FORMULATION AND OPTIMIZATION OF POLYMERIC NANO DRUG DELIVERY SYSTEM OF ACYCLOVIR USING 3² FULL FACTORIAL DESIGN

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ABSTRACT: Acyclovir is an antiviral drug, used for treatment of herpes simplex virus infections with an oral bioavailability of only 10 to 20% (limiting absorption in GIT to duodenum and jejunum), half-life about 3 hrs, soluble only at acidic pH (pKa 2.27). Polymeric nano drug delivery systems of acyclovir have been designed and optimized using 3² full factorial design. Poly (lactic-co-glycolic acid) (PLGA) (50:50) was used as polymers and Pluronic F68 used as stabilizer. Nanoparticles were prepared by solvent deposition method. From the preliminary trials, the constraints for independent variables X₁ (amount of PLGA) and X₂ (amount of Pluronic F68) have been fixed. The derived polynomial equations for particle size and % drug entrapment were verified by check point formulation. The prepared formulations were further evaluated for drug content, in vitro drug release pattern, and short-term stability and drug-excipient interactions. The application of factorial design gave a statistically systematic approach for the formulation and optimization of nanoparticles with desired particle size and high entrapment efficiency. Drug: polymer ratio and concentration of stabilizer were found to influence the particle size and entrapment efficiency of acyclovir loaded PLGA nanoparticles. The release was found to follow first order release kinetics with fickian diffusion mechanism for all batches. These preliminary results indicate that acyclovir loaded PLGA nanoparticles could be effective in sustaining drug release for a prolonged period.

KEYWORDS: Acyclovir, PLGA, nanoparticles, 3² factorial design, sustained release.

INTRODUCTION:
Herpes simplex virus (HSV) is a member of family of herpes viridae, a DNA virus. There are two types of Herpes Simplex Viruses (HSV). viz HSV type 1 and type 2. HSV type 1 is the herpes virus that is usually responsible for cold sores of the mouth, the so called “fever blisters”. HSV type 2 is the one that most commonly causes genital herpes. The infection causes painful sores on the genitals in both men and women. Herpes sores provide a way for HIV to get past the body’s immune defenses and make it easier to get HIV infection. A recent study found that people with HSV had three times the risk of becoming infected with HIV as compared to people without HSV. Currently the treatments available for herpes simplex are conventional tablets and topical gel for application on outbreaks. The drugs that are commonly used for herpes simplex are Acyclovir, Valaclovir and Famciclovir.

Acyclovir, the first agent to be licensed for the treatment of herpes simplex virus infections, is the most widely used drug for infections such as cutaneous herpes, genital herpes, chicken pox, varicella zoster infections. Acyclovir is currently marketed as capsules (200 mg), tablets (200, 400 and 800 mg) and topical ointment. Oral acyclovir is mostly used as 200 mg tablets, five times a day. In addition, long term administration of acyclovir (6 month or longer) is required in immunocompromised patient with relapsing herpes simplex infection. The presently available conventional therapy is associated with a number of drawbacks such as highly variable absorption and low bioavailability (10–20%) after oral administration. Furthermore, with increase in dose, there is decrease in bioavailability. Moreover, because the mean plasma half-life of the drug is 2.5 h, five times a day administration is required. In order to make oral therapy of acyclovir more patient compliant there is a need of using different approaches like matrix tablets, nanoparticles and polymeric films.

The main problem with the therapeutic effectiveness of acyclovir is its absorption which is highly variable and dose dependent thus reducing the bioavailability to 10–20%. Acyclovir is soluble in acidic pH and is predominantly absorbed from upper gastro intestinal tract (GIT) to duodenum to jejunum regions.
There are indications of its active absorption from the duodenum and jejunum regions of GIT.

The inherent shortcomings of conventional drug delivery and the potential of nanoparticles as drug delivery systems have offered tremendous scope for researchers in this field and is fast moving from concept to reality. Nanoparticles may be used for oral administration of gut-labile drugs or those with low aqueous solubility. These colloidal carriers have the ability to cross the mucosal barrier as such. In addition to the potential for enhancing drug bioavailability via particle uptake mechanisms, nanoparticulate oral delivery systems also have slower transit times than larger dosage forms increasing the local concentration gradient across absorptive cells, thereby enhancing local and systemic delivery of both free and bound drugs across the gut. These colloidal carriers are expected to develop adhesive interactions within the mucosa and remain in the gastrointestinal tract, while protecting the entrapped drug from enzymatic degradation, until the release of the loaded drug or their absorption in an intact particulate form. Most evidence suggests that the favored site for uptake is the Peyer’s patches (PP) lymphoepithelial M cell. It has been shown that microparticles remain in the Peyer’s patches, whereas nanoparticles disseminate systemically. Nanoparticles bind to the apical membrane of the M cells, followed by rapid internalization and shuttling to lymphocytes wherein size and surface charge play a crucial role for their uptake, and these properties are influenced by the stabilizer used.

Appropriate selection of the polymeric matrix is necessary in order to develop a successful nanoparticulate delivery system. Biodegradable polymers have received much attention in recent years. Poly(lactide-co-glycolide) (PLGA) has been most extensively used because of its biocompatibility and biodegradability with degradation products formed at a slow rate, thus not affecting the normal cell function. PLGA degrades in vivo to lactic and glycolic acids, which are subsequently eliminated as carbon dioxide and water via the Krebs cycle. The release of drug from the nanoparticles depends on polymer degradation, which is governed by the nature of copolymer composition and its molecular weight. For this study, we used PLGA 50:50, which is known to hydrolyze at a faster rate than those containing a higher proportion of polyactic acid. This article reports the design of biodegradable nanoparticles containing acyclovir for oral delivery.

The present investigation, Poly (lactic-co-glycolic acid) (PLGA) (50:50) was used as polymers and Pluronic F68 used as stabilizer. Nanoparticles were prepared by solvent deposition method. From the preliminary trials, the constraints for independent variables X_1 (amount of PLGA) and X_2 (amount of Pluronic F68) have been fixed. The derived polynomial equations for Particle size and % drug entrapment were verified by check point formulation. The prepared formulations were further evaluated for drug content, in vitro drug release pattern, short-term stability and drug-excipient interactions. The application of factorial design gave a statistically systematic approach for the formulation and optimization of nanoparticles with desired particle size and high entrapment efficiency.

**MATERIALS AND METHODS:**

Acyclovir was a gift sample from Ajanta Pharmaceutical Limited, Mumbai; Poly (D, L Lactide-co-Glycolide) (PLGA 50:50 and PLGA 85:15) were obtained as gift samples from Indena Ltd., Italy; Pluronic F 68 was procured from StridesArco Lab, bangalore.; Acetone, Cellophane Membrane, were purchased from SD Fine Chem. Ltd., Mumbai. All other reagents and chemicals used in this study were of Analytical Grade.

**METHODS:**

**Formulation of poly (lactic-co-glycolic acid) (PLGA) Nanoparticles:**

PLGA nanoparticles were prepared by the solvent deposition method. Acyclovir was dissolved in neutral water (pH = 7) kept at 35-40°C containing a hydrophilic stabilizer (Pluronic F68) at various concentrations. PLGA was solubilized in acetone (40ml) at various concentrations. The organic phase was poured into the aqueous solution drop wise, under stirring (RPM 5000) for 2 hrs, thus forming a milky colloidal suspension. The organic solvent was then evaporated by using a Rota evaporator. The resultant dispersion was dried using a freeze drying method.

**Experimental Design:**

The formulations were fabricated according to a 3² full factorial design, allowing the simultaneous evaluation of two formulation variables and their interaction. The experimental designs with corresponding formulations are outlined in table-1. The dependent variables that were selected for study were particle size (Y_1), and % drug entrapment (Y_2).

**In Vitro Characterization of PLGA Nanoparticles**

**I) Determination of particle size**

The particle size and size distribution of the acyclovir loaded PLGA (50:50) nanoparticles were characterized by laser light scattering using Particle size Analyzer (Malvern Mastersizer Hydro-2000 SM, UK). The obscuration level was set between 7 to 11 %, distilled water was used as medium.

**II) Determination of Encapsulation Efficiency**

**i) Estimation of Free Drug**

The free drug (per 100mg of formulation) was estimated by taking said quantity of formulation in dialysis bag (cellophane membrane, molecular weight cut off 10000-12000 Da, Hi-Media, India ) which was tied and placed into 100ml water (pH = 7) kept at 37°C on magnetic stirrer. At predetermined time intervals, 5ml of the samples were withdrawn by means of a syringe. The volume withdrawn at each interval was replaced with...
same quantity of fresh water (pH=7) maintained at 37 °C. The samples were analyzed for free drug by measuring the absorbance at 252nm using UV-visible spectrophotometer (Shimadzu UV-1700) after suitable dilution. Above described process of withdrawing sample and analysis was continued till a constant absorbance was obtained.

**ii) Estimation of encapsulated drug**

Encapsulated drug (per 100mg of formulation) was estimated by taking residue formulation remained in dialysis membrane after estimation of free drug content, as described above. Quantity left behind in dialysis membrane was added to acetone (10ml) to dissolve PLGA and filtered. Residue remaining on filter paper was dissolved in 100ml of water (pH=7) kept at 37 °C and after removing supernatant, sample was analyzed for drug content by measuring the absorbance at 252nm using UV-visible spectrophotometer (Shimadzu UV-1700) after suitable dilution. The percentage of drug entrapped and the percentage of free drug are calculated by following Eq. 16.

Formulae to calculate % free drug and % drug entrapment

1) % free drug = 
   \[ \frac{\text{Amount of free drug present in 100mg of formulation}}{\text{Total amount of drug present in 100mg of formulation}} \times 100 \]

2) % Drug entrapment = 
   \[ \frac{\text{Amount of encapsulated drug present in 100mg of formulation}}{\text{Total amount of drug present in 100mg of formulation}} \times 100 \]

**III) Statistical Analysis:**

The results from factorial design were evaluated using PCP Disso 2000 V3 software. Step-wise backward linear regression analysis was used to develop polynomial equations for dependent variables particle size (Y1) and % drug entrapment (Y2) which bear the form of equation-1:

\[ Y = b_0 + b_1X_1 + b_2X_2 + b_{12}X_1X_2 + b_{11}X_1^2 + b_{22}X_2^2 \] ……..1

Where Y is dependent variable, b0 arithmetic mean response of nine batches, and b1 estimated coefficient for factor X1. The main effects (X1 and X2) represent average result of changing one factor at a time from its low to high value. The interaction term (X1X2) shows how the response changes when two factors are simultaneously changed. The polynomial terms (X1^2 and X2^2) are included to investigate non-linearity. The validity of the developed polynomial equations was verified by preparing check point formulation (C).

**IV) Drug Release Study:**

A quantity of selected factorial formulations equivalent to 25 mg of the drug (table no.3 indicates drug content per 100mg of formulation) was taken in the dialysis bag (cellulose membrane, molecular weight cut off 10000-12000 Da, Hi-Media, India ). The dialysis bag was then suspended in a flask containing 100 ml of 0.1 N HCl on a magnetic stirrer at 37±0.5 °C at 100 rpm. Required quantity (5ml) of the medium was withdrawn at specific time periods (1, 2, 3, 4, 6, 8, 10, 12, 24, 32 hours) and same volume of dissolution medium was replaced in the flask to maintain a constant volume. The withdrawn samples were filtered and 5 ml filtrate was made up to volume with 100 ml of 0.1 N HCl. The samples were analyzed for drug release by measuring the absorbance at 252nm using UV-visible spectrophotometer (Shimadzu UV-1700).

**V) Drug-Polymer Interaction Studies:**

Differential scanning calorimetry (DSC) is one of the most powerful analytical techniques, which offers the possibility of detecting chemical interaction. Acyclovir (pure drug), PLGA, and physical mixtures of drug and polymer at different ratios (1:1, 1:1.5, 1:2, 1:2.5) were kept at 40 ±2°C/75±5% RH. Samples at 0, 1, 2, 3 and 6 months were withdrawn and sent for testing.

**VI) SEM Photomicrographs**

The morphology of nanoparticles was examined by scanning electron microscopy (SEM, JSM-5310LV scanning microscope Tokyo, Japan). The nanoparticles were mounted on metal stubs using double-sided tape and coated with a 150 Å layer of gold under vacuum. Stubs were visualized under scanning electron microscope.

**RESULTS AND DISCUSSION:**

Out of all factorial formulations developed by the above described method, formulations, F4, F5, F7, F8 and F9 were found to be free flowing i.e. non sticky but formulations, F1, F2, F3 and F6 were found to be sticky. All formulation were white and powdery in appearance.

**Particle Size and Entrapment Efficiency:**

The particle size affects the biopharmaceutical, physicochemical and drug release properties of the nanoparticles. A graphical representation of the particle size of PLGA nanoparticles obtained is given in Fig. 3. Particle size is an important parameter because it has a direct relevance to the stability of the formulation. Larger particles tend to aggregate to a greater extent compared to smaller particles, thereby resulting in sedimentation. The amount of stabilizer used also has an effect on the properties of nanoparticles. If the concentration of stabilizer is too low, aggregation of the polymer will take place, whereas, if too much stabilizer is used, drug incorporation could be reduced as a result of the interaction between the drug and stabilizer.

The effect of the concentration of the polymers tested is negative or positive. A positive effect would imply that increasing the concentration causes the emulsion to have larger droplets, hence leading to larger particles. A negative effect means that increasing the
Pluronic F68 i.e. 0.25% . Drug entrapment efficiency respectively with constant concentration of stabilizer (Pluronic F68) i.e. 0.25% . Drug entrapment efficiency of factorial formulations F3, F6, F9 where drug: polymer ratio increased from 1:1, 1:2 and 1:3 showed reduced or insignificant change in the drug entrapment efficiency.

This can be explained by observing drug entrapment efficiency of factorial formulations F3, F6, F9 where drug: polymer ratio increased from 1:1, 1:2 and 1:3 respectively with constant concentration of stabilizer (Pluronic F68) i.e. 0.25% . Drug entrapment efficiency increased from 84.67% to 94.5% and then decreased it to 90.06%. It is also observed that as percentage of stabilizer increased from 0.25% to 1% entrapment efficiency and particle size decrease significantly, the same can be explained with respective to factorial formulation F1, F4, F7 and F2, F5, F8 where it is observed that as drug: polymer ratio increases, entrapment efficiency increased significantly but further increase in drug: polymer ratio has negative or insignificant effect on drug entrapment. For factorial formulation F1, F2, F3 where Drug: Polymer ratio is constant i.e.1:2 and concentration of stabilizer decreased from 1% to 0.25%, drug entrapment efficiency increased from 64.06% to 90.06% and particle size increased from 1230nm to 1500nm. Thus it can be concluded that the stabilizer had greater influence on both dependent parameters (particle size and drug entrapment) as compared to drug: polymer ratio, which shows positive effect on particle size and insignificant effect on drug entrapment efficiency.

In-Vitro Drug release study

Based on highest % drug entrapment and lowest particle size (below 1100 nm) batches F5, F8, and F9 were selected to carry out the drug release study.

Drug release from nanoparticles and subsequent biodegradation are important for developing successful formulations. The release rate of nanoparticles depends upon
i) desorption of the surface-bound/adsorbed drug ; ii) diffusion through the nanoparticle matrix; iii) diffusion (in case of nanocapsules) through the polymer wall; iv) nanoparticle matrix erosion; and v) a combined erosion/diffusion process. Thus, diffusion and biodegradation govern the process of drug release.

It is generally anticipated from a bulk eroding polymer such as 50:50 PLGA to give an initial burst release followed by a controlled release, in contrast to the release pattern observed in other controlled release systems, for example sustain release tablets, pellets and beads. In cases where there is an initial burst effect, the high initial release may be attributed to the presence of crystals of free and weakly bound drug on the surface of the particulate carriers.

The mechanism of drug release from nanoparticles is determined by different physical–chemical phenomena.

concentration causes the emulsion to be more stable, hence leading to smaller particles. From Fig.3 and Fig. 4 and Table-1, it is revealed that as drug: polymer (Acyclovir:PLGA) ratio increased from 1:1 to 1:2 particle size increased significantly and drug entrapment also increased but there after, further increase in drug: polymer ratio showed reduced or insignificant change in the drug entrapment efficiency.

In equation (3) positive sign for coefficient of X indicates that % drug entrapment increases when concentration of stabilizer (Pluronic F 68) increases from 1% to 0.25%, drug entrapment efficiency increased significantly but further increase in drug: polymer ratio has negative or insignificant effect on drug entrapment. For factorial formulation F1, F4, F7 and F2, F5, F8 where it is observed that as drug: polymer ratio increases, entrapment efficiency increased significantly but further increase in drug: polymer ratio has negative or insignificant effect on drug entrapment. For factorial formulation F1, F2, F3 where Drug: Polymer ratio is constant i.e.1:2 and concentration of stabilizer decreased from 1% to 0.25%, drug entrapment efficiency increased from 64.06% to 90.06% and particle size increased from 1230nm to 1500nm. Thus it can be concluded that the stabilizer had greater influence on both dependent parameters (particle size and drug entrapment) as compared to drug: polymer ratio, which shows positive effect on particle size and insignificant effect on drug entrapment efficiency.

The exponent n has been proposed as indicative of the release mechanism. In this context, n ≤ 0.43 indicates Fickian release and n = 0.85 indicates a purely relaxation controlled delivery. Intermediate values 0.43 < n < 0.85 indicate an anomalous behavior (non-Fickian kinetics corresponding to coupled diffusion/polymer relaxation).

The average percentage release was fitted into different release models: zero order, first order, Higuchi’s square root plot, and Hixson Crowell cube root plot. The models giving a correlation coefficient close to unity were taken as the order of release. In vitro drug release data of all selected factorial formulations was subjected to goodness of fit test by linear regression analysis according to zero order and first order kinetic equations, Higuchi’s, Hixson Crowell and Korsmeyer-Peppas models to ascertain the mechanism of drug release. From various parameters determined for drug release from nanoparticles based on Peppas Model, Higuchi Model and Diffusion profile, it is evident that values of ‘r’ for Higuchi plots of factorial formulations F5, F8 and F9 are 0.8477, 0.9343 and 0.8322 respectively, for first order plots 0.9125,0.8952 and 0.8834 respectively and those of ‘n’ (Diffusion exponent) values of Peppas equation are 0.2547, 0.2512 and 0.3637 respectively. This data reveals that drug release follows first order release kinetics with fickian diffusion mechanism. Drug release for selected factorial formulations F5, F8, F9 are 24.27%, 29.85% and 16.14% respectively after 1 hr. These formulation show initial burst release followed by a controlled release. Kinetic exponent ‘n’ for these formulations indicate diffusion through the nanoparticle matrix as well matrix erosion. Finally, it can be concluded that the different drug release rates may be attributed to different sizes of the nanoparticles. It is expected as the particle size of PLGA nanoparticle is smaller, their surface area will be more and the drug release is faster.

Development of Polynomial Equations:

From the data of Experimental design and Parameters (table-1) for factorial formulations F1 to F9, polynomial equations for two dependent variables (particle size and % drug entrapment) have been derived using PCP Disso 2000V3 software. The equation derived for particle size is:

\[ Y_1 = 1.0193 + 0.3567 X_1 - 0.1500 X_2 \]  \[ \ldots 2 \]

The equation derived for % drug entrapment is:

\[ Y_2 = 83.49 + 3.6750 X_1 - 14.095X_2 \]
\[ - 6.2283 X_1^2 - 3.6983 X_2^2 \]  \[ \ldots 3 \]

In equations (2) negative sign for coefficient of \( X_2 \) indicates that the particle size of nanoparticles increases when concentration of stabilizer (Pluronic F 68) is decreased and positive sign for coefficient of \( X_1 \) indicate positive effect of polymer concentration (PLGA) on particle size.

In equation (3) positive sign for coefficient of \( X_1 \) indicates that the % drug entrapment increases when concentration of polymer (PLGA) increases and negative sign for coefficient of \( X_2 \) indicates that % drug
entrainment of nanoparticles increases when concentration of stabilizer (Pluronic F 68) decreases.

Validity of the above equations was verified by designing check point formulation (C). The particle size and % drug entrainment predicted from the equations derived and those observed from experimental results are summarized in table-2. The closeness of predicted and observed values for particle size and % drug entrainment indicates validity of derived equations for dependent variables.

Response Surface Plots:

Graphical presentation of the data can help to show the relationship between response and independent variables. Graphs gave information similar to that of the mathematical equations obtained from statistical analysis. The response surface graphs of particle size and % drug entrainment are presented in figures-1, and 2. respectively.

The response surface plots illustrated that as concentration of as polymer (PLGA) increases, the value of dependent variable i.e. particle size increases and as concentration of stabilizer (Pluronic F 68) increases the value of dependent variable i.e. particle size decreases. Similarly the response surface plots for % drug entrainment shows positive effects of independent variable i.e. polymer concentration (PLGA) and negative effect of other independent variable i.e. concentration of stabilizer (Pluronic F 68).

Drug-Polymer Interaction Studies:

Differential Scanning Calorimetry (DSC) gives information regarding the physical properties like crystalline or amorphous nature of the samples. The DSC thermogram of Acyclovir (Fig.6.B) shows an exothermic peak at 267.03 corresponding to its melting temperature, which was not detected in the thermograms for Acyclovir nanoparticles of PLGA 50:50 (Fig.6 A). It has been shown by a couple of authors that when the drug does not show its exothermic peak in the formed nanoparticles, it is said to be in the amorphous state. Hence it could be concluded that in both the prepared PLGA nanoparticles the drug was present in the amorphous phase and may have been homogeneously dispersed in the PLGA matrix.

Conclusion

Acyclovir loaded nanoparticles were prepared by the solvent deposition method. The application of factorial design gave a statistically systematic approach for the formulation of nanoparticles with desired particle size and high entrapment efficiency. Drug: polymer ratio and concentration of stabilizer were found to influence the particle size and entrapment efficiency of acyclovir loaded PLGA nanoparticles but the concentration of stabilizer had greater influence on both dependent variables (Particle size and Drug entrapment) as compared to Drug:Polymer rato. In vitro drug release study of selected factorial formulations (F5, F8, F9) showed 58.48 %, 66.54%, and 62.92% release respectively in 32 hrs. The release was found to follow first order release kinetics with fickian diffusion mechanism for all batches. These preliminary results indicate that acyclovir loaded PLGA nanoparticles could be effective in sustaining drug release for a prolonged period.

### Table-1: Experimental design and Parameters for 3² Full Factorial Design Batches

<table>
<thead>
<tr>
<th>Batch Code</th>
<th>Variable level in Coded Form</th>
<th>Particle size (nm)</th>
<th>% drug entrainment ±SD**</th>
<th>% Free Drug ±SD**</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>X₁</td>
<td>X₂</td>
<td></td>
<td></td>
</tr>
<tr>
<td>F1</td>
<td>+1</td>
<td>+1</td>
<td>1230</td>
<td>64.06±1.2</td>
</tr>
<tr>
<td>F2</td>
<td>+1</td>
<td>0</td>
<td>1310</td>
<td>81.32±0.52</td>
</tr>
<tr>
<td>F3</td>
<td>+1</td>
<td>-1</td>
<td>1500</td>
<td>90.06±1.69</td>
</tr>
<tr>
<td>F4</td>
<td>0</td>
<td>+1</td>
<td>1030</td>
<td>66.24±1.64</td>
</tr>
<tr>
<td>F5</td>
<td>0</td>
<td>0</td>
<td>1074</td>
<td>82.36±0.84</td>
</tr>
<tr>
<td>F6</td>
<td>0</td>
<td>-1</td>
<td>1130</td>
<td>94.5±0.46</td>
</tr>
<tr>
<td>F7</td>
<td>-1</td>
<td>+1</td>
<td>395</td>
<td>54.36±1.73</td>
</tr>
<tr>
<td>F8</td>
<td>-1</td>
<td>0</td>
<td>580</td>
<td>74.36±1.64</td>
</tr>
<tr>
<td>F9</td>
<td>-1</td>
<td>-1</td>
<td>925</td>
<td>84.67±0.98</td>
</tr>
<tr>
<td>C</td>
<td>-0.5</td>
<td>-0.5</td>
<td>887</td>
<td>81.19±0.94</td>
</tr>
</tbody>
</table>

**All the tests were carried out in triplicate
C check point batch.

* For PLGA (50:50) (X₁) transformed levels in drug: polymer ratios are: −1= 1:1; ‘0’=1:1.5, +1=1:2, −0.5=1:1.25
# For surfactant (Pluronic F68) (X₂) transformed levels in % are:−1=0.25; ‘0’=0.5, +1= 1 , −0.5=0.3
Table-2: parameters of check point formulation

<table>
<thead>
<tr>
<th>Formulation</th>
<th>Predicted values</th>
<th>Observed values</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Particle Size</td>
<td>% drug entrapment</td>
</tr>
<tr>
<td></td>
<td>nm</td>
<td></td>
</tr>
<tr>
<td>C</td>
<td>915</td>
<td>86.23</td>
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</table>

Table - 3. Various parameters determined for drug release from nanoparticles based on Peppas Model, Higuchi Model and Diffusion profile

<table>
<thead>
<tr>
<th>Formulation</th>
<th>Correlation Coefficient, (First Order)</th>
<th>Kinetic Exponent, ‘n’</th>
<th>Correlation Coefficient, (Higuchi model)</th>
<th>T 50% hrs</th>
</tr>
</thead>
<tbody>
<tr>
<td>F5</td>
<td>0.9125</td>
<td>0.2547</td>
<td>0.8477</td>
<td>12.8</td>
</tr>
<tr>
<td>F8</td>
<td>0.8952</td>
<td>0.2512</td>
<td>0.9343</td>
<td>8.8</td>
</tr>
<tr>
<td>F9</td>
<td>0.8834</td>
<td>0.3637</td>
<td>0.8322</td>
<td>11</td>
</tr>
</tbody>
</table>

Figure no-1: Response surface plot showing effect of factorial variables on particle size.
Fig no-2 Response surface plot showing effect of factorial variables on % drug entrapment.

Fig no -3 Comparison of particle size of factorial formulations and check point formulation.
Fig no -4 Comparison of drug entrapment of factorial formulations and check point formulation

Fig-5. Average % Drug release of F 5, F 8, F 9 Formulations
Fig. 6-DSC Thermographs of A- Formulation, B- Drug, C- Polymer, D- Physical Mixture

Fig. 7. SEM photomicrograph of PLGA (Uncoated) Nanoparticles (×10,000). Scale bar = 50 µm
REFERENCES:


