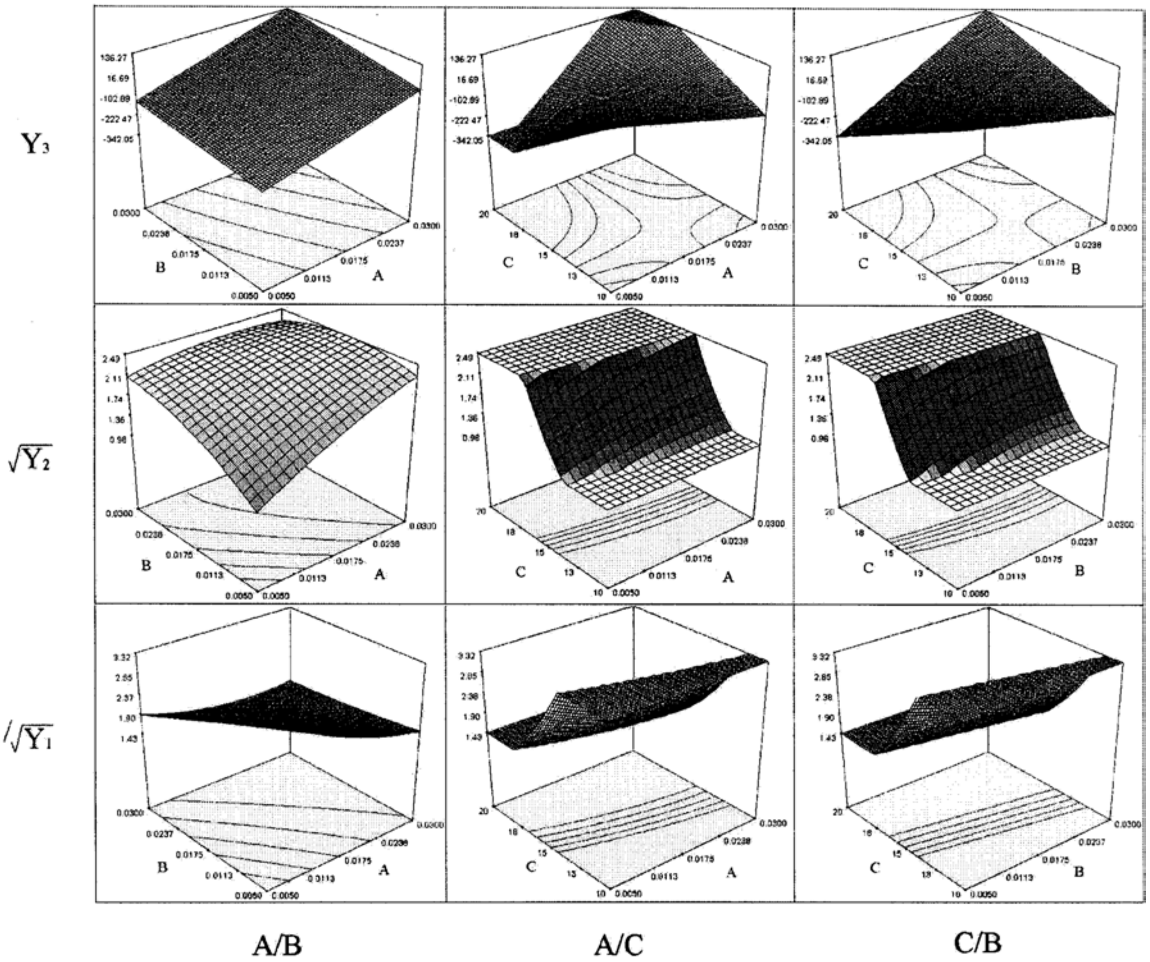

Figure 5. Response surface plots

the feasible response values in the factor space. Regions that do not fit the optimization criteria are displayed in grey colour. Any part of the design space that is not shaded satisfies the multiple constraints on the responses, and is designated yellow.

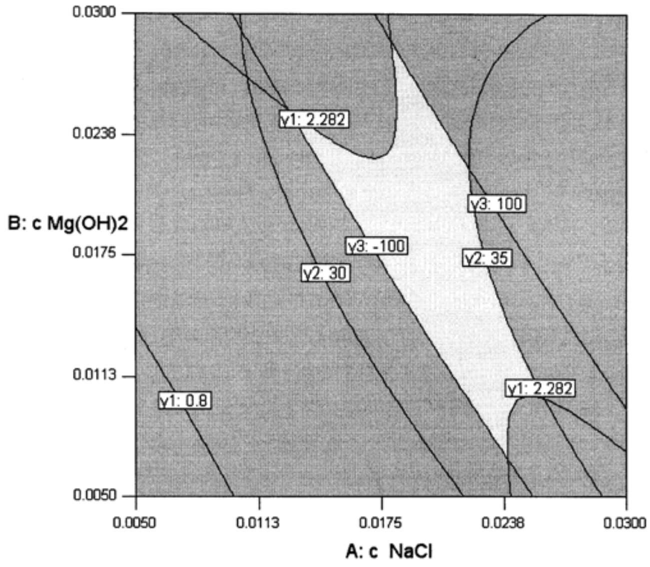

Figure 6. Graphical optimisation

The optimal design space was defined using acceptable nominal values of the responses Y_1 – Y_3 listed in Table 8. Our goal was to optimise the process for the nominal values of the responses Y_1 – Y_3 listed in Table 9. Even though the nominal value for the yield stress point was outside the optimal design space and the prediction of the responses was associated with a certain uncertainty, we found that the predicted values were in good concordance with the observed values (low residual values). The optimal calculated parameters were:

Concentration of Na^+ ions: 0.0094 mol/L
Concentration of Mg^{2+} ions: 0.0300 mol/L
Content of $AlPO_4$: 17%

From the values presented in Table 9 it can be concluded that the optimised combination of the investigated parameters can be used to obtain $AlPO_4$ of the suspension required quality.

CONCLUSION

It was shown that the rheological properties of aluminium phosphate suspensions can be successfully optimised in a way that adequate thixotropy, viscosity and yield stress point are achieved, by using response surface methodology and statistical optimisation techniques.

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