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Research Article

Enhancing dissolution of artesunate from immediate release tablets using a green granulation technique

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ABSTRACT

Artesunate is a poorly soluble drug and liable to aqueous hydrolysis. This study aims to formulate Artesunate as an immediate release tablet through optimization of the melt granulation technique to improve the dissolution of the drug. Three different meltable binders were used (Polyethylene Glycol PEG 6000, Poloxamer 188 and Gelucire 50/13) for granulation step in high shear mixer prior tablets compression step applying Box-Behnken experimental design to determine the significant variables and their interactions that impact dissolution of Artesunate. Optimization mathematical models showed that by increasing binder concentration, D50 was increased, and narrow particle size distribution with minimum fines percentage was produced. Higher binder concentration and impeller speed resulted in retarding tablets dissolution. PEG 6000 and Poloxamer 188 based tablets showed faster disintegration and dissolution than Gelucire 50/13 based tablets, as well as tablets prepared by wet granulation due to hydrophilic pore forming. Melt granulation technique using a low level of PEG 6000 and Poloxamer 188 not only enhanced the dissolution of Artesunate from their immediate release tablets in comparison to traditional wet granulation technique but also maintained the stability of the product under accelerated conditions of heat and moisture.

Keywords: *pore-forming; crystalline; Box-Behnken design; hot melt granulation; modeling; dissolution.*

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1. INTRODUCTION

Melt granulation with no need for solvents and drying step is a successful alternative to the traditionally adopted wet granulation **[1-3]**. It is a technique by which non-meltable solids are bound together by a meltable binder through heating without any solvent. For accomplishing this process, no special equipment is required except a high shear mixer or a fluid bed granulator where the product temperature is raised above the melting point of the binder. The most prominent advantages of melt granulation are environment-friendly, improving solubility,

and hence bioavailability of poorly water-soluble drugs, saving manufacture steps, and costs, whereas the drawbacks include the non-suitability for heat-labile actives and the high energy required for the heating process **[4, 5]**. The addition of a binder to the powder mixture to be granulated can be either by keeping the binder within the mixture, and heat up all (melt-in procedure), or pouring the molten binder directly to the pre-heated powder mixture (spray-on or pump-on procedure) **[2, 6]**.

Different meltable binders can be employed in melt granulation technique either hydrophilic or hydrophobic. Hydrophilic binders are used for immediate release granules such as polyethylene glycols (PEGs) 2000–20000 **[1, 2, 4-7]**, Poloxamer 188 **[2, 8]**, and Poloxamer 407 **[6]**, esters of polyethylene glycols and polyoxylglycerides **[8, 9]**. Whereas the hydrophobic binders are mainly applied for sustained release applications including fats such as glyceryl palmitostearate **[7]**, glyceryl monostearate **[2, 10]**, and glyceryl behenate **[11]**, waxes such as beeswax, carnauba wax, microcrystalline wax, stearic acid **[12]**, cetostearyl alcohol **[13]**, and hydrogenated castor oil **[2, 11, 12]**. Many researchers investigated the process parameters, and formulation variables affecting the quality and mechanisms involved in the growth of melt granules prepared in a high shear mixer **[3, 14]**. The identified critical variables were impeller speed **[1]**, massing time **[1]**, binder content **[4, 5, 15]**, binder type **[2, 6]**, binder particle size **[7, 16]**, binder rheology **[4, 15]**, product and jacket temperature **[2, 8]**, and properties of solid non-meltable particles **[17]**. Researchers employed the melt granulation technique to enhance the dissolution of poorly water-soluble drugs by the application of hydrophilic meltable binders to improve their bioavailability and in-vivo performance **[8, 9, 18-20]**.

Artesunate, a poorly water-soluble drug (BCS class II), is commonly used as anti-malarial and very promising in the treatment of different types of cancer. Artesunate is a semi-synthetic derivative of artemisinin, a naturally occurring sesquiterpene endoperoxide which is isolated from the “quinghaosu” or sweet wormwood plant (*Artemisia annua*) and has been used worldwide for many years as anti-malarial **[21-23]**.

Artesunate exhibits a pH-dependent solubility due to the presence of a carboxylic acid group (-COOH) in its chemical structure. The main pharmaceutical issue with artesunate containing formulations is stability. Artesunate degradation is related to the main moisture, light, acidic, and basic conditions. Artesunate undergoes hydrolysis in aqueous solution and is rapidly converted to dihydroartemisinin, with a half-life of 26 min and 10 h at pH 1.2 and 7.4, respectively at room temperature **[24]**. Therefore, a solvent-free granulation technique is preferred over the traditional wet granulation.

At the best of authors’ knowledge, this is the first contribution in artesunate tablet formulation development via melt granulation technique in which no solvents are used hence the drying step will not be required, few research papers discussed artesunate tablets development. However, some authors conducted research to improve artesunate dissolution from tablets by applying other techniques as solid dispersion and cyclodextrin complexation **[25, 26]**.

The aim of this study is to optimize melt granulation process as an alternative technique to the traditional wet granulation process and define the best combination of formulation and process variables that lead to best tablets quality attributes with focus on dissolution enhancement of artesunate from its immediate release tablets.

2. MATERIALS AND METHODS

2.1. Materials

Artesunate (IPCA, India) was used as a model drug, microcrystalline cellulose (Avicel®PH101, FMC Corporation, DE, USA) was used as extra-granular compression aid, Croscarmellose sodium (Amishi drugs and chemicals, India) was used as disintegrant, Lactose monohydrate 200 mesh (Pharmatose®200M, DFE-Pharma, Veghel, Netherlands) was used as diluent, meltable binders were PEG 6000 (Ineos, Switzerland) in flakes form with melting range (55–63 °C), Gelucire® 50/13 which is a mixture of glycerides and PEG esters of fatty acids (Gattefossé, France) in pellets form with melting range (46–51 °C), Poloxamer 188 (Kolliphor®P 188, BASF Corporation, Germany) in spherical pellets form with melting range (52–57 °C), Magnesium stearate (Kemilub® EM-F-V, Union Derivan, Spain) was used as a tablet lubricant, and Aerosil 200 (Wacker chemie, Germany) was used as a granular flow enhancer.

2.2. Methods

2.2.1. Binary mixture stability study

A 3-months artesunate-excipient compatibility screening study was carried out in ratios of drug to excipient (1:1) for the main fillers lactose and avicel; and (4:1) for the remaining excipients: croscarmellose sodium, magnesium stearate, and aerosil. Samples of binary mixtures were mixed and filled in amber colored sealed vials with a rubber stopper and closed with aluminum caps then stored at 40 °C. Samples of these mixtures were withdrawn and checked for any physical change and analyzed for artesunate content using HPLC, at time intervals of 1, 2 and 3 months.

2.2.2. Preparation of granules

Melt granulation of a mixture of artesunate, lactose, and croscarmellose sodium was carried out in (Rotolab®, Zanchetta, Lucca, Italy), which is 1.8 liters laboratory scale high shear mixer equipped with an electrically heated jacket, three blades impeller, a top drove chopper, and a product temperature sensor. The batch size of each trial was 310 g without a binder (150 g artesunate, 150 g lactose, and 10 g croscarmellose sodium). The process consisted of three steps: mixing, granulation, and cooling. Powders were discharged manually into granulator bowl, then mixed at 120 rpm, then the heatable jacket was switched on, and the temperature was set to 10-15 °C higher than the melting point of the employed binder which was added in an amount corresponding to 7, 11 and 15% of the batch size 310 g. Powder mixture temperature was continuously monitored by the probe which was fixed to the bowl lid and dipped into the powder mass. The binder amount was added once the mixture temperature became 5 °C below the binder melting point **[1]**. Preliminary runs were attempted to investigate the feasibility of carrying out the trials without failure or limitations by challenging the border levels of the selected independent variables, with focus on binder concentration using PEG 6000 as a meltable binder. These runs supported the choice of upper and lower binder concentration levels, their results were mentioned later in the results and discussion section. A melt-in method as a binder addition technique was preferred to avoid the flaws of the pour-on method, and the hot molten binder handling and application [3]. Once the temperature of the mixture reached the binder melting point, the granulation step started at the defined speed and time for each run without the chopper. The cooling step was performed by discharging the hot granules manually onto a stainless steel tray at room temperature. The cooled granules were pressed manually through 1000 µ sieve for de-agglomeration of lumps and oversized granules. A wet granulation trial was carried out using 40 g absolute alcohol as solvent

(water was not used to avoid artesunate hydrolysis), powder mixture included 150 g artesunate, 150 g lactose, 10 g croscarmellose sodium, and 46.5 g milled PEG 6000 (mixed as fine powder) at 15% of the total powder mixture. Granulation was performed by three steps, a premix of powder at 120 rpm for 2 min, followed by alcohol addition within 2 min at 200 rpm, then final wet massing at 600 rpm for 2 min. Wet granules were discharged manually onto a stainless steel tray and dried in an oven at 50 °C till loss on drying of a sample reached 2.5% at 120 °C. Dried granules were screened through 1000 micron sieve for de-agglomeration of big sized granules. The aim of the wet granulation trial was to compare the wet granules dissolution results to those of melt granules.

2.2.3. Experimental design

Three sets of experiments were carried out by employing three different binders as a formulation variable (Polyethylene glycol 6000, Poloxamer 188 and Gelucire 50/13). STAVEX®5.2 software (Aicos, Switzerland) was used to generate a Box-Behnken design. Each set of experiments included 13 runs comprised of 3 independent variables (binder concentration, impeller speed and granulation time) with 3 levels as shown in **table 1**. The dependant factors under study were particle size distribution (D10, D50, D90, span value, and fine%), granules bulk and tapped densities, granules flowability (Carr's index and Hausner ratio), dissolution of both granules and tablets at different time points 5,10,15,30 and 45 min.

Table 1. Box-Behnken design of the process and formulation variables.

Run no.	Binder concentration (%)	Impeller speed (rpm)	Granulation time (min.)
1	7	300	6
2	15	300	6
3	7	900	6
4	15	900	6
5	7	600	3
6	15	600	3
7	7	600	9
8	15	600	9
9	11	300	3
10	11	900	3
11	11	300	9
12	11	900	9
13	11	600	6

2.2.4. Characterization of the prepared granules

2.2.4.1. Particle size distribution

The distribution of the granules size was evaluated by a dry method using Mastersizer 2000 (Malvern Instruments, Worcestershire, UK) as an average of three measurements for each

granulation sample. Results were presented as D10, D50, D90, and span (D90-D10)/D50.

2.2.4.2. Determination of true density

The true density was determined by a gas displacement pycnometer (model Ultrapycnometer 1000, Quantachrome Instruments, USA). Density calculated was the mean of three measurements for each sample.

2.2.4.3. Determination of Carr's index and Hausner ratio

The granules were weighed and poured into a 50 mL graduated cylinder. The bulk densities of granules were calculated before tapping the cylinder manually until no more volume decreasing was noticed, and then the tapped densities were calculated. The bulk and tapped densities were obtained as an average of triplicate measurements for each granulation sample, and were used to calculate the Hausner ratio and Carr's index by applying the following equations:

$$\text{Compressibility (Carr's) index} = \frac{\text{tapped density-bulk density}}{\text{tapped density}} \text{ X } 100$$

$$\text{Hausner ratio} = \frac{\text{tapped density}}{\text{bulk density}}$$

Granules are judged according to the scale of flowability mentioned in USP pharmacopeia **[27]**.

2.2.4.4. Determination of flow rate

The flow rate of granules was evaluated using the automatic powder flow tester (model PTG-S4, Pharma Test, Hainburg, Germany); and the results were obtained as an average of three measurements.

2.2.4.5. Determination of artesunate assay in granules

The test was carried out by dispersing a known amount of granules equivalent to 100 mg artesunate into methanol in 100 mL amber-colored volumetric flask. The preparation was shaken and filtered; the filtrate was analyzed using WATERS HPLC with UV detection at 210 nm, the column was Thermo HYPERSIL BDS C18 (150 x 4.6 mm, 5 µm), and the mobile phase was 50% acetate buffer pH 5.5, 30% acetonitrile and 20 % methanol using 1.5 ml/min flow rate and 50 µl sample injection volume while column temperature was 30 °C, against a standard preparation of 100 mg artesunate dissolved into 100 mL methanol. Assay results of each granulate was an average of two tests.

2.2.4.6. Determination of in vitro dissolution

The test was performed using a dissolution tester (model VK7010, Varian Inc., USA) equipped with an autosampler (model VK8000). The applied test parameters were apparatus II rotating at 100 rpm, granules were dispersed into 900 mL 0.1 M sodium acetate buffer prepared by dissolving 82.0 g sodium acetate anhydrous into 10 L of distilled water, then adding few drops of glacial acetic acid to adjust pH to 5.5, the temperature was maintained at 37±0.1 °C. Dissolution media samples were withdrawn at predetermined time points of 5, 10, 15, 30, and 45 min. Withdrawn samples were filtered and analyzed using HPLC, the analytical parameters were Hypersil BDS C18, 5 µm 150 X 4.6 mm under 30 °C column temperature, mobile phase 0.1 M sodium acetate buffer (pH= 5.5) to acetonitrile to methanol (50:30:20%,v/v) with flow rate 1.5 mL/min, the injection volume was 50 µL, and UV detection at 210 nm **[24]**. The same procedure was applied for the determination

of artesunate dissolution from their corresponding tablets.

2.2.4.7. Fourier transform infrared spectroscopy (FTIR)

The FTIR spectra were obtained on an FTIR spectrometer (Nicolet iS10, Thermo Scientific, USA) over the range 400 - 4000 cm^{-1}.

2.2.4.8. X-ray powder diffraction

Powder diffraction patterns were recorded on an X-ray diffractometer (XPERT-PRO, PANalytical, Netherlands, Holand) with Cu as tube anode; the diffractograms of API, physical mixtures, and melt granules were recorded under the following conditions: voltage 40 kV, 35 mA, angular range 5, and fixed divergence slit.

2.2.4.9. Scanning electron microscopy (SEM)

The shape and surface of melt granules fraction (250-355µm), as well as pure artesunate powder, were examined using scanning electron microscope (Model Quanta 250 FEG Field Emission Gun, FEI company, Netherlands), with accelerating voltage 30 KV. Samples were sputter-coated with gold using a vacuum evaporator (Emitech, England) before the examination.

2.2.4.10. Differential scanning calorimetry (DSC)

DSC analysis of artesunate, excipients, and the produced granules were performed using differential scanning calorimeter (DSC 4000, PerkinElmer Corp., Waltham, MA, USA). Accurately weighed samples (3–7 mg) were placed in manually crimped aluminum pans scanned at a heating rate of (10 °C /min) over a temperature range of 30-220 °C in nitrogen atmosphere.

2.2.5. Compression of granules

Melt granules were compressed into tablets after the addition of 5% croscarmellose sodium as an extragranular disintegrant followed by 1% magnesium stearate as a lubricant for all runs. A rotary tablet press (model XL-100, KORSCH, Berlin, Germany), equipped with only two sets of 10 mm round flat punches, was employed to obtain tablets containing 100 mg Artesunate with an average weight of 300 mg. The main compression force was 11.5±1.0 KN, turret speed was 25 rpm and edge thickness was fixed to 1.5 mm. The amount of extragranular avicel was varied according to the variable granules weight, which was directly related to the used binder concentration level in order to obtain a constant tablet weight containing 100 mg artesunate per tablet as shown in **table 2**.

Table 2. Composition of artesunate (100 mg) tablets with three different binder concentrations.

Binder concentration	7%	11%	15%
Each tablet contains:			
Granules containing 100 mg artesunate	221.13 mg	229.40 mg	237.67 mg
Avicel PH 101	60.87 mg	52.6 mg	44.33 mg
Croscarmellose sodium	15.0 mg	15.0 mg	15.0 mg
Magnesium stearate	3.0 mg	3.0 mg	3.0 mg
Total	300 mg	300 mg	300 mg

2.2.6. Tablets properties

The friability of 10 tablets from each run was determined using a friabilator (model PTF-20E, Pharma Test, Hainburg, Germany) rotating 100 times. The crushing strength of ten tablets from each run was measured with a hardness tester (model 8 M, Pharmatron AG, Switzerland).The disintegration time of six tablets from each run was tested using the disintegration test apparatus (model ZT 220, Erweka, Germany). Water at 37 °C was used as a medium, and the basket was raised and lowered at a constant frequency of 30 ± 1 strokes/min. *In vitro* dissolution and assay test were also carried out for tablets in the same way as granules.

2.2.7. Stability testing of artesunate tablets

Tablets of single run from each binder set of trials were packed in Alu/Alu cold forming blisters, then stored under ambient (30 °C /65% RH), and accelerated (40 °C /75% RH) conditions for 3 and 6 months to be tested later for drug assay and dissolution.

3. RESULTS AND DISCUSSION

3.1. Preliminary granulation study

The preliminary trials of melt granulation using PEG 6000 as meltable binder at concentrations of 5% and 20% at 900 rpm for 9 min with the application of the same procedure mentioned in section of preparation of granules showed very fine granules and dusty mixture after cooling step indicating that 5% is insufficient binder quantity to consider as a binder concentration low level in the statistical design of experiment. On the other hand, 20% binder showed severe overwetting and very large balls formation supporting the selection of a lower concentration as the high level in the design.

Based on the findings, 7% and 15% concentrations of PEG 6000 were tested using the same procedure and resulted in reasonable quality granules with no physical limitations encouraging the selection of these percentages as the upper and lower levels of binder concentration.

3.2. Binary mixture stability study

Table 3 shows three consecutive months of assay results of binary mixtures of excipients with artesunate. There were no signs of any physical change or significant chemical degradation.

Table 3. Assay of Artesunate content in binary mixtures stored at 40 ^{o}C/30% RH.

	Assay (%) of Artesunate content after		
Sample name	**1st month**	**2nd month**	**3rd month**
Artesunate only	98.73	99.01	98.98
Artesunate: Lactose monohydrate	94.11	95.83	95.77
Artesunate: Avicel 101	101.29	100.66	101.4
Artesunate: Croscarmellose sodium	97.67	99.65	98.86
Artesunate: Magnesium stearate	99.83	100.66	99.38
Artesunate: Aerosil 200	100.07	101.45	98.87

3.3. Particle size distribution

The particle size distribution of all melt granules was measured and compared and presented in **table 4**. D10 is the particle size which 10% of particles exist below it, D50 is the particle size which 50% of particles exist below it, while D90 is the particle size which 90% of particles exist below it. Span is the width of particles distribution and calculated by using D10, D50 and D90 terms, which is the difference between D10 and D90 divided by D50, low span values indicates narrow particle size distribution and vice versa. D50 was the smallest for Gelucire 50/13 (125-621 µm) and the largest for Poloxamer 188 (183-946 µm). Poloxamer 188 granules showed lower percentages of fines and larger D10 values than both Gelucire 50/13 and PEG 6000. Regardless of binder type, high binder concentration (15%) showed granules with narrow particle size distribution (low span values), noticeable high D10, D50 values and less fines percentage, while low binder concentration (7%) showed granules with wide particle size distribution (high span values), noticeable low D10, D50 values, and more fines percentage. This finding is in agreement with what was reported in the literature **[7, 19]**. Masic et al. found that increasing binder concentration of PEG 2000, and Precirol ATO 5 up to 15% significantly reduced particle size distribution and controlled fines fraction. Also, Perissutti et al. reported that the mean geometric diameter increased as the PEG content increased. This can be explained by the abundance of liquid droplets of the meltable binder at high concentration that allows more particles fusion and granular growth. **Table 5** shows the suggested models for D10, D50, D90, span, and fines percentage for all binders. Binder concentration was a common significant variable ($P<0.05$) affecting nearly all particle size terms of the three used binders. Regarding PEG 6000, by increasing binder concentration, D50 and D90 were increased with good ($R^2= 0.9574$), and mediocre ($R^2= 0.8986$) model fitting respectively, while fines % and span values were decreased which emphasizes a good granulation efficiency. Similar results were found for Poloxamer 188, whereby increasing binder concentration, D50 was increased with very good model fitting ($R^2= 0.9809$), span and fines % values were decreased. Also for Gelucire 50/13, by increasing the binder concentration, D10, D50, and D90 values were increased significantly with good model fitting. The values of D50 for Gelucire granules were not only increased by binder concentration but also did by increasing both impeller speed and granulation time mainly at high binder concentration, **Fig.1**. It was reported that the yield of 2 mm size fraction of paracetamol pellets prepared using stearic acid as a binder was very sensitive to small variations of impeller speed and massing time **[28]**.

3.4. Determination of True density

Results of true density are ranging from 1.353 to 1.52 g/mL as shown in **table 6**. Generally, melt-granulation yields denser granules compared with wet granulation due to the remaining of the binder liquid into granules structure, whereas the binder solvent is evaporated in case of wet granulation **[5, 17]**.

3.5. Determination of Carr's index and flow rate of granules

As shown in **table 7**, Carr's index values of PEG 6000 granules ranged from (11.35 – 28.1%) indicating flow character from good to poor, Poloxamer 188 granules ranged from (10.41-29.6%) indicating flow character ranged from good to poor, and Gelucire 50/13 granules ranged from (15.54-24.57%) indicating flow character from good to passable according to mentioned ranges in scale of flowability in USP. Hausner ratio values of PEG 6000 granules ranged from (1.13-1.39), Poloxamer 188 granules (1.12-1.42),

and Gelucire 50/13 granules (1.18-1.33) indicating the same flow character conclusion deduced from carr's index values, flow rate results ranged from (1.9-3.6 seconds for 20 g of granules). All melt granules exhibited good apparent flowability before and after addition of extragranular excipients. Regardless of the binder type or concentration, no glidant was needed to improve granules flow upon addition and mixing of extragranular excipients; powder mixtures filled the dies evenly. Statistical analysis showed that in PEG 6000 trials binder concentration was the sole significant factor (p= 0.0366), reducing Carr's index values was achievable by increasing binder concentration, the number of fine particles was reduced due to higher agglomeration of particles, hence better flow, and confirmed good flow behavior of granules.

Table 4. Particle size distribution of melt granules.

	PEG 6000 (mean ± SD)					Poloxamer 188 (mean ± SD)					Gelucire 50/13 (mean ± SD)				
Run No.	**D10**	**D50**	**D90**	**Span**	**Fines (%)**	**D10**	**D50**	**D90**	**Span**	**Fines (%)**	**D10**	**D50**	**D90**	**Span**	**Fines (%)**
1	45±3	241±10	1189±48	4.7±0.31	28.6±2.52	55±5	220±15	1069±34	4.7±0.21	28.5±2.21	33±3	125±14	428±28	3.2±0.32	43.5±3.11
2	160±17	665±84	1390±36	1.8±0.22	3.7±0.86	266±13	684±41	1326±55	1.5±0.40	0.2±zero	156±6	407±20	930±41	1.9±0.31	6.4±2.04
3	49±2	173±13	1140±50	6.3±0.41	32.1±1.81	66±4	183±22	987±31	5.0±1.11	26.3±2.51	38±2	138±8	389±17	2.5±0.12	39.1±1.63
4	90±5	746±23	1449±26	1.8±0.30	11.4±1.31	572±19	946±33	1502±40	1.0±0.30	0.4±0.12	333±9	621±16	1071±23	1.2±0.20	2.8±0.82
5	38±4	139±25	1049±55	7.3±0.54	40.3±2.50	54±4	199±13	1065±24	5.1±1.62	30.0±2.12	35±3	130±10	460±12	3.3±0.31	42.0±1.71
6	84±6	390±16	1184±28	2.8±0.22	12.6±1.51	276±10	631±17	1245±19	1.5±0.10	0.8±0.21	148±2	411±13	836±19	1.7±0.12	7.0±1.33
7	46±5	159±12	1147±35	6.9±0.20	35.2±1.43	64±2	194±9	1105±11	5.4±0.21	26.6±1.40	38±2	145±6	513±13	3.3±0.22	38.0±1.22
8	106±8	670±26	1366±21	1.9±0.11	9.9±1.90	133±8	729±21	1390±23	1.7±0.13	7.8±1.61	192±10	544±15	1030±33	1.5±0.13	5.7±1.80
9	56±6	281±14	1293±34	4.4±0.71	24.4±2.1	121±3	484±29	1247±52	2.3±0.24	6.9±1.03	58±2	192±9	805±21	3.9±0.40	24.8±2.11
10	90±3	307±10	1222±19	3.7±0.32	13.6±1.71	156±5	352±11	888±21	2.1±0.12	7.8±1.12	68±3	244±11	510±19	1.8±0.12	17.7±1.22
11	56±2	200±9	1143±22	5.4±0.53	27.6±2.22	130±8	390±12	1189±21	2.7±0.10	4.8±0.81	62±4	191±7	477±10	2.2±0.20	23.1±0.70
12	102±6	414±15	1319±20	2.9±0.22	10.6±1.33	319±12	555±23	961±34	1.2±0.22	0.0±zero	135±10	336±15	612±29	1.4±0.50	7.5±1.12
13	72±6	218±12	1109±29	4.8±0.91	20.2±2.12	161±7	446±13	1245±33	2.4±0.41	2.0±0.51	70±5	261±17	638±28	2.2±0.32	16.6±1.31

* D10, D50, and D90 are displayed in micrometers (µm).
* *Fines % are below 105 µm*

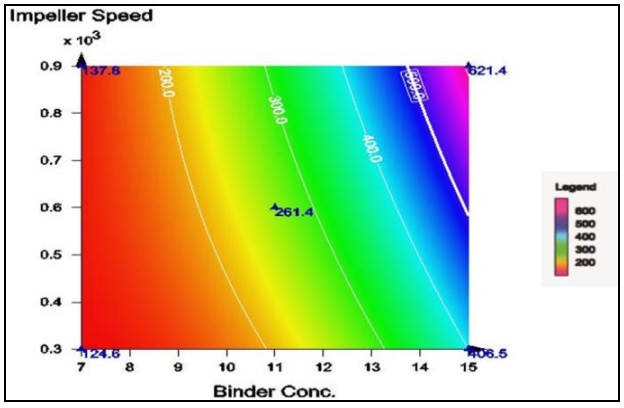

Fig. 1. A contour plot showing the effect of binder concentration and impeller speed on D50 of Gelucire 50/13 melt granules.

Table 5. Models suggested for particle size distribution terms showing the independent variables: binder concentration (B), impeller speed (S), granulation time (T), and their interactions.

Binder trial	Equation	Goodness of fit	
		R^2	R^2 corrected
PEG 6000	**D10** = -64.28 + 11.41 B + 0.01263 S + 5.947 T + 0.2019 B^2+ 0.0001174 S^2- 0.7716 T^2 - 0.01537 BS+ 0.2671 BT+ 0.003522 ST	0.7839	0.1355
	D50 = 1602 - 187.4 B - 1.878 S - 54.82 T + 8.688 B^2+ 0.001105 S^2- 1.886 T^2 +	0.9574	0.8295
	D90 = 2255 - 83.37 B - 2.220 S - 70.99 T+ 3.920 B^2+ 0.001337 S^2+ 1.639 T^2+ 0.02260 BS + 1.764 BT + 0.06896 ST	0.8986	0.5943
	Span = 2.051+ 0.07569 B+ 0.01713 S+ 0.1298 T - 0.01495 B^2 - 9.292E-06 S^2+ 0.02178 T^2- 0.000335 BS - 0.01219 BT - 0.0004975 ST	0.9156	0.6623
	Fines =74.18 - 6.796 B + 0.03864 S- 2.741 T+ 0.1313 B^2- 3.713E-05 S^2+ 0.2435 T^2 + 0.0008708 BS + 0.05062 BT - 0.001761 ST	0.8683	0.4731
Poloxamer 188	**D10** = 212.8 - 6.202 B -1.568 S+ 68.92 T+ 0.9068 B^2+ 0.0007173 S^2- 4.819 T^2+	0.8618	0.4472
	D50 = 635.3 - 20.09 B - 1.537 S - 17.6 T + 1.743 B^2+ 0.000386 S^2- 3.930 T^2 + 0.06234 BS + 2.155 BT + 0.08251 ST	0.9809	0.9237
	D90 = 1233 - 79.94 B + 0.009489 S + 90.84 T+ 3.326 B^2- 0.0008554 S^2- 10.71 T^2+ 0.0538 BS + 2.188 BT + 0.03663 ST	0.8085	0.234
	Span =11.36 - 1.701 B + 0.008078 S + 0.2481 T + 0.06245 B^2- 3.967E-06 S^2 - 0.0005278 T^2 - 0.0001815 BS - 0.002333 BT - 0.0003636 ST	0.9864	0.9456
	Fines =150.3 - 20.65 B+ 0.0004729 S- 5.350 T+ 0.7245 B^2+ 2.125E-06 S^2+ 0.3051 T^2+ 0.0003896 BS+ 0.2165 BT- 0.001592 ST	0.9839	0.9357
Gelucire 50/13	**D10** = 488.2 - 68.87 B - 0.7092 S + 2.415 T + 2.899 B^2+ 0.0002659 S^2- 1.43 T^2+ 0.03587 BS+ 0.829 BT+ 0.01765 ST	0.9543	0.8173
	D50 =530.2 - 82.6 B - 0.403 S - 8.339 T + 3.995 B^2- 3.075E-05 S^2- 2.013 T^2 + 0.04203 BS + 2.449 BT + 0.02579 ST	0.9956	0.9824
	D90 = 1321 - 95.95 B - 0.8745 S - 81.77 T+ 5.482 B^2- 0.0002362 S^2- 1.782 T^2+ 0.03782 BS + 2.932 BT + 0.1196 ST	0.9307	0.7229
	Span =8.025 - 0.2615 B - 0.003150 S - 0.5462 T + 0.004633 B^2- 5.069E-07 S^2+ 0.02179 T^2 - 1.792E-05 BS - 0.002729 BT + 0.0003689 ST	0.9044	0.6175
	Fines =122.8 - 12.6 B - 0.01066 S - 1.206 T + 0.353 B^2+ 8.472E-06 S^2+ 0.1069 T^2+ 0.0001583 BS+ 0.05667 BT - 0.00235 ST	0.9867	0.9466

**Significant factors with (P<0.05) are underlined in the mathematical models.*

Table 6. True density of PEG 6000, Poloxamer 188, and Gelucire 50/13 granules

Run No.	Binder conc. (%)	PEG6000 (mean ± SD)	Poloxamer 188(mean ± SD)	Gelucire 50/13 (mean ± SD)
1	7.0	1.439± 0.0008	1.433± 0.0020	1.408± 0.0027
2	15.0	1.431± 0.0022	1.353± 0.0003	1.374± 0.0023
3	7.0	1.468± 0.0010	1.406± 0.0012	1.411± 0.0010
4	15.0	1.416± 0.0005	1.371± 0.0004	1.382± 0.0015
5	7.0	1.520± 0.0006	1.415± 0.0007	1.418± 0.0012
6	15.0	1.461± 0.0007	1.376± 0.0003	1.383± 0.0035
7	7.0	1.482± 0.0014	1.406± 0.0001	1.408± 0.0005
8	15.0	1.501± 0.0052	1.388± 0.0025	1.384± 0.0004
9	11.0	1.465± 0.0004	1.405± 0.0011	1.400± 0.0038
10	11.0	1.450± 0.0007	1.394± 0.0019	1.399± 0.0005
11	11.0	1.468± 0.0005	1.397± 0.0003	1.397± 0.0003
12	11.0	1.458± 0.0005	1.388± 0.0010	1.411± 0.0061
13	11.0	1.469± 0.0005	1.388± 0.0015	1.383± 0.0011

Table 7. Flowability results of all granules' types expressed as Carr's index, Hausner ratio, and flow rate values

	PEG 6000			Poloxamer 188			Gelucire 50/13		
Run no.	Carr's Index (mean ± SD)	Hausner Ratio (mean ± SD)	Flow rate (sec./20 g) (mean ± SD)	Carr's Index (mean ± SD)	Hausner Ratio (mean ± SD)	Flow rate (sec./20 g) (mean ± SD)	Carr's Index (mean ± SD)	Hausner Ratio (mean ± SD)	Flow rate (sec./20 g) (mean ± SD)
1	23.69±2.12	1.31±0.03	2.80±0.12	23.50±2.21	1.31±0.03	3.50±0.62	24.57±3.21	1.33±0.02	3.60±0.23
2	21.14±1.71	1.27±0.02	2.20±0.00	18.42±1.42	1.23±0.02	2.10±0.00	16.20±2.00	1.19±0.01	2.20±0.06
3	23.11±1.60	1.30±0.02	2.50±0.06	23.26±2.03	1.30±0.03	2.70±0.12	18.15±2.12	1.22±0.03	3.10±0.06
4	11.35±3.42	1.13±0.04	1.90±0.00	10.41±0.52	1.12±0.00	2.00±0.06	15.54±1.13	1.18±0.01	2.30±0.06
5	28.10±2.03	1.39±0.04	3.20±0.22	13.68±1.20	1.16±0.01	3.50±0.35	21.14±2.41	1.27±0.02	3.60±0.11
6	19.55±2.54	1.24±0.04	2.10±0.11	24.66±3.10	1.33±0.02	2.30±0.15	19.29±1.72	1.24±0.02	2.40±0.06
7	23.14±1.30	1.30±0.03	2.60±0.02	25.96±2.61	1.35±0.03	3.30±0.46	20.56±2.63	1.26±0.03	3.40±0.72
8	19.41±0.16	1.24±zero	2.30±0.63	25.06±3.11	1.33±0.04	2.30±0.06	15.76±1.21	1.19±0.02	2.20±0.06
9	20.24±1.70	1.25±0.03	2.80±0.23	29.26±2.33	1.41±0.03	2.30±0.06	19.74±0.83	1.25±0.01	2.50±0.12
10	19.81±2.41	1.25±0.04	2.20±0.00	20.92±1.70	1.26±0.01	2.20±0.06	18.11±1.30	1.22±0.02	2.50±0.11
11	21.22±2.82	1.27±0.04	2.40±0.06	29.61±2.90	1.42±0.03	2.10±0.00	18.38±2.22	1.23±0.03	2.30±0.06
12	21.38±2.40	1.27±0.04	2.10±0.06	18.62±0.92	1.23±0.01	2.20±0.06	22.20±1.91	1.29±0.02	2.40±0.06
13	23.26±1.61	1.30±0.02	2.20±0.00	18.19±1.70	1.22±0.02	2.50±0.14	24.00±1.73	1.32±0.02	2.40±0.06

3.6. Determination of artesunate assay in granules

All assay results were between (90-110%) indicating good content uniformity of artesunate in all granules regardless binder type as shown in **table 8**. Results emphasized the efficiency of the high shear mixer to maintain the homogeneity of powder mixture in a melt granulation process **[3]**.

Table 8. Assay results of Artesunate in melt granules.

Run No.	PEG 6000 (mean ± SD)	Poloxamer 188 (mean ± SD)	Gelucire 50/13 (mean ± SD)
1	92.7 ± 2.5	100.4 ± 0.8	99.9 ± 1.2
2	99.6 ± 1.9	100.9 ± 1.5	95.2 ± 2.1
3	97.5 ± 1.1	97.6 ± 1.0	99.7 ± 0.9
4	97.1 ± 2.0	99.6 ± 2	103.7 ± 2.3
5	101.7 ± 1.5	99.1 ± 0.7	102.3 ± 1.9
6	98.7 ± 2.1	102.5 ± 2.1	101.5 ± 1.6
7	91.8 ± 1.6	99.6 ± 0.8	100.9 ± 3.1
8	93.9 ± 0.6	100.2 ± 1.8	101.6 ± 1.0
9	93.0 ± 1.0	101.4 ± 3.1	97.8 ± 1.6
10	95.3 ± 2.6	100.6 ± 1.4	99.1 ± 2.6
11	94.6 ± 1.2	104.9 ± 2.2	100.3 ± 0.9
12	94.3 ± 1.8	108.6 ± 1.1	100.0 ± 0.5
13	98.5 ± 0.9	107.9 ± 1.9	101.8 ± 1.3

3.7. FTIR analysis

FTIR analysis was performed to determine if there is any interaction between the granules components. **Fig. 2** shows the spectra of Artesunate, lactose monohydrate, binders, and the three different melt granules. FTIR spectrum of pure crystalline Artesunate powder shows a characteristic peak at 3278 cm^{-1} due to (O-H stretching vibrations), a broad peak of (C-H aliphatic stretching vibrations) in a range 2860-2960 cm^{-1}, and a distinct strong sharp peak of (C=O stretching vibrations) at 1756cm^{-1} **[22]**. Characteristic peaks of artesunate, binders, and lactose are evident in the FTIR spectra of all melt granules, suggesting the lack of solid state interactions.

3.8. X-ray powder diffraction

XRPD was performed to detect any change in the crystallinity of artesunate after melt granulation. **Fig. 3** shows the diffraction pattern of pure artesunate, lactose monohydrate, a physical mixture of (PEG 6000, lactose, and artesunate), and the melt granules. Artesunate is a crystalline material as demonstrated by multiple sharp and intense diffraction peaks. Melt granules diffraction peaks of all binders did not show a significant reduction in crystallinity of artesunate which was maintained in both granules and physical mixture as well. The low intensity of artesunate diffractions peaks in melt granules and physical mixtures may be attributed to the dilution of the API in the powder mixture. Some researchers reported that melt granulation changed crystalline drugs into an amorphous

structure with improved solubility such as Ibuprofen, Ketoprofen as well as solid dispersions formulations of Artemether **[3, 29, 30]**. However, others reported the absence of solid state modifications of Praziquantel within melt granules **[8]**. In another study, there was no change of Griseofulvin crystallinity, and it was stated that the dissolution improvement of the drug was due to the hydrophilic character of the water-soluble binders **[9]**. Diazepam dissolution was improved via melt granulation due to the formation of the carrier controlled system in which the dissolution rate was dependent only on the dissolution rate of carrier **[20]**. Our findings are in accordance with the studies where drug crystallinity was not changed hence mitigating any stability concerns of amorphous conversion. On the other hand, a high dissolution rate from the hot melt granules was shown as will be presented later.

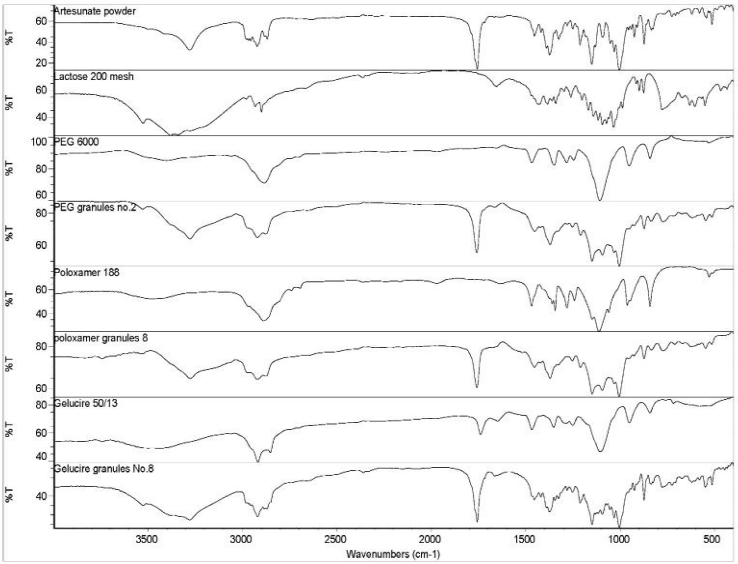

Fig. 2. Comparison of FTIR spectra of Artesunate powder, lactose monohydrate, PEG 600, Poloxamer 188, Gelucire 50/13, and their corresponding granules at 15% binder concentration.

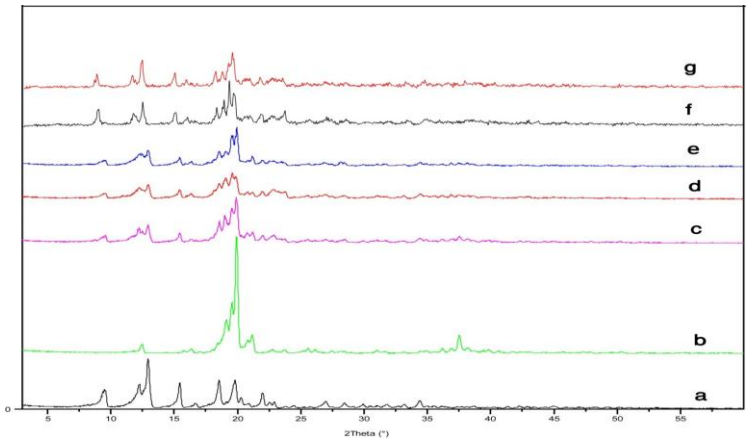

Fig. 3. X-Ray Diffractograms showing a) Artesunate b) Lactose monohydrate c) Artesunate, Lactose monohydrate, and PEG 6000 physical mixture d) 15% PEG 6000 granules e) 7% PEG 6000 granules f) 15% Gelucire 50/13 granules g) 15% Poloxamer 188 granules.

3.9. Scanning Electron Microscopy

Fig. 4 shows SEM micrographs that depict Artesunate powder as irregular particles with agglomerates in different sizes. Both PEG 6000 and Poloxamer 188 granules were shown as relatively spherical particles with a smooth surface, whereas Gelucire 50/13 granules seemed also spherical but with a rough and irregular surface, This may be attributed to better solubility or dispersion of artesunate particles in PEG 6000 and Poloxamer 188 than Gelucire 50/13. This seems to affect dissolution result as shown later under the dissolution section.

3.10. Differential scanning calorimetry

Fig. 5 shows DSC thermogram of artesunate exhibiting a characteristic sharp endothermic peak corresponding to melting transition at 144.43 °C, directly followed by a broad exothermic peak corresponding to recrystallization at 159.32 °C, followed by degradation of the molecule after 190 °C. Lactose monohydrate thermogram depicts a dehydration endothermic peak at about 140 °C **[2]**, which coincide with the melting peak of artesunate. However, the broad recrystallization peak of artesunate remains unchanged supporting the absence of incompatibility between artesunate and lactose monohydrate. Thermograms of a physical mixture of (PEG 6000, Lactose monohydrate, and artesunate) and PEG 6000 granules are quite similar indicating that melt granulation process does not impact the thermal behavior of artesunate in granules, and this confirms PXRD results of unaltered crystallinity of artesunate within melt granules. Poloxamer 188 granules as well as Gelucire 50/13 granules thermograms show the characteristic thermal peaks of artesunate in addition to early melting endothermic peaks of binders at about 52 °C and 45 °C respectively. No significant differences were found between the obtained thermograms of granules and their corresponding physical mixtures suggesting lack of interactions.

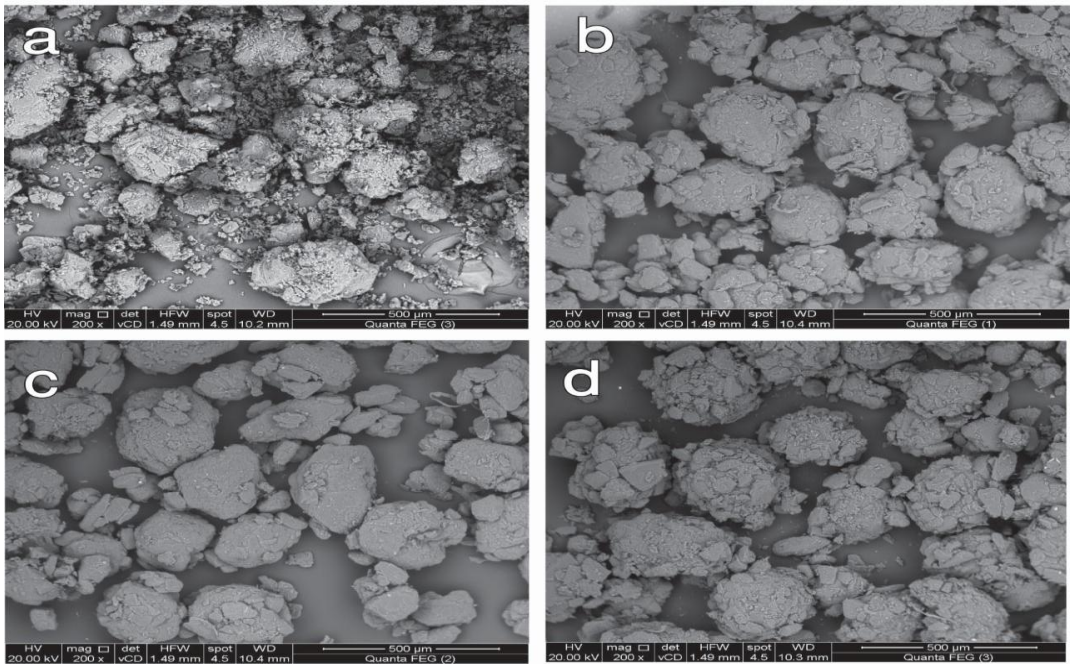

Fig. 4. SEM micrographs of a) Pure crystalline Artesunate, b) Melt granules with 15% PEG 6000, c) Melt granules with 15% Poloxamer 188 and d) Melt granules with 15% Gelucire 50/13. Magnification 200X

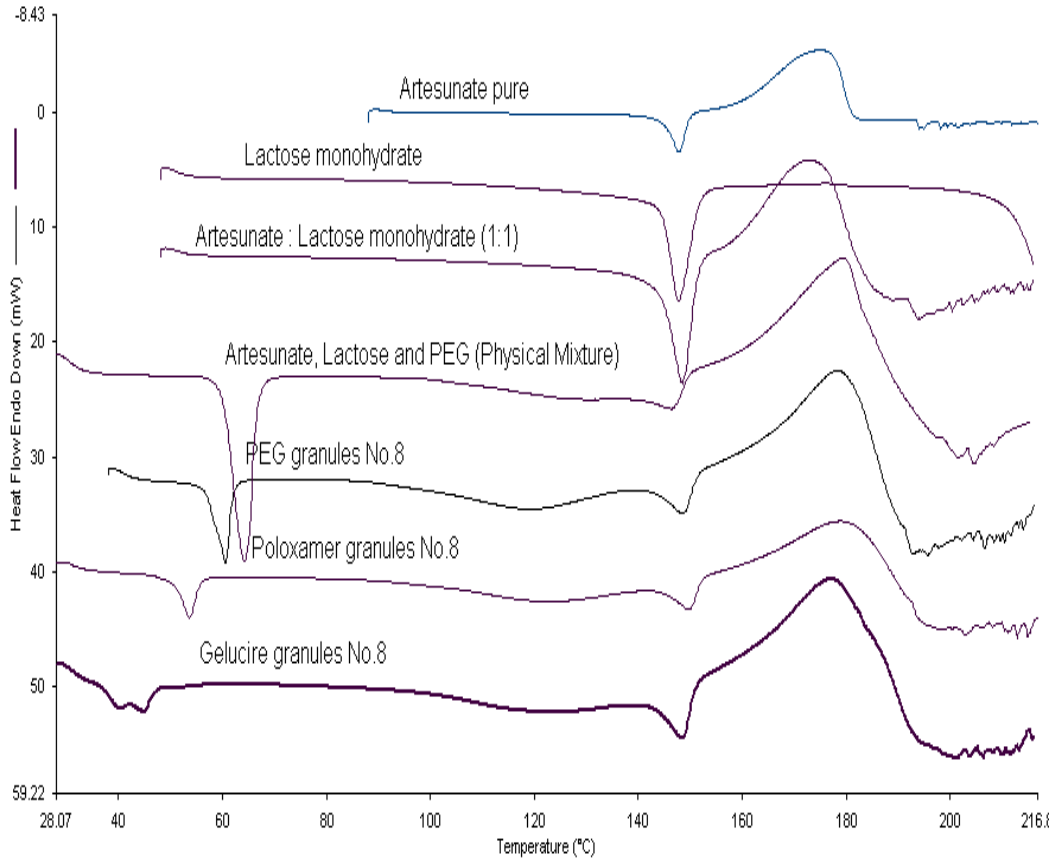

Fig. 5. DSC thermograms of artesunate, PEG 6000 physical mixture, and melt granules

3.11. Granules in-vitro dissolution

Table 9 shows dissolution results of all melt granules at 5, 10 and 15 min, Dissolution results of melt granules at 15 minutes time point showed the superiority of PEG 6000 and Poloxamer 188 over Gelucire 50/13 in improving artesunate dissolution from melt granules due to their more hydrophilic nature in comparison to Gelucire 50/13.

3.12. Tableting properties

Tableting properties of the three binders were comparable; hardness values ranged from (37N-79N), thickness values were almost constant (3.12 ± 0.005 mm), and friability values ranged from (0.06-0.56%). Disintegration values were highly variable (1.7 to 14.25 min), but complied with USP pharmacopeia requirements for the disintegration of immediate-release tablets. This variability is due to the dependency of disintegration on the type and concentration of the binder used. It was noticed that artesunate tablets compressed from melt granules did not disintegrate but gradually eroded, a similar finding of disintegration behavior of compressed melt granules was reported in the literature **[19, 31]**. This can be attributed to the solubility rate of the re-solidified binder matrix that surrounded powder particles, as the matrix could not be destroyed by extragranular disintegrant **[19]**. At the same binder level, Artesunate tablets manufactured with PEG 6000 showed the fastest disintegration time followed by Poloxamer 188, then Gelucire 50/13, which could be attributed to the hydrophilicity and faster erosion of the binder matrix within tablet mass. PEG 6000 polymer is

very soluble in water; Poloxamer 188 is a non-ionic polyoxyethylene–polyoxypropylene copolymer in which the polyoxyethylene segment is hydrophilic which makes it freely soluble in water. Gelucire 50/13 is a mixture of stearic acid esters of glycerol and PEG, its hydrophilic-lipophilic balance (HLB) is 13 which facilitates dispersion in warm water **[32]**. The hydrophobic portion in both Poloxamer and Gelucire structures makes them acting as a surfactant that improves solubilization of poorly soluble compounds yet delays binder water dispersion when compared to PEG that lacks hydrophobic portions. **Table 10** shows very good model fitting of tablets disintegration time data of Poloxamer and Gelucire trials while that of PEG is mediocre. Binder concentration was the sole significant factor increasing tablet disintegration time e.g. by increasing the concentration of PEG 6000 (P= 0.0084). While in case of Poloxamer 188, increasing binder concentration (P= 0.0003), impeller speed (P= 0.0104) and granulation time (P= 0.0152) retarded tablet disintegration. On the other hand, only binder concentration (P= 0.0002), and its quadrate B^2 (P= 0.0167) increased disintegration time in case of Gelucire. Binder concentration was a common significant factor in increasing tablet disintegration time (P<0.05) in all binders. The higher the binder concentration in melt granulate based tablet, the slower the rate of disintegration. A study showed longer disintegration time occurs by increasing the PEG content in tablets pressed from granules of different size ranges **[5]**. As deduced from **table 10** models, **Fig. 6** shows that increasing both binder concentration and impeller speed for Poloxamer 188 binder tablets resulted in an increase in disintegration time. On the other hand, impeller speed had no significant effect on PEG 6000 and Gelucire 50/13 binder tablets as shown in **table 10**. It was found that an increased mixing time and an increased impeller speed induced a larger pellet size and a lower porosity **[28]**. Certain granule porosity is required in order to allow water to penetrate tablets during disintegration.

Table 9. Dissolution results of melt granules expressed as percentage drug dissolved

	PEG 6000 (mean ± SD)			**Poloxamer 188 (mean ± SD)**			**Gelucire 50/13 (mean ± SD)**		
Run No.	**5 min**	**10 min**	**15 min**	**5 min**	**10 min**	**15 min**	**5 min**	**10 min**	**15 min**
1	*73.3±6.3*	*88.4±2.3*	*94.9±1.2*	*95.0±9.2*	*88.8±4.8*	*96.8±1.8*	*58.4±0.8*	*70.1±0.2*	*76.1±2.2*
2	*77.9±3.1*	*88.1±3.0*	*90.6±0.7*	*71.0±2.9*	*84.0±0.3*	*89.8±0.5*	*59.6±0.8*	*72.3±0.3*	*79.1±2.9*
3	*76.6±3.1*	*88.5±2.7*	*92.0±1.9*	*82.9±7.4*	*87.9±3.9*	*90.4±7.6*	*62.2±1.9*	*71.7±0.6*	*77.9±0.5*
4	*88.9±0.7*	*94.1±4.2*	*95.2±2.1*	*82.5±0.7*	*88.5±0.7*	*92.9±0.8*	*64.1±0.8*	*73.9±0.6*	*79.3±2.8*
5	*77.2±2.3*	*91.8±1.9*	*97.0±0.5*	*70.4±1.8*	*80.6±2.3*	*85.8±1.6*	*54.2±1.3*	*67.6±0.7*	*72.8±0.6*
6	*73.0±0.9*	*87.0±2.6*	*93.1±1.7*	*74.0±0.1*	*83.6±2.9*	*90.1±1.9*	*56.5±0.2*	*67.4±2.1*	*72.8±0.3*
7	*72.8±0.1*	*83.3±3.5*	*86.2±2.9*	*72.7±1.9*	*82.7±1.0*	*87.3±0.6*	*54.4±1.6*	*66.4±5.3*	*72.4±0.7*
8	*93.3±1.3*	*96.9±5.1*	*102.3±2.2*	*87.7±1.5*	*90.8±1.2*	*90.7±0.4*	*58.0±0.2*	*70.5±0.7*	*73.7±2.1*
9	*90.0±3.1*	*85.4±1.1*	*87.7±1.9*	*73.0±1.4*	*81.4±1.7*	*83.3±2.8*	*62.8±2.8*	*79.1±1.7*	*87.3±6.8*
10	*74.1±2.5*	*87.0±4.1*	*92.4±0.5*	*71.5±0.03*	*82.0±0.03*	*88.9±0.3*	*52.2±6.8*	*77.4±6.6*	*74.8±8.2*
11	*70.5±0.7*	*84.3±3.1*	*86.5±2.1*	*70.6±0.5*	*82.3±0.03*	*87.3±0.1.8*	*57.2±1.8*	*75.6±3.0*	*79.9±6.7*
12	*86.4±2.7*	*81.3±0.4*	*83.6±1.1*	*73.3±2*	*85.4±2.7*	*86.1±0.8*	*67.6±1.4*	*77.4±1.5*	*94.7±1.3*
13	*70.1±1.7*	*80.2±0.8*	*94.6±4.1*	*70.0±1.6*	*88.6±5.7*	*100.0±5.4*	*52.1±6.2*	*76.5±1.7*	*77.4±8.0*

Table 10. Models suggested for each binder showing artesunate tablet disintegration time as a function of the independent variables and their interactions.

		Goodness of fit	
Binder type	**Model**	**R^2**	**R^2 corrected**
PEG 6000	Disintegration =5.975 - 0.8650 B - 0.003775 S - 0.3217 T + 0.03219 B^2+ 1.278E-06 S^2- 0.02667 T^2 + 0.0004BS+ 0.08333 BT+ 5.556E-05 ST.	0.9475	0.79
Poloxamer 188	Disintegration= 0.8659 + 0.5147 B- 0.006156 S- 0.5825 T-0.01B^2+ 2.694E-06 S^2 + 0.02389 T^2+ 0.0004104 BS + 0.03792 BT + 0.0002306 ST.	0.9936	0.9743
Gelucire 50/13	Disintegration= -10.57+ 2.973 B+ 0.007256 S- 0.4710 T- 0.08844 B^2- 1.917E-06 S^2 + 0.0275 T^2 - 0.0004563 BS+ 0.02354 BT- 0.0001389 ST.	0.9948	0.9792

**Significant factors with (P<0.05) are underlined in the mathematical models.*

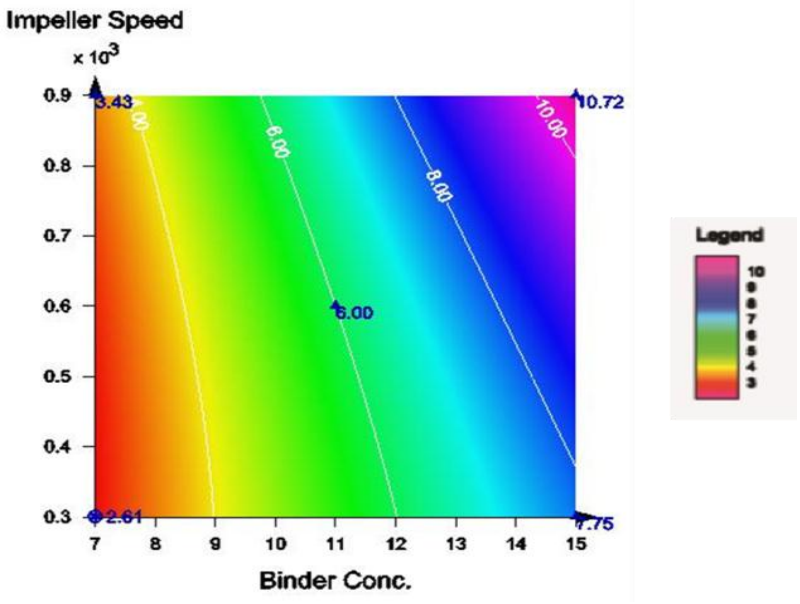

Fig. 6. A Contour plot showing the effect of binder concentration and impeller speed on tablet disintegration time for Poloxamer 188.

3.13. Tablets dissolution at 10 min (T10) point

Dissolution after 5 min was mainly impacted by binder concentration, P<0.05, whereas T10 was impacted by more than one factor, that's why T10 was selected to elucidate the impact of independent factors on the dissolution of artesunate from tablets, **table 11**. It was noticed that dissolution results of tablets were retarded upon compression of granules in trials 4 and 8 for all binders, where both trials showed over granulation due to high binder concentration (15%) combined with high impeller speed (600-900 rpm), and long granulation time (6-9 min). **Fig. 7** shows tablet dissolution profiles of trial no.5 of the three binders based tablets, granulation trial using absolute ethanol as solvent, and for pure artesunate powder. At 10 min point (T10), PEG 6000 and Poloxamer 188 tablets exhibited higher dissolution results than artesunate powder and wet granulation; results of

PEG 6000 and Poloxamer 188 were 95.66% and 88.71% respectively, whereas pure artesunate powder was 60.48% and 84.9% for wet granulation. Dissolution from Gelucire 50/13 tablets was 68.85%, which was lower than PEG 6000 and Poloxamer 188 results. This finding is in agreement with other studies which showed that the presence of Gelucire 50/13 in a system resulted in an extended release due to its hydrophobic nature **[13]**.

The hydrophobic moiety in the Gelucire structure slows down its water dispersion rate relative to the water-soluble PEG 6000 and Poloxamer 188. Gelucire was reported to be less efficient than Poloxamer when used for olanzapine solid dispersion. Dissolution differences among formulation were attributed to different carriers behavior in the aqueous system rather than difference in the solid state of olanzapine **[33]**. It was stated clearly that monolithic Gelucire structures are prone to prolonged erosion times, thereby reducing drug dissolution **[34]**. This is in accordance with our findings of the low efficiency of Gelucire to improve artesunate dissolution compared to the fast dissolution of Poloxamer and PEG systems.

Regarding the wet granulation trial, although tablets showed faster dissolution in comparison to the pure artesunate powder, PEG 6000 containing tablets prepared by hot melt granulation dissolved faster than those prepared by wet granulation. This finding emphasizes the superiority of the melt granulation process for dissolution enhancement in comparison to wet granulation. The explanation for the artesunate dissolution enhancement from melt granulated tablets is the improved wetting of artesunate particles through intimate contact between a hydrophilic binder and the hydrophobic drug as there is a better particle coating for the drug by the melted binder. Wetting is an important mechanism that contributes to the enhancement of the dissolution rate of BCS class II drugs **[8, 9, 20, 35]**. It was reported that Diazepam dissolution was enhanced from PEG 3000 and Gelucire 50/13 melt granules due to the formation of carrier controlled systems in which dissolution rate depends mainly on the dissolution rate of the applied carrier **[20]**. As mentioned above, artesunate crystals in the binder matrix are as islands scattered in water brought by the pore-forming binders, which facilitates fast water diffusion. In case of utilizing PEG 6000 and Poloxamer 188, dissolution rate was improved by the increase in artesunate wettability due to the hydrophilic nature of both binders, and the surface active property of Poloxamer 188 enhancing aqueous solubility. In addition, the prevention of drug particles aggregation within prepared granules contributed to the enhanced dissolution rate of artesunate due to high exposed surface area as reported in a study on carvedilol **[2]**. Poloxamer 188 showed superiority over PEG 4000 in carvedilol dissolution enhancement due to Poloxamer amphiphilic structure that assisted polymeric micelles formation, which enhanced drug solubility. A similar finding was reported for Poloxamer 188, which improved praziquantel dissolution significantly more than PEG **[8]**. Another study showed that Gelucire 50/13 retarded salbutamol sulfate release from a semi-solid matrix, however PEG inclusion with Gelucire improved dissolution results by increasing water uptake resulting in fast erosion of the matrix **[13]**. Regarding PEG 6000 and Poloxamer 188 trials, regression coefficients revealed very good fitting of prediction models for dissolution at T10 whereas, for Gelucire trials, the model was mediocre as shown in **table 11**. Binder concentration was a common significant factor that affected tablet dissolution results at (T10) whatever the binder type. A higher binder concentration resulted in a lower percentage of dissolved artesunate from tablets at T10. This finding is in accordance with what was reported by Masic et al. **[7]**. As explained under particle size, increasing binder concentration leads to the fusion of particles, and increase of geometric

diameter with less surface area exposed for dissolution media. For both PEG and Poloxamer trials, higher binder content, higher impeller speed, and longer granulation time reduced percent drug dissolved at T10 significantly. **Fig. 8** shows the contour plots of binder concentration and impeller speed effect on artesunate tablets dissolution for PEG 6000 and Poloxamer 188. The negative effect of impeller speed on dissolution was more significant at higher binder concentrations. Regarding Gelucire 50/13 trials, only binder concentration (P= 0.0119) was the only retarding factor on artesunate dissolution at T10, **table 11**. It is observed that tablet dissolution results are corresponding to their disintegration results; hence, dissolution was retarded at high binder concentration.

Table 11. Models suggested for each binder showing Artesunate tablet dissolution time at T10 as a function of the independent variables and their interactions

Binder type	Model	Goodness of fit	
		R^2	R^2corrected
PEG 6000	Dissolution at 10 min = 50.91+ 8.357 B+ 0.05872 S- 2.927 T- 0.2175 B^2- 2.061E-05 S^2+ 0.7158 T^2- 0.004171BS- 0.5160 BT - 0.0005583 ST.	0.9907	0.9627
Poloxamer 188	Dissolution at 10 min = - 37.75 + 10.83 B + 0.1644 S + 16.75 T - 0.4322 B2 - 8.197E-05 S2 - 1.132 T2 - 0.006965 BS - 0.2056 BT - 0.002667 ST.	0.9869	0.9475
Gelucire 50/13	Dissolution at 10 min = 91.35 - 8.340 B + 0.06412 S + 3.209 T + 0.2352 B2 - 5.256E-05 S2 - 0.4633 T2 - 0.0006938 BS + 0.2098 BT + 8.889E-05 ST.	0.9315	0.726

**Significant factors with (P<0.05) are underlined in the mathematical model of each binder.*

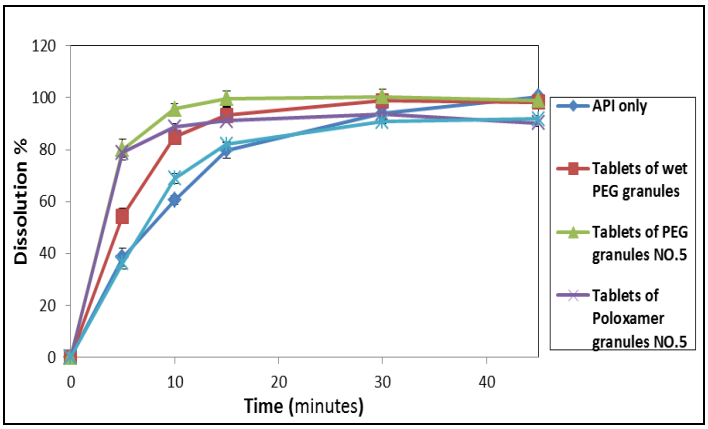

Fig. 7. Dissolution results of pure artesunate powder; wet granulated tablets of PEG 6000, and hot melt granulated tablets of the three binders in acetate buffer pH 5.5

Table 12. Assay results of Artesunate tablets stored at ambient and accelerated conditions after 3 and 6 months.

Time	**PEG 6000 (**mean ± SD**)**		**Poloxamer 188 (**mean ± SD**)**		**Gelucire 50/13 (**mean ± SD**)**	
	Ambient	*Accelerated*	*Ambient*	*Accelerated*	*Ambient*	*Accelerated*
0 month	101.7 ± 1.5		100.4 ± 0.8		99.7 ± 0.9	
3 months	97.94±1.3	100.22±0.7	100.27±1.8	102.46±2.2	104.08±3.5	102.33±2.2
6 months	92.25±0.9	93.95±2.1	96.25±2.8	100.86±1.1	99.5±1.6	94.47±1.1

Table 13. Dissolution results (T15) of Artesunate tablets after 3 months storage at ambient and accelerated conditions.

Time	**PEG 6000 (**mean ± SD**)**		**Poloxamer 188 (**mean ± SD**)**		**Gelucire 50/13 (**mean ± SD**)**	
	Ambient	Accelerated	Ambient	Accelerated	Ambient	Accelerated
T15 at 0 month	97.0±0.5		96.8±1.8		77.9±0.5	
T15 after 3 months	99.75±2.3	89.08±1.5	98.62±3.1	97.63±0.6	85.63±1.1	83.29±1.7
T15 after 6 months	98.44±4.1	93.76±3.02	97.5±2.2	95.16±7.0	82.53±3.7	49.99±3.01

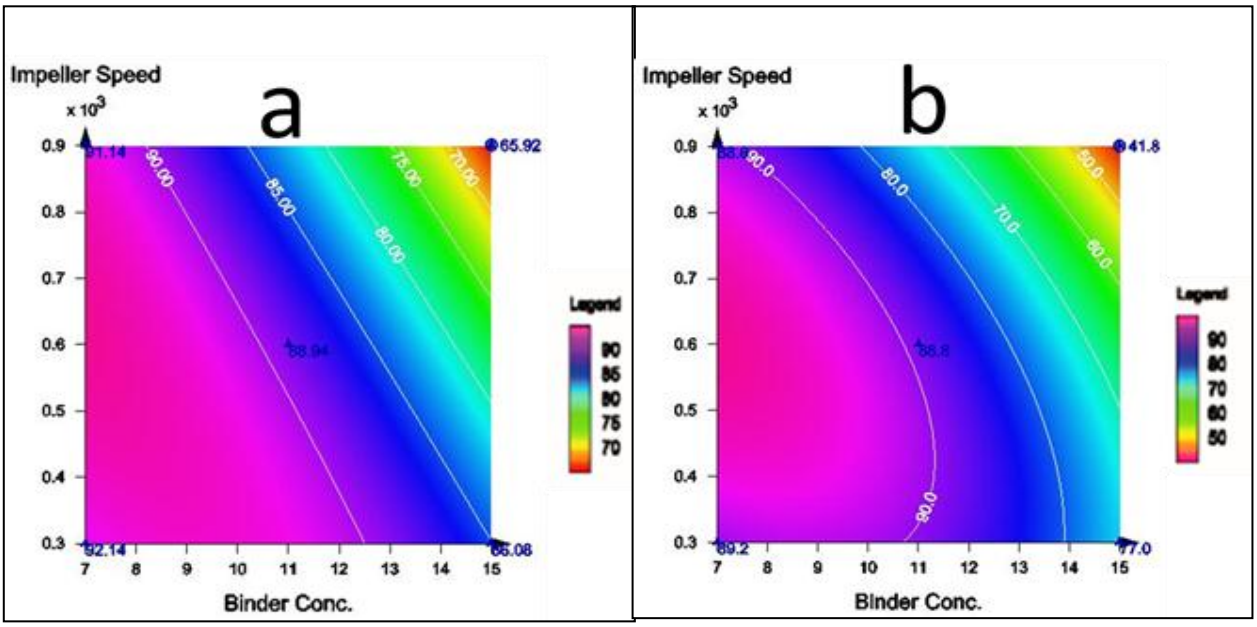

Fig. 8. Contour plots showing the effect of binder concentration and impeller speed on the dissolution of Artesunate from tablets after 10 min (T10) a) PEG 6000 b) Poloxamer 188.

3.14. Stability testing of artesunate tablets

Tablets of Gelucire 50/13 run no.3, Poloxamer 188 run no.1, and PEG 6000 run no.5 were tested for assay and dissolution after three and six months storage at ambient (30 °C -65% RH), and accelerated conditions of temperature and relative humidity (40 °C -75% RH). **Table 12** shows

assay results indicating good stability of artesunate tablets after three and six months. There was no physical change of tablets or a significant decrease in assay values at both ambient and accelerated storage conditions. **Table 13** shows the dissolution results at 15 min (T15) of artesunate tablets after three and six months storage under ambient (30 °C -65% RH), and accelerated (40 °C -75% RH) conditions. By comparing dissolution results of the three batches at zero time, and after three and six months storage, results of all tablets were similar and were not changed even at extreme conditions of heat and moisture except for Gelucire that showed a decrease in dissolution after 6 months at accelerated conditions.

4. CONCLUSION

Tailored drug delivery through DoE approach was achieved for an immediate release tablet of artesunate. Mathematical models showed a very good prediction of responses. The dissolution of artesunate from immediate release tablets was enhanced by the application of melt granulation in comparison to granulation. Furthermore, melt granulation showed stable tablets until six month due to keeping the drug crystalline state. Binder concentration and impeller speeds were the most critical formulation and process factors respectively. At high values of such parameters, dissolution was retarded. Due to hydrophilic nature and drug wetting enhancement by pore-forming, PEG 6000 and Poloxamer 188 were the meltable binders of choice to enhance Artesunate dissolution from their immediate release tablets.

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Conflict of interest

The authors declare no conflict of interest in the work provided in the paper.

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