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PREPARATION OF PARAFFIN MICROSPHERES BY SUSPENSION USING PVP AS A STABILIZING AGENT

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Abstract: The present work aimed at the production of paraffin microspheres, which have the function of acting as a porogenic agent in the manufacture of porous scaffolds. The experimental tests were carried out in suspended systems. Subsequently, the influence of 3 process factors was evaluated using experimental planning techniques: A) Concentration of sodium sulfate as coagulant (3 and 9 g/L), B) Concentration of poly(vinyl pyrrolidone) (PVP) as stabilizer (4 and 12g/L) and C) Agitation (500 and 600 rpm). The best results obtained for the diameter of the paraffin particles were from 750 to 550 mm, for the 3 dependent variables d_{10}, d_{32} and d_{43}, obtained with agitation from 580 to 640 rpm (levels 0.9 to 1.2). Since to obtain particles with smaller diameters, the increase in the concentration of PVP is important together with the increase in agitation.
Keywords: Microspheres, paraffin, PVP, suspension.

INTRODUCTION

Tissue Engineering consists of the development and manipulation of cells, tissues or organs obtained synthetically to replace or support the function of damaged parts of the human body. Since, for tissue growth in complex three-dimensional form, reproducing the form and function of the tissue, it is necessary to use a support that allows cell growth, this support being called scaffold (Ma and Langer, 1995; Liu and Ma, 2004; Machado and Santos, 2009). There are studies on the production of scaffolds from PLGA (Shum et al., 2005), poly (DL-lactic-co-glycolic acid) and alpha-TCP (alpha-tricalcium phosphate) matrices using paraffin particles obtained by suspension (Machado and Santos, 2009).

Suspension systems are used to obtain micro-particles of substances derived from petroleum, which have a low melting point, such as paraffin. The use of stabilizing agents, in a suspension, has the function of preventing the coalescence of the organic droplets suspended in this phase and also giving a spherical shape, stabilizing the formed droplets (Machado et al., 2007). The agent forms a film around the paraffin droplet, having its hydrophobic part facing the droplet and hydrophilic part facing the continuous medium.

There are several analysis methods for determining the size and size distribution of a particulate solid. In this work, the methods of Mean Linear Diameter (d_{10}), Superficial Mean Diameters (d_{32}) and Volumetric Mean Diameters (d_{43}) were used for the analysis of results.

MATERIAL AND METHODS

OBTAINING PARAFFIN MICROSPHERES

To obtain paraffin in the form of microspheres, a glass container (V=1 L) containing 500 mL of deionized water was used, where different concentrations of the stabilizer poly(vinyl pyrrolidone), PVP, (Vetec) and additions sodium sulfate (Na2SO4) coagulant (Vetec). This solution was heated to 60°C and commercial paraffin (Vetec) was added and awaited for its complete melting (65°C). The system was placed under mechanical stirring for 3 min., and then rapid cooling was performed. The beads were then washed with distilled water and vacuum filtered.

After drying at room temperature, the microspheres were separated by means of sieving in different granulometric ranges. Mean diameter and dispersion were calculated for each trial.

EXPERIMENTAL PLANNING FOR PVP

Experimental planning is a tool used in

many analysis processes, formulations of new operations systems and improvement of usual operations systems. In obtaining paraffin, the influence of the following variables was evaluated: A) Sulfate concentration, B) Stabilizer concentration (PVP) and C) System agitation; with their respective levels of variation shown in Table 1. A 2^3 full factorial experimental design was adopted.

	encoded levels	
	-1	+1
conc. Sodium Sulfate (g/L)	3	9
conc. Stabilizer (g/L)	4	12
Agitation (rpm)	400	600

Table 1 - Factors and levels studied in the experimental design.

Initially, the experimental matrix to be used for screening the factors and their levels was determined. Subsequently, experiments were carried out to verify the influence of these in the paraffin granulometry (d_{10}, d_{32} and d_{43}). The results obtained were analyzed using the Statistica® software using the Response Surface Methodology (MSR) in order to obtain the optimal operating conditions.

RESULTS ANDDISCUSSION

A full 2^3 factorial design was used to assess the influence of the variables:

A) Sulfate concentration, B) Stabilizer concentration (PVP) and C) Agitation of the system, with predefined codified levels, on the cut diameter of the paraffin particles. Table 2 shows the complete planning matrix 2^3 and the responses obtained for each trial.

Rehearsal	Factors			Answers		
	conc. Sodium Sulfate (g/L)	conc. of PVP (g/L)	Agitation (rpm)	d_{10}(µm)	d_{32}(µm)	d_{43}(µm)
1	3(-1)	4(-1)	400(-1)	512,518	908,907	1059,714
two	9 (+1)	4(-1)	400(-1)	535,804	829,909	975,566
3	3(-1)	12 (+1)	400(-1)	425,830	723.010	893,304
4	9 (+1)	12 (+1)	400(-1)	432,394	755,376	927,354
5	3(-1)	4(-1)	600(+1)	339,172	547,911	735,389
6	9 (+1)	4(-1)	600(+1)	370,469	558,561	725,134
7	3(-1)	12 (+1)	600(+1)	337,934	555,702	735,237
8	9 (+1)	12 (+1)	600(+1)	345,280	575,221	787,760

*(-1) and (+1) are the levels of variation of the factors.

Table 2 - Experimental planning matrix 2^3, with their respective answers.

MEAN LINEAR DIAMETER - D10

Through the results obtained in Table 2, an analysis of the effects of the three independent variables studied on the d_{10} responses obtained in the experiments and their respective statistical indices was carried out, which is presented in Table 3.

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	It is made	Standard deviation	Student's t test	level p	-95% Confidence Limit	+95% Confidence Limit
Mean/Interactions	412.43	0.90	456.42	0.00	400.94	423.91
(A) Conc. of Sulfate (g/L)	17.12	1.81	9.47	0.07	-5.84	40.09
(B) Conc. of PVP (g/L)	-54.13	1.81	-29.95	0.02	-77.09	-31.17
(C) Agitation (rpm)	-128.42	1.81	-71.06	0.01	-151.39	-105.46
(1) AB	-10.17	1.81	-5.63	0.11	-33.13	12.79
(2) CA	2.20	1.81	1.22	0.44	-20.76	25.16
(3) BC	40.92	1.81	22.64	0.03	17.95	63.88

Table 3- Calculations of the effects and respective statistical indices in relation to the d10 response.

It can be seen from Table 3 that the main effects of PVP concentration (B) and agitation (C) and the second order interaction (BC) are statistically significant for the adopted confidence level of 95%, since the fact that the interval of confidence does not contain the number zero indicates that the factor under analysis does not have a null effect, that is, it is considered significant at the confidence level tested. Also, a value for the coefficient of determination (R^2) of 0.9998 was obtained.

Regarding the analysis of the algebraic sign of the effects found, these are in accordance with the knowledge that one has of the phenomena involved. With regard to agitation (C) an increase in this contributes to a smaller average linear diameter of the particles present at the end of the process of obtaining the paraffin microspheres. Thus, as in relation to the concentration of PVP, which acts as a stabilizer of the particles, preventing their grouping, which with its increase induces the obtaining of particles with a smaller diameter, these two variables having an indirectly proportional relationship with the response. In the case of sulfate concentration, an increase in the amount of this variable influences obtaining a larger paraffin diameter, having a directly proportional interaction with the response.

SAUTER'S MEAN DIAMETER – D^{32}

Also, through the results obtained in Table 2, an analysis of the effects of the study variables on the d32 response obtained in the experiments and their respective statistical indices, which is presented in Table 4, can also be carried out.

	It is made	Standard deviation	t testin Student	level p	-95% Confidence Limit	+95% Confidence Limit
Mean/Interactions	681.82	12.81	53.22	0.01	519.04	844.61
(A) Conc. of Sulfate (g/L)	-4.12	25.62	-0.16	0.90	-329.69	321.46
(B) Conc. of PVP (g/L)	-58.99	25.62	-2.30	0.26	-384.57	266.58
(C) Agitation (rpm)	-244.95	25.62	-9.56	0.07	-570.53	80.63
(1) AB	30.06	25.62	1.17	0.45	-295.52	355.64
(2) CA	19.20	25.62	0.75	0.59	-306.38	344.78
(3) BC	71.22	25.62	2.78	0.22	-254.36	396.80

Table 4 - Calculations of the effects and respective statistical indices in relation to the d32 response.

It can be seen from Table 4 that, through the analysis of the effects of this planning, it was verified that only the average is statistically significant for the adopted confidence level of 95%, and the other effects, as well as for the response of the cutting diameter d32 are not significant. However, the value of the coefficient of determination (R2) was 0.9906.

Regarding the main effects, the most significant value found was that of the agitation factor, as it presents a higher absolute value than the effects of other factors such as sulfate concentration and PVP concentration. For the secondary effects arising from the interactions that occur between the factors under study, it is observed that the interactions (B) and (C) showed higher absolute values, results similar to those observed for diameter d_{10}.

VOLUMETRIC AVERAGE OF DIAMETERS - D43

Also, through the results obtained in Table 2, an analysis of the effects of the study variables on the d_{43} response obtained in the experiments and their respective statistical indices, which is presented in Table 5, can also be carried out.

It can be seen from Table 5 that the main effect of agitation (C) is statistically significant for the adopted confidence level of 95%, since the fact that the confidence interval does not contain the number zero indicates that the factor under analysis does not have a null effect, that is, to be considered significant at the confidence level tested. Also, a value for the coefficient of determination (R^2) of 0.9966 was obtained.

Regarding the analysis of the algebraic sign of the effects found, these are in accordance with the knowledge that one has of the phenomena involved, as observed in d_{10} and d_{32}.

	It is made	Standard deviation	Student's t test	level p	-95% Confidence Limit	+95% Confidence Limit
Mean/Interactions	854.93	6.93	123.41	0.01	766.91	942.95
(A) Conc. of Sulfate (g/L)	-1.96	13.85	-0.14	0.91	-178.00	174.08
(B) Conc. of PVP (g/L)	-38.04	13.85	-2.75	0.22	-214.08	138.00
(C) Agitation (rpm)	-218.10	13.85	-15.74	0.04	-394.14	-42.06
(1) AB	45.24	13.85	3.27	0.19	-130.80	221.28
(2) CA	23.09	13.85	1.67	0.34	-152.95	199.13
(3) BC	69.27	13.85	5.00	0.13	-106.77	245.31

Table 5- Calculations of the effects and respective statistical indices in relation to the d43 response.

ANALYSIS OF CONTOUR LINES FOR THE 3 CUTTING DIAMETERS

Figures 1, 2 and 3 show the level curves corresponding to the response surface generated by the empirical model for diameters d_{10}, d_{32} and d_{43}.

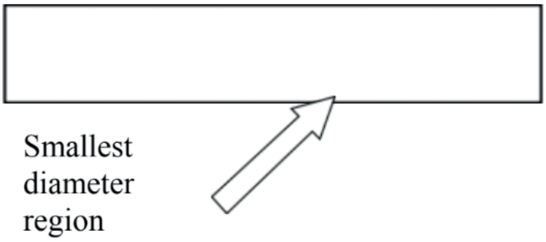

Figure 1: Contour curves for agitation and PVP concentration factors for diameter d_{10} (sulfate concentration level 0 = 6 g/L).

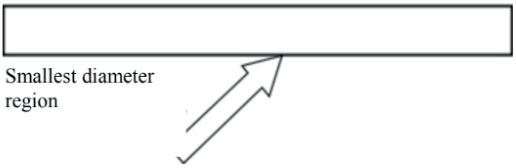

Figure 2: Contour curves for agitation and PVP concentration factors for diameter d_{32} (sulfate concentration level 0 = 6 g/L).

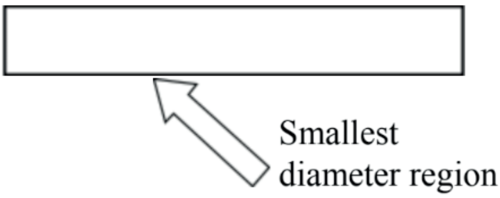

Figure 3: Contour curves for agitation and PVP concentration factors for diameter d_{43} (sulfate concentration level 0 = 6 g/L).

As shown in Figures 1, 2 and 3, it can be verified, mainly, that with the increase of the agitation of the system, the diameter of the paraffin particles, after the process for the 3 diameters d_{10}, d_{32} and d_{43}, is at a minimum level in the optimal region of process in stirring 580 to 640 rpm (levels 0.9 to 1.2). In order to obtain larger particles, the PVP concentration factor is important together with the agitation factor, which is not observed in areas with smaller particle diameters. The model for the process of obtaining paraffin for d_{10}, d_{32} and d_{43} is represented by Equations 1, 2 and 3, respectively.

$$d10 = 412.425 + 8.561\ A - 27.066\ B - 64.211\ C - 5.084\ AB + 1.099\ BC + 20.459\ BC \quad (1)$$

$$d32 = 681.825 - 2.058\ A - 29.497\ B - 122.476\ C + 15.029\ AB + 9.600\ BC + 35.610\ BC \quad (2)$$

$$d43 = 854.932 - 0.979\ A - 19.019\ B - 109.052\ C + 22.622\ AB + 11.546\ BC + 34.637\ BC \quad (3)$$

where: "A" is the concentration of sodium sulfate (g/L); B is the PVP concentration (g/L); C is the agitation (rpm); and AB, AC, and BC are the second-order interactions of factors A, B, and C.

CONCLUSIONS

The tests showed that sodium sulfate does not significantly interfere with the PVP film formed around the particles. Increasing PVP concentration and agitation parameter tended to decrease the mean particle diameter and increasing sodium sulfate concentration had little effect on the value.

The best results obtained for the paraffin particle diameter were 750 to 550 μm, for the 3 dependent variables d_{10}, d_{32} and d_{43}, obtained with agitation from 580 to 640 rpm (levels 0.9 to 1.2). The values obtained for the average linear diameters (d_{10}) and the average Sauter diameters (d_{32}) produced the highest amount of non-ideal particles to be used in the fabrication of scaffolds (50 to 125 μm) (Ítala et al., 2001). A significant increase in the PVP concentration and agitation used in the experiments is necessary to reach the desired granulometry. However, such an addition can make the system economically unfeasible.

REFERENCES

ITALA, A. I.; YLANEN H. O.; EKHOLM C.; KARLSSON K. H.; ARO H. T. Pore diameter of

more than 100 micron is not requisite for bone ingrowth in rabbits. *J. Biomed. Mater. Res*., v. 58, p. 679-683, 2001.

LIU, X.; MA, P.X. Polymeric Scaffolds for Bone Tissue Engineering. *Annals Biomed. Eng*., v. 32, p. 477–486, 2004.

MACHADO, F.; LIMA, E. L.; PINTO, J. C. Uma revisão sobre os processos de polimerização em suspensão. *Polím. Ciên. Tec*., v. 17, p. 166-179, 2007.

MACHADO, J.L.M.; SANTOS, L.A. Obtenção e utilização de microesferas de parafina para confecção de arcabouços teciduais em cimento de α-fosfato tricálcico. *Cerâmica*, v. 55, p. 216-222, 2009.

MA, P.X.; LANGER, R. Degradation, structure and properties of fibrous nonwoven poly(glycolic acid) scaffolds for tissue engineering. In: *Polymers in medicine and phar*macy. p 99–104. R. Mater. Res. Soc., PA, EUA, 1995.

SHUM, A. W. T.; LI, J.; MAK, A. F. T. Fabrication and structural characterization of porous characterization of porous biodegradable poly(DL-lactic-co-glycolic acid) scaffolds with controlled range of pore sizes. *Polym. Degrad. Stab.,* v. 87, p. 487-493, 2005.