ARTICLE INFO

A B S T R A C T

The objective of this research is to develop controlled release system of water insolvable Olanzapine (OZ), using Glycerol monooleate (GMO) and Polyethylene glycol (PEG 300) and to study the effects of initial drug loading and different additives on rheological characteristics.

Rheological characterization of all formulations was performed to determine rheological property and viscosity, flow and oscillatory rheological characterization of gel system were calculated and influence of different gel structure was studied to define the rheological characteristics of samples. The best model to explain flow behaviour of all formulation was Cross' model and viscosity was obtained according to the constant shear rate.

Shear rheology is comprehensively applied to consider polymeric bulky liquid crystalline phases to reveal order–disorder transitions (ODT) and order–order (OOT) to explain dynamic phenomena (Kossuth et al., 1999, Lodge et al., 2003, Ruokolainen et al., 2005). Viscoelastic behavior of cubic phases in block copoly As regards, the rheological behavior of self-assembled low-molecular weight liquid crystalline phases has been considered.

The unusual feature with these systems is the actual absence of involvements between hydrophobic chains, which creates the relaxation phenomena to happen in short time scales, generically of the order of seconds. Since this occurs to be identically in the typical time scale of the drug release process, rheological characterization appears to be one of the most related techniques that permit obtaining intelligence in the perception or release characteristics of liquid crystalline materials (Mezzenga et al., 1995).

When behaviour with the rheological characterization of liquid crystalline phases a first recognition has to be made related to the viscoelastic behaviours considered. Whereas no comprehensive study has been performed at deformations for hexagonal and lamellar phases, in the non-linear viscoelastic behaviour, models based on the attendance of liquid crystalline slip planes within which surfactant can be diffused for cubic phases (Jones and McLeish, 1995) and micellar cubic phases (Rodriguez-
Abreu et al., 2004) Although most of the researches focused on the linear-viscoelastic behaviour, explanation of rheological characterization of liquid crystalline phases has basically been completed by studying the storage, \( G' \), and loss modulus, \( G'' \). Alteration in the slope of \( G' \) and \( G'' \) versus composition and temperature has been displayed to be a valuable method to discover OOT and ODT (Radiman et al., 2004, Hamley et al., 2002, Messé et al., 2002). Cubic-to-cubic, cubic-to-hexagonal and hexagonal-to-isotropic fluid transmission has been distinguished by this procedure, taking benefit of the differences in the inherent rigidity of the other phases (Pitzalis et al., 2000).

By investigating the \( G' \) and \( G'' \) depending on shear frequency, attempts have been constructed to establish rheological behaviour of particular liquid crystalline phases (Feng et al., 2004, Habas and Pavie, 2004, Moros et al., 2001, Wang et al., 2005, Mezzenga et al., 2005) Current regards tend to agree that the bicontinuous cubic phases are the most rigid liquid crystalline phases, continued by the reversed hexagonal phase, which is a soft viscoelastic fluid, and the lamellar phase which can be explained as a plastic fluid.

In this research, a series of preliminary test were used to define the general rheological behaviour of prepared matrices.

**MATERIALS AND METHODS**

**Materials**

GMO was purchased from Danisco, Denmark. PEG 300 was obtained from Merck, Germany. Olanzapine was purchased from Sobhan, Iran and other chemical materials were of analytical grade and applied as received.

**Experimental design**

Response surface methodology (RSM) is syntax of mathematical and statistical methods and it offers the best regression mathematical model that has been selected and checked by multiple statistical parameters.

Hence, a Box-Behnken statistical plan was used with 3 factors including; weight ratio of GMO/Water (at 0.2 to 0.4 w/w), weight ratio of PEG (300)/GMO (at 0.2 to 0.6 w/w), and percentage of OZ (at 2 to 4 %). Based on Box-Behnken plan with 3 factors, 17 runs were needed to investigate the main effects and the interaction of three factors on responses.

**Preperation of gels of Olanzapine loaded**

The molten GMO (1 gr) and PEG (300) at various ratio (0.2, 0.4, 0.6 w/w) and OZ (2, 3, 4%) were blended by vortex, and then pH of solutions were evaluated (the indicated pH of all samples are shown to be below 4).

Water at three ratio (water/GMO: 0.2, 0.3, 0.4 w/w) was blended to the solution and mixed by vortex for 10 min and cumulated for 48h to equilibrate at room temperature. After 48 hrs, the clarity of phases of gel samples was corroborated by assay according to their isotropic nature.

**Rheological studies**

Flow and oscillatory rheological properties of gel systems were measured by using a Plate--and-Plate geometry with radius of 25 mm and experiment conditions were kept constant during the experiments. In all experiments, a new gel was used and allowed to rest after 30 min to receive temperature equilibration.

**Flow behaviour**

Flow behaviour surveyed the response of gels to an applied strain or stress (Norling et al., 1992, Zarzycki et al., 2010) and generally rheological studies can be classified in to two types: elasticity properties and viscous or plastic behaviour. Investigating behaviour, all gels were tested in shear rate between 0.01 s\(^{-1}\) and 100 s\(^{-1}\). The goal of setting equation was correlated between the shear stress and shear rate.

Considering this, by using the mathematical models can be analyzed the results of experimental (Bezerril et al., 2006):

\[
\begin{align*}
1- \text{Hershel-Balkly} & \quad \tau = \tau_0 + k (\dot{\gamma})^n \quad \text{(Eq.1)} \\
2- \text{Cross} & \quad \tau = \left(\tau_0 - \tau_{\text{cy}}\right) \left(1 + k (\dot{\gamma})^\beta\right) + \tau_{\text{mf}} \quad \text{(Eq.2)} \\
3- \text{Ostwald} & \quad k (\dot{\gamma})^n \quad \text{(Eq.3)} \\
4- \text{Casson} & \quad \tau^{0.5} = k (\dot{\gamma})^{0.5} \quad \text{(Eq.4)}
\end{align*}
\]

Where: \( \tau \) is shear stress (pa), \( \dot{\gamma} \) is shear rate (s\(^{-1}\)), \( \tau_0 \) is yield stress (pa), \( \eta \) is plastic viscosity (pa.s) and \( K \) is consistency index (pa.s\(^n\)) and \( n \) is flow index.

The shear rate (s\(^{-1}\)) and viscosity (pa.s) can be fitted to power low constitutive equation:

\[
\eta = m \gamma^n \quad \text{(Eq.5)}
\]

Two constant were required: \( m \) is consistency index, \( n \) is flow index

\( n = 1 \rightarrow \text{Newtonian Flow} \)

\( n < 1 \rightarrow \text{Shear Thinning Flow (Pseudo-Plastic)} \)

\( n > 1 \rightarrow \text{Shear Thickening Flow (Dilatant)} \)

**Oscillatory frequency sweep**

Oscillatory frequency sweep test was carried out at low amplitude with angular velocity (w) in range of 0.01 to 100 Hz and temperature was fixed in 37°C (isothermal condition). All measurement were carried out in triplicate. Loss or viscous modulus (\( G'' \)) demonstrates the viscous energy dissipation and large amount of this modulus illustrates viscous characteristic of sample (Martinez-Ruvalcaba et al., 2007) and loss tangent is the content of energy lost to saved energy (tan\( \delta = G''/G' \)). A value of tan\( \delta < 1 \) is a sign of elastic property.

Graph of \( G' \) and \( G'' \) versus frequency, also allow exploiting the longest relaxation time, \( \tau_{\text{max}} \), determined as the reverse of the frequency at which crossover of \( G' \) and \( G'' \) happens. There is a generic consensus on the physical concept of \( \tau_{\text{max}} \), ascribed to the specification diffusion time of the lipid molecules at the lipid–water interface. Alternatively, \( \tau_{\text{max}} \) can be distinguished as the time measure for relaxation to equilibrium formation of the liquid crystalline lipid-water interface, disturbed by shear or orthogonal deformations. Therefore, \( \tau_{\text{max}} \) supplies a typical order of value for many diffusion processes occur in liquid crystalline phases, and can be
applied to investigate the release kinetics of active molecules via the hydrophobic /hydrophilic interface.

<table>
<thead>
<tr>
<th>MODEL Sample</th>
<th>Independent variables (factors)</th>
<th>Hershel-Baldy</th>
<th>Cross 1</th>
<th>Ostwald</th>
<th>Casson 1</th>
</tr>
</thead>
<tbody>
<tr>
<td>F1</td>
<td>Water/GMO:0.3</td>
<td>$\tau_0 = 13.635$</td>
<td>$n = 0.30389$</td>
<td>$R^2 = 0.99436$</td>
<td>$\tau_0 = 0.79516$</td>
</tr>
<tr>
<td>F2</td>
<td>PEG 300/GMO:0.4</td>
<td>$\tau_0 = 36.688$</td>
<td>$k = 0.5987$</td>
<td>$R^2 = 0.99971$</td>
<td>$\tau_0 = 80.609$</td>
</tr>
<tr>
<td>F3</td>
<td>Percentage of OZ:3%</td>
<td>$\tau_0 = 0.99727$</td>
<td>$k = 0.83535$</td>
<td>$R^2 = 0.99753$</td>
<td>$\tau_0 = 6.898$</td>
</tr>
<tr>
<td>F4</td>
<td>Percentage of OZ:2%</td>
<td>$\tau_0 = 7.9633$</td>
<td>$k = 20.631$</td>
<td>$R^2 = 0.99776$</td>
<td>$\tau_0 = 19.743$</td>
</tr>
<tr>
<td>F5</td>
<td>PEG 300/GMO:0.6</td>
<td>$\tau_0 = 3.6744$</td>
<td>$n = 0.30389$</td>
<td>$R^2 = 0.92605$</td>
<td>$\tau_0 = 0.99753$</td>
</tr>
<tr>
<td>F6</td>
<td>Percentage of OZ:4%</td>
<td>$\tau_0 = 19.743$</td>
<td>$k = 20.631$</td>
<td>$R^2 = 0.99776$</td>
<td>$\tau_0 = 19.743$</td>
</tr>
<tr>
<td>F7</td>
<td>Percentage of OZ:3%</td>
<td>$\tau_0 = 3.6744$</td>
<td>$n = 0.30389$</td>
<td>$R^2 = 0.92605$</td>
<td>$\tau_0 = 0.99753$</td>
</tr>
<tr>
<td>F8</td>
<td>Percentage of OZ:2%</td>
<td>$\tau_0 = 3.6744$</td>
<td>$n = 0.30389$</td>
<td>$R^2 = 0.92605$</td>
<td>$\tau_0 = 0.99753$</td>
</tr>
<tr>
<td>F9</td>
<td>Percentage of OZ:4%</td>
<td>$\tau_0 = 3.6744$</td>
<td>$n = 0.30389$</td>
<td>$R^2 = 0.92605$</td>
<td>$\tau_0 = 0.99753$</td>
</tr>
<tr>
<td>F10</td>
<td>PEG 300/GMO:0.4</td>
<td>$\tau_0 = 3.6744$</td>
<td>$n = 0.30389$</td>
<td>$R^2 = 0.92605$</td>
<td>$\tau_0 = 0.99753$</td>
</tr>
<tr>
<td>F11</td>
<td>Percentage of OZ:3%</td>
<td>$\tau_0 = 3.6744$</td>
<td>$n = 0.30389$</td>
<td>$R^2 = 0.92605$</td>
<td>$\tau_0 = 0.99753$</td>
</tr>
<tr>
<td>F12</td>
<td>Percentage of OZ:2%</td>
<td>$\tau_0 = 3.6744$</td>
<td>$n = 0.30389$</td>
<td>$R^2 = 0.92605$</td>
<td>$\tau_0 = 0.99753$</td>
</tr>
<tr>
<td>F13</td>
<td>Percentage of OZ:4%</td>
<td>$\tau_0 = 3.6744$</td>
<td>$n = 0.30389$</td>
<td>$R^2 = 0.92605$</td>
<td>$\tau_0 = 0.99753$</td>
</tr>
<tr>
<td>F14</td>
<td>Percentage of OZ:3%</td>
<td>$\tau_0 = 3.6744$</td>
<td>$n = 0.30389$</td>
<td>$R^2 = 0.92605$</td>
<td>$\tau_0 = 0.99753$</td>
</tr>
<tr>
<td>F15</td>
<td>Percentage of OZ:2%</td>
<td>$\tau_0 = 3.6744$</td>
<td>$n = 0.30389$</td>
<td>$R^2 = 0.92605$</td>
<td>$\tau_0 = 0.99753$</td>
</tr>
<tr>
<td>F16</td>
<td>Percentage of OZ:4%</td>
<td>$\tau_0 = 3.6744$</td>
<td>$n = 0.30389$</td>
<td>$R^2 = 0.92605$</td>
<td>$\tau_0 = 0.99753$</td>
</tr>
<tr>
<td>F17</td>
<td>Percentage of OZ:3%</td>
<td>$\tau_0 = 3.6744$</td>
<td>$n = 0.30389$</td>
<td>$R^2 = 0.92605$</td>
<td>$\tau_0 = 0.99753$</td>
</tr>
</tbody>
</table>

The study of $\tau_{max}$ alterations with composition and temperature has been shown to be the most calculable 2005. Mezzenga et al., 2005), also an alternative procedure to establish phase diagrams, since alters in the
slope of $r_{\text{max}}$ versus composition and temperature obtain all phase transitions, the regions of phase symbiosis, and also OOT enforced by the attendance of host molecules in either the hydrophobic or hydrophilic phases (Mezzenga et al., 2005).

RESULTS AND DISCUSSION

Rheological studies

Flow behaviour

The rheological properties of GMO systems depend on weight ratio water/GMO and PEG/GMO and also percentage of Olanzapine. All gels show a pseudo-plastic and shear thinning behaviour. Many physical gel, with hydrophilic polymer network or spherical micellar gel, exert these properties (Bezerril et al., 2006, Martinez-Ruvalcaba et al., 2007)

Table 1 shows constant parameters and $R^2$ correlation coefficient for various rheological models. Regarding the table, it is of importance to note that, the highest values of the correlation coefficients are recorded for casson 1 model by taking the mentioned models into account, and the specific rheological parameters are calculated, either.

The yield stress is the minimum stress above which fluid was observed and yield stress affected on semi-solid behaviour when Olanzapine injected in to the body. This Table shows that enhanced weight fraction of water leads to an increase of yield stress value and declines by increasing amount of PEG, but percentage of Olanzapine has no great effect on yield stress.

Influence of percentage of Olanzapine on flow behaviour

The data obtained from experiments were plotted in shear stress against shear rate, as shown in Figure 1.

![Figure 1: Effect of percentage of Olanzapine on the shear stress](image1)

From these plots, it can be illustrated that all gels have non-Newtonian behaviour, and also these results observed from the plotting of shear rate against viscosity diagrams. In Figure 1, the slope of diagram increases with increment of Olanzapine percentage and the mentioned effect may be due to the hydrophilic nature of PEG, which reduces the availability of water to mesophases. The fitting results, as shown in Table 1, confirm that the gel with low concentration of Olanzapine has smoother increasing in shear stress and in the cross 1 model $K$ and $n$ decreases.

Influence of weight ratio PEG/GMO on flow behaviour

Figure 2, illustrates the relationship between the shear stress and shear rate, when the fraction of water and Olanzapine maintained constant and the weight ratio of PEG/GMO varied. In the first increasing of shear rate, shear stress has a steep decrease and then with slow slope of shear rate, it increased. Based on the fact that PEG was a hydrophilic material, therefore increasing fraction of PEG lead to lamellar phase with low viscosity. It should be noted that as the fraction of water and percentage of Olanzapine in the matrices increased, viscosity was found to increase, and in contrast, by increasing the amount of PEG, viscosity raised. In Figure 2, with increasing the shear rate, viscosity decreased, which is also indicated in Table 1. When the weight ratio of PEG/GMO rises from 0.2 to 0.4, $k$ and $N$ decline, because gels with the cubic formation convert to lamellar phase, and increasing percentage of Olanzapine in stable condition leads to an enhancement of $N$. Also, with increasing weight ratio of water/GMO, viscosity increased sharply, and therefore $k$, $N$ raised significantly due to formation of cubic phase with high viscosity and also because of the splitting the network resulted from the induced pressure and reducing the viscosity with sharp slope.

![Figure 2: Effect of weight ratio PEG/GMO on the shear stress](image2)

The apparent viscosity profiles for several gels with various formulations are given in Table 2. All gels presented a non-Newtonian and shear thinning behaviour. In the case of consistency, gels with weight ratio of water/GMO: 0.4 and PEG/GMO: 0.4 and percentage of 2% had higher consistency index than all other systems. This indicated that they had more cohesive structure than other gels. The shear thinning behaviour was more pronounced at higher amounts of water.

<table>
<thead>
<tr>
<th>weight ratio water/GMO</th>
<th>weight ratio PEG/GMO</th>
<th>% Olanzapine</th>
<th>$K$</th>
<th>$N$</th>
<th>$R^2$</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.3</td>
<td>0.4</td>
<td>3%</td>
<td>38.416</td>
<td>0.086</td>
<td>0.9919</td>
</tr>
<tr>
<td>0.3</td>
<td>0.2</td>
<td>3%</td>
<td>104.08</td>
<td>0.118</td>
<td>0.9856</td>
</tr>
<tr>
<td>0.3</td>
<td>0.6</td>
<td>2%</td>
<td>42.93</td>
<td>-0.27</td>
<td>0.9971</td>
</tr>
<tr>
<td>0.3</td>
<td>0.6</td>
<td>4%</td>
<td>100.78</td>
<td>-0.333</td>
<td>0.9964</td>
</tr>
<tr>
<td>0.2</td>
<td>0.4</td>
<td>2%</td>
<td>39.844</td>
<td>-0.008</td>
<td>0.9888</td>
</tr>
<tr>
<td>0.4</td>
<td>0.4</td>
<td>2%</td>
<td>2524.6</td>
<td>-0.191</td>
<td>0.9951</td>
</tr>
</tbody>
</table>

Viscosity is major parameter in entrapment of drug and release from prepared gel formulation. Weight fraction of water/GMO and PEG/GMO and also percentage of Olanzapine have been affected on viscosity and drug release pattern. Sample $F_1$ and $F_{10}$ have shown highest and least viscosities, respectively, and the highest viscosity of sample $F_2$ resulted to accumulative drug release at 5th and 164th (19% and 89.2%, respectively) and the least viscosity of sample $F_{10}$ resulted to faster rate of drug release (44.74% and 89.2%, respectively).
Oscillatory frequency sweep

By oscillatory measurement, it can be defined that the linear viscoelasticity region exists, and inducing stress had no significant effect on structure of network in this range. Also, critical oscillatory stress ($\tau_c$) is a point in which linear relationship between strain and shear stress is overset (Barry, 1974).

Influence of weight ratio PEG/GMO on Oscillatory frequency sweep

Figure 3 shows that the destruction of the gel structure (cross-over of storage modulus and loss modulus) turns up at oscillatory stress values and the point of destruction in Fig 3- a is higher than 1400 pa and the frequency at the crossover can be considered as the relaxation time of gel system.

For the gel with weight ratio of PEG/GMO: 0.4 (w/w), the crossover point is 45 (1/s), while it is 200(1/s) for the sample with weight ratio PEG/GMO: 0.2 (w/w), which illustrates more relaxation time at increasing weight fraction of PEG in the matrices. This alteration of crossover point means long-range elastic behaviour, and thus the gel elasticity rises by increasing the amount of PEG.

The rheogram (Figure 3) of the GMO system showed that $G'\tilde{\omega}$ values are higher than their corresponding $G''\tilde{\omega}$ for both gels, which demonstrates that semisolid and rigid microstructure of samples and gel with weight ratio of PEG/GMO: 0.4 (w/w) and PEG/GMO:0.2 (w/w) have minor gap between $G'\tilde{\omega}$ and $G''\tilde{\omega}$ in comparison with other gels (difference between $G'\tilde{\omega}$ values and $G''\tilde{\omega}$ indicated intensity of viscoelasticity properties) and this phenomena can be explained with hydrophilic nature of PEG.

Influence of weight ratio water/GMO on Oscillatory frequency sweep

The crossover point was separated to a liquid like ($G'' > G'$) region from gel, like($G' > G''$). For the samples with weight ratio water/GMO of 0.2(w/w), $G''\tilde{\omega}$ values are higher than their corresponding $G'\tilde{\omega}$, which means that liquid like formation and $G'\tilde{\omega}$ values of the other samples are higher than $G''\tilde{\omega}$, indicating gel configuration. For the sample 4-a, crossover point is 143 (1/s) and for the sample 4-b, it is 100 (1/s), which indicates that reducing weight fraction of water leads to the increase of time relaxation.

CONCLUSION

Rheological behaviour showed that all the prepared gel had a non-Newtonian, pseudo plastic characteristics also appropriate model to explain the type of flow was Cross 1. In the case of consistency, gel with weight ratio water/GMO: 0.4 and PEG/GMO: 0.4 and percent 2%, had higher consistency index than all other systems, that indicated had more cohesive structure than other gels.

References


