CHAPTER 6

DISCUSSION

6.1 Preformulation studies

The preformulation is the preliminary study for investigation and detail understanding of the physicochemical and chemical dynamics of drug substances through stability under the conditions to select correct form of drug and non-drug components for developing optimal drug delivery system\textsuperscript{151}. Based on the preformulation results, proper excipients are used in the manufacturing of protocol products and to optimize the release of drug for an extended period at the site of action.

6.1.1 Development calibration curves of aceclofenac sodium and dexibuprofen by UV-visible spectrophotometry

The development of standard calibration curves of aceclofenac sodium and dexibuprofen in SGF pH 1.2, pH 6.8 and 7.4 PBS were generated by UV-visible spectrophotometric technique at absorption maxima 275nm for aceclofenac sodium and 264.5 nm for dexibuprofen

The standard curves of aceclofenac sodium reveal that the drug obeys beer’s law in concentration range of 2 – 20\(\mu\)g/ml. The linear regression equation was generated and used to calculate the amount of drug present in the formulated dosage forms i.e. SGF pH 1.2 \(y = 0.021x - 0.00232, R^2 = 0.9990\), pH 6.8 PBS \(y= 0.058x - 0.05963, R^2=0.9995\), and pH 7.4 PBS \(y= 0.098x + 0.12615, R^2 = 0.9980\).

The standard curves of dexibuprofen indicate that the drug obeys beer’s law in concentration range of 50-350 \(\mu\)g/ml. The linear regression equation was generated and used to calculate the amount of drug present in the formulated dosage forms i.e.
SGF pH 1.2 (y= 0.016x – 7.10852, $R^2= 0.9981$), pH 6.8 PBS (y= 0.0452x – 2.8328, $R^2=0.9995$), and pH 7.4 PBS (y=0.652x + 0.6743) $R^2 = 0.9994$).

6.1.2 Saturation solubility studies

The dissolution study of dosage forms containing poorly water soluble drugs is necessary to modifications in the dissolution medium to increase the aqueous solubility. Therefore, there are many approaches used to define increase in the solubility of such type of drugs among them addition of surfactants is necessary for solubilizing such type of drugs, because various surfactants are present in the GI-fluid\textsuperscript{152}. Aceclofenac sodium is practically insoluble in water. The table 5.1.3 explains that the solubility of aceclofenac sodium in 0.1N HCl is very scarcely compared to distilled water. The solubility of aceclofenac sodium significantly increases with increasing the concentration of sodium lauryl sulfate (SLS) in 0.1N HCl as well as pH of the dissolution media. Aceclofenac sodium shows sufficient solubility in 0.1N HCl with 2%w/v of SLS up to 0.487mg/ml which adequately maintains the sink condition. Therefore, 2% w/v used in the dissolution medium for in-vitro drug release studies for initial 2 hours. The solubility of aceclofenac sodium was more in 1%w/v of calcium chloride solution than in double-distilled water, which induces certain amount of drug release when prolonged exposés of the beads in curing medium during the manufacturing process.

The microemulsion is a monophasic liquid formulation at ambient temperature when added into aqueous phase. The single phase was formulated by using oil, surfactants, co-surfactants and aqueous solution of drug that has good solvent properties allow solubilizing the drug in solution\textsuperscript{142}. In primary studies certain physicochemical properties of microemulsion were assessed (droplet size) by using different concentrations of oil, surfactant and co-surfactant. The efficiency of micro-
emulsification was estimated by determining the rate of emulsification and droplet size distribution. The droplet sizes of emulsion were dictates the performance of microemulsification because the rate and extent of drug release as well as absorption mainly influenced by droplet size.\textsuperscript{155} By increasing the surfactant and co-surfactant concentration from 2:1 to 1:1, decreases the average diameter of emulsion formed with 10% (v/v) oil (Labrafac PG) but the average diameter slightly increases with above 10% (v/v) of oil which induces the formation of large size beads. Moreover, the spontaneous emulsion formation was not efficient with less than 2:1 concentration of surfactant: co-surfactant and was used in the preparation of microbeads. The results are not mentioned in the present protocol study.

The solubility of dexibuprofen showed good solubility with surfactants. Among them the Labrasol a medium-length alkyl chain surfactant with HLB 14 was selected as a surfactant. Moreover, the miscibility of selected oils with Labrasol at 2:1 volume ratio was investigated by clarity of oil/surfactant mixture. The castor oil, cotton seed oil, sesame oil, peanut oil and Labrafil 2125 CS were poorly miscible with Labrasol, whereas Labrafac PG was well miscible and formed clear solution. Labrafac PG showed higher drug solubility than other oils. Thus, Labrafac PG was selected as an oily vehicle due to its good emulsion-forming with surfactant and co-surfactant. Further, Span 80 (HLB 6) was selected as a co-surfactant for its good solubility and influences the distribution of gelling agent (SA) for preparing homogenous polymeric dispersion.

6.1.3 FT-IR spectroscopy

The figures of FTIR spectra, observed that the characteristic absorption peaks of pure aceclofenac sodium was obtained at 3276.5, 2915.5, 1716.5, 1589.3, 1279.6 and 749.4cm\textsuperscript{-1} corresponding to NH- stretching, C=O stretching of –COO and –
COOH group respectively. The characteristic absorption peaks of SA show around 3077.15, 2914.98, 1615.34, 1359.60, and 745.05 cm\(^{-1}\) reflective of O–H, C–H, COO– (asymmetric), COO– (symmetric), C–O–C stretching respectively. The physical mixture of aceclofenac sodium and SA characteristic peaks were obtained at 2820.28, 1591.72, 1384.91, 1102.03 and 766.4 cm\(^{-1}\). The characteristic absorption peaks of pure dexibuprofen were obtained at 3087.56, 2994.16, 1707.56, 1460.7, 13620.10 and 705.5 cm\(^{-1}\) corresponding to O–H, C–H, C=O C–C, C–O stretching and OH- bending. The SA microbeads containing aceclofenac and dexibuprofen caused a shift in the O–H, COO– (asymmetric), and COO–(symmetric) stretching peaks to lower wave numbers due to the presence of an ionic bond between the calcium ion and carboxyl groups of sodium alginate\(^{154}\).

The characteristic IR peaks of Clay (HAS) were observed at 2924.18, 2336.84, 1628.19, 1503.56 and 818.81 cm\(^{-1}\) and physical mixture of dexibuprofen and clay (HAS) IR peaks at 2956.97, 2640.01, 1708.99, 1509.35 and 861.24 cm\(^{-1}\). The addition of HAS in the dexibuprofen-loaded beads caused a change in the carboxylate peaks of SA. The Si–O–Si stretching peak of clay (HAS) at 1015.24 cm\(^{-1}\) became narrow and moved to a lower wave number 1009.15 cm\(^{-1}\) due to the interaction between SA and dexibuprofen.

The characteristic absorption peaks of LBG were observed in the region of 3186.4, 2925.6, 1705.6, 1528.4, 1450.5, 1350.25 and 774.5 cm\(^{-1}\). The characteristic peak at 3186.4 cm\(^{-1}\) represents O–H stretching vibration and 2925.6 cm\(^{-1}\) corresponding to C–H stretching of the –CH\(_2\) groups. The peaks found at 1705.6 to and 1528.4 cm\(^{-1}\) represent the ring stretching of galactose and mannose of LBG. On other hand, the peaks observed in the region of 1350.25 to 1450.5 cm\(^{-1}\) are corresponding to symmetrical deformations of CH\(_2\) and COH groups. The lower wave
number observed at around 774.5 cm\(^{-1}\) are due to ring stretching and ring deformation of \(\alpha\)-D-(1-4) and \(\alpha\)-D-(1-6) linkages. The physical mixture of aceclofenac sodium and LBG characteristic peaks at wave numbers 3124.2, 2856.5, 1710.8, 1538.2 and 774.5 cm\(^{-1}\) corresponding to NH- stretching, C=O stretching of –COO and –COOH groups respectively.

FTIR spectra of HPMC K15M show the characteristic peaks at wave numbers 2815.33, 1706.56, 1485.32, 1385.36 and 782.60 cm\(^{-1}\) corresponding to C-H stretching, C=C stretching in the aromatic ring and peaks at 1485.32, 1385.36 cm\(^{-1}\) which can be assigned to the C-H deformation. FTIR spectrum of physical mixtures of aceclofenac sodium and HPMC K15M showed absorption peaks at wave numbers 2932.86, 1718.33, 1507.52, 1252.82 and 748.45 cm\(^{-1}\) corresponding to C-H stretching and C-H deformation. The FT-IR spectra of the physical mixture of dexibuprofen and HPMC K15M shows the characteristic peaks at wave number 3185.50, 2985.35, 1735.25, 1495.65, 1405.25 and 715.36 cm\(^{-1}\) corresponding to O-H, C-H, C=O C-C, C-O stretching and OH- bending.

The characteristic absorption peaks of Kollidon SR were observed in the region of 3315.42, 2996.52, 1745.55, 1695.30 1480.50, 1375.66 and 694.25 cm\(^{-1}\) and represents O-H, C-H, C=O stretching vibration. The peaks observed at 1375.66 corresponding to CH\(_2\) wagging and 694.25 cm\(^{-1}\) represents skeletal C–H rocking. The physical mixture of Kollidon SR and dexibuprofen shows characteristic absorption peaks in IR-spectra at wave numbers 3319.50, 2909.11, 1710.92, 1585.5, 1418.32 and 669.53cm\(^{-1}\) corresponding to O-H, C-H, C=O stretching vibration and C-H deformation.

The IR spectra’s of individual polymer and physical mixtures of drug and polymer were compared with the spectra of the pure drugs. The spectral data suggests
that the major peaks for drugs are obtained as nearer value and there were no considerable changes in IR peaks in all physical mixtures of drug and polymer. This indicates that the drugs were molecularly dispersed in the polymers or in drug loaded formulations thus thereby indicating the absence of any interactions.

6.1.4 Differential scanning calorimetry (DSC)

DSC is a well-established method often used as a qualitative technique to characterize physical and chemical changes in either enthalpy or heat capacity of a crystalline drug in the polymer matrix during the manufacturing process.

From the figures 5.1.23 to 5.1.26, the pure aceclofenac sodium shown a sharp endothermic peak at 158.50°C followed by corresponding meting point. However, the drug-loaded microbeads and matrix tablets were shown a sharp endothermic peak in between the range of 193 to 195°C. The extra obvious peak of the pure aceclofenac sodium at 158.50°C was not observed in the thermograms of prepared microbeads and matrix tablets. The results indicate that there were no changes in thermal behavior of drug during the manufacturing process and it was molecularly dispersed in different hydrogel matrices.

From the figures 5.1.27 to 5.1.31, the thermogram of pure dexibuprofen shows a sharp endotherm at 55.50°C followed by 400°C corresponding its meting point. The drug loaded formulated microbeads and matrix tablets show a sharp endotherm at 400°C. The distinctive endothermic peak of dexibuprofen at 55.50°C was absent in the formulated microbeads and matrix tablets. There was no appreciable change in the melting endothermic peaks of drug loaded formulations compared to pure drug. It may indicate that there are no changes in thermal behavior of drug in the manufacturing process and the drug was molecularly dispersed in different hydrogel matrices.

6.1.5 X-Ray Powder diffractometry (X-RPD)
The distribution of the drug in the polymeric matrix is very important to maintain the theoretical potency and stability in the manufacturing process. However, the drug can crystallize during the formulation resulting decreased aqueous solubility rate due to its polymorphic changes such as particle size, shape, density, melting point etc. The XRPD is an important technique in pharmaceutical field because to investigate the fundamental physical features about the crystalline nature of solid substances. The X-ray powder diffraction patterns of pure drug were compared with drug-loaded microbeads and matrix tablets. The XRPD scan of plain aceclofenac sodium and dexibuprofen shown sharp intense peaks of crystallinity whereas the drug-loaded microbeads and matrix tablets exhibited halo pattern with less intense followed by denser peaks. This result indicates that decrease in the degree of crystallinity due to partial amorphization of the drug in the polymeric matrix. Thus, there were no appreciable changes in the crystallinity of drug during the manufacturing process.

6.2. Characterization and evaluation of aceclofenac sodium microbeads

The certain manufacturing variables of the formulated batches of drug-loaded microbeads were investigated i.e. concentration of drug to polymer ratio, concentration calcium chloride, curing time, stirring speed on physicochemical properties and in-vitro drug release of microbeads..

Prior to develop the dosage forms, it is necessary to control the manufacturing variables to produce optimum formulated products. Suppose, aceclofenac sodium loaded microbeads are formulated with 0.5 percent of SA which are cured for 2 hours at 2000 rpm in 0.5 percent of calcium chloride solution. The formed microbeads were not spherical and had a flattened base at the points of contact with the drying vessel. This indicates that at low sodium alginate concentration the particles were composed
of loose net-works structure collapsed during drying. Increase in the concentration of SA tends to make the particles more spherical and formed dense matrix structure which prevents collapse of microbeads in the curing process. But increasing the concentration of drug to polymer ratio above 1:15 forming high viscous polymer dispersion and did not pass easily in the needle during the manufacturing process. Moreover, the formation of small tail at one end of beads which significantly affects the flow properties and particle size distribution. This explains that the optimum concentration of drug to polymer ratio [1:10], calcium chloride [4%w/v], cross-linking time [2 hours] and stirring speed [2000 rpm] could influences the properties of microbeads such as size, average diameter, recovery, encapsulation efficiency, size distribution swelling behavior and the release characteristics.

6.2.1 Micromeritic properties of aceclofenac sodium microbeads

The rheological parameters like angle of repose, bulk density and tapped density of all microbeads confirms better flow and packaging properties. All the formulations showed excellent flowability represented in terms of angle of repose (<40°)\textsuperscript{20}. The sodium alginate concentration has significant positive effect on the angle of repose. The particle size increases by increasing the concentration of SA as a result decreases the angle of repose. However, higher calcium chloride concentration, cross-linking time and stirring speed could influence the formation of smaller beads and show an increased angle of repose. Bulk and tapped density of microbeads obtained a good acceptable range indicating good packability. The density of the microbeads increases by increasing the concentration SA. This indicates that the beads formed at high polymer concentration are more compact and less porous than those prepared at low polymer content. Carr’s index and Hausner’s ratio values are obtained in
acceptable ranges that explain the formulated microbeads have excellent compressibility and good flow properties.

6.2.2 Morphology of aceclofenac sodium by SEM

The external morphology of formulated microbeads before and after drying is represented photographically in figure 5.2.1 and 5.2.2. The SEM of the dried drug-loaded microbeads and their surface morphology are given in figure 5.2.3 to 5.2.6. Form the electron micrographs, clearly explains the drug loaded microbeads were discrete and spherical in shape with a rough outer surface, bigger crevices, pores and visible large wrinkles formed in the internal structure. The overall surface morphology of the SA microbeads explains that the formation of large matrix bridges contribute the formation of thick hydrodynamic layer which influences the slower release of the incorporated drug.

6.2.3 Effect of drug to polymer ratio:

The table 5.2.2 explains that the percentage yield of microbeads was observed in decreasing order from 88.30 to 73.40. It clearly explains that by increasing polymer ratio in the formulations significantly lower the product yield, due to the formation of high viscous polymer dispersion which may be lost during manufacturing process.

The mean particle size of drug loaded microbeads (F1-F5) was obtained in the range of 596.45±1.04 to 880±1.23. It was found that the particle size distribution of all the formulations was within a narrow size but the mean particle size was different among the different batches. The results indicate that the mean particle size was considerably increased with increase in the amount of SA due increase in relative viscosity, the formation of large droplets during addition of polymer solution in to the calcium chloride solution.
The actual drug content in formulations F1 to F5 was obtained in the range of 56.92± 0.39 to 72.60±0.62 mg/100mg and the percentage of drug entrapment efficiency was found in the range of 63.24±0.66 to 98.90±0.86 respectively. The actual drug content was high by increase in the concentration of SA due to formation of high viscous gel which leads to better precipitation of polymer at the external phase of the droplets. The entrapment efficiency increased progressively with increase in the concentration of SA. This result indicates that the formation of larger beads entrapping greater amount of aceclofenac sodium due to the greater availability of active calcium binding sites in the polymeric chains. In other hand, the greater degree of cross-linking reaction takes place consequently to form dense matrix which minimizes the loss of drug in curing media.

The “Swelling-Dissolution-Erosion” process is highly complex. In systems based on SA cross-linked with calcium chloride, the osmotic pressure gradient that exists between the alginate gel and the environment comprises an important factor in the swelling process. The swelling ratio of the beads is dependent on the pH of the solution\textsuperscript{155}.

The figure 5.2.7 and 5.2.8 explains that the increase in the drug to polymer ratio significantly increases the swelling behavior of microbeads in higher pH level. Generally, SA is a polyelectrolyte that can exhibit swelling depends on the ionic strength, ionic composition and pH of the medium\textsuperscript{156}. The low swelling observed in SGF pH1.2 is probably due to the formation of insoluble alginic acid by proton-calcium ion exchange to form thick gel polymeric net-work which reduces the water penetration into interior parts of beads. The swelling of beads was ultimately increased in pH 4.8 and pH6.8 at the end of 4 hours. It has been reported that the swelling can be enhanced by relaxation of the cross-linked polymeric net-work due to the
interchanging of Ca$^{2+}$ ions within the beads by the presence of phosphate ions in higher pH level. Thus, under acidic conditions swelling of the calcium alginate beads occurs very slowly and followed by increasing under neutral pH conditions.

The effects of drug-polymer ratio on aceclofenac sodium release from different batches (F1-F5) of microbeads are reported in table 5.2.4 and figure 5.2.9. The in-vitro drug release from alginate microbeads was pH dependent and the drug release from the formulations F1 to F5 was observed negligible drug release in SGF pH 1.2 (<5%w/w) this may be due to the stability of alginate at lower pH and conversion of calcium alginate to the insoluble alginic acid$^{157}$. On the other hand, the solubility of aceclofenac sodium is very less in this environment and may affect the drug release. The maximum amount of aceclofenac sodium release (>70% w/w) from the microbeads in pH 6.8 PBS at 6 hours followed by sustaining in pH 7.4 PBS up to 12 hours. As the drug to polymer ratio increases the release rate from the microbeads decreases in higher pH levels due to the extent of swelling and the formation of thick gel layer which acts as a barrier for the penetration of water retarding the diffusion of drug from the swollen alginate beads. However, the steady state drug release was achieved after an initial lag time and it was directly proportional to the concentration of SA. The first phase might be for the negligible dissociation of SA beads in PBS mainly based on drug diffusion through the small pores and cracks. (Reported in SEM figures) The second phase exhibited a burst-like release pattern, which was accompanied by the polymeric relaxation and erosion at alkaline pH level. The contents are released in a sustained manner by both diffusion and slow erosion of polymer matrix.
6.2.4 Effect of stirring speed

From the table 5.2.5, the total percentage yield of drug loaded microbeads decreases by increasing the stirring speed, because some of the microbeads collapse at higher stirring speed due to high turbulence caused attrition between impeller of stirrer and container wall. The size of the spherical matrix could be easily controlled by varying the stirring speed of the system. The mean particle size was observed in the range of 784.60±1.08 to 716.80±0.96 µm. The mean particle sizes of microbeads were tremendously decreased with increasing the rotational speed. At a stirring speed of 500 rpm, the mean particle diameter and the size distribution of the beads increased significantly. This low stirring speed might have decreased the uniformity of the mixing force throughout the polymeric mixture and the particles were found to settle at the bottom of vessel resulting in a wider diameter of the final beads.

Consequently at higher stirring speed at 2500 rpm increased mechanical shear might have been influenced in the formation of lesser diameter of microbeads. The present research protocol fixed the stirring speed higher than 2000 rpm did not reduce the mean diameter, because high turbulence caused frothing and adhesion to container wall which changes the shape and size of the microbeads reported in SEM micrographs.

Increasing the stirring speed 500 to 2500 rpm the actual drug content and percentage of drug entrapment efficiencies were found to be in the range of 64.90 ± 0.78 to 69.54 ± 0.64 mg/100mg and 87 ± 0.90 to 90.56 ± 0.36 respectively. There was no significant change on encapsulation efficiency of drug with increase in the speed of agitation.

The figure 5.2.11, illustrates that the aceclofenac sodium release from the microbeads in SGF pH 1.2 for initial 2 hours was obtained as less than 10%w/w due to
poor solubility of drug in this environment. The maximum amount of aceclofenac sodium release (>70 % w/w) from the microbeads in pH 6.8 PBS at 6 hours followed by sustaining in pH 7.4 PBS up to 12 hours. When the stirring rate was increased, the drug release was found to be faster. This may be due to the reduction in the size of microbeads, which provide a large surface area to increase the drug release.

6.2.5 Effect of crosslinking agent

From the table 5.2.8, the total percentage yield of aceclofenac sodium-loaded microbeads (F11- F15) was observed in the range of 72.40 to 77.60. The concentration of calcium chloride increases from 1 to 5%w/v in the curing media which gradually increases the yield. Increasing the concentration of calcium chloride in the curing media progressively decreases the mean particle size and found in the range of 746.60 to 688.56±1.2µm. It has been stated that when a drop of SA solution comes in contact with calcium ions gelation occurs instantaneously and also Ca^{2+} ions penetrates into interior of droplets and ultimately water is squeezed out resulting in contraction of beads\textsuperscript{158}.

The actual drug content and entrapment efficiency were gradually increases in higher levels of calcium chloride in the curing media. From the results indicate that by increasing calcium chloride concentration produces beads with higher levels of Ca^{2+} ions which increase the compactness and forms insoluble dense matrices resulting in more drug entrapment in the microbeads. On other hand further increase in the concentration of calcium chloride above (5%w/v) do not enhance the drug loading. This could be due to probably saturation of Ca^{2+} ions involved in crosslinking process.

The figure 5.2.12, Illustrates that increasing in the concentration of calcium chloride (F11-F15) produces the beads with higher levels of Ca^{2+} ions which are
reduces the swelling of the beads in acidic medium. The swelling behavior of beads in pH 4.8 and 6.8 PBS gradually increases due to ionic exchange between the phosphate ions in the buffer and higher level of Ca$^{2+}$ ions within the beads.

The figure 5.2.13, illustrates that the aceclofenac sodium release from the microbeads in SGF pH 1.2 for initial 2 hours was obtained as less than 10% w/w due to poor solubility of drug in this environment. The maximum amount of aceclofenac sodium release (>70 % w/w) from the microbeads in pH 6.8 PBS at 6 hours and steady state release was observed in pH7.4 PBS up to 12 hours. The results indicate that rate and extent of drug release decreases gradually with increase in the concentration of calcium chloride, because a tight gel junction is formed between the residues of alginate with calcium ions which take longer time to diffuse the drug into bulk of the medium.

6.2.6 Effect of crosslinking time

From the table 5.2.11, the total percentage yield of aceclofenac sodium-loaded microbeads was observed in the range of 75.65 to 77.70. Based on the resulted data it can be concluded that the effect of crosslinking time on the yield of microbeads is insignificant.

The effect of cross-linking time at a particular stirring speed on particle size of microbeads was recorded and by increasing the cross-linking time decreases the mean particle size, possibly due to more Ca$^{2+}$ ions available for binding with carboxylic groups of alginate that influences compactness of particulate system formed smaller size of beads. Moreover, batch F-11 prepared with 0.5 hours cross-linking time was noted insufficient to cross-linking process and formed irregular size microbeads.

The variation in the cross-linking time affects actual drug content and drug entrapment efficiencies of the formulated microbeads. The prolonged cross-linking
time results decrease in the drug loading and entrapment efficiency, since the solubility of aceclofenac sodium was slightly higher in calcium chloride than in distilled water. Prolonged exposure in the curing medium caused greater loss of drug through weakly cross-linked SA beads. However, the constant drug loading was achieved at 2 hours but there were no significant changes of drug entrapment efficiency observed at above 2 hours curing time. This could be due to the formation of tight junction between calcium ions and the active sites on the guluronic acid chain. Consequently, the drug was entrapped in highly bound calcium alginate matrix which prevents the further diffusion of drug in the curing medium.

The figure 5.2.14, explains that the prolonged cross-linking time shows maximum swelling with increased pH level. The result suggests that the maximum extent of cross-linking influences the formation of compact beads, which are rehydrated to a greater extent. The sequestering action of phosphate ions in higher pH media on Ca$^{2+}$ ions may have been contributed to the swelling equilibrium of SA microbeads.

The figure 5.2.15, illustrates that the aceclofenac sodium release from the microbeads in SGF pH 1.2 for initial 2 hours was obtained less than 10%w/w due to poor solubility of drug in this environment. The maximum amount of aceclofenac sodium release (>70 % w/w) from the microbeads in pH 6.8 PBS at 6 hours and steady state release was observed in pH7.4 PBS up to 12 hours. An increase in the cross-linking time from 0.5-2.5 hours progressively decreases drug release from the microbeads due to formation of tight mesh alginate net-work by penetration of Ca$^{2+}$ ions to the interior of the microbeads. Faster drug release was observed with 0.5 and 1 hours which can be attributed to the poor binding of drug into the polymer matrix.
Increasing the cross-linking time more than 2 hours there was no significant change in the drug release.

6.2.7 Kinetics of drug release;

All the formulations F1 - F5 showed first-order mechanism with highest correlation coefficient 0.9814 and 0.9430 compared to Higuchi matrix and followed by zero-order. The obtained data of in-vitro drug release was also correlated to Korsemeyer – Peppas model in order to find out coefficient of correlation (r) and diffusion coefficient (n) value, which describes the drug release mechanism. The kinetic data was best fitted to Korsmeyer and Peppa’s model and good regression coefficient was observed. The values of coefficient of correlation (r) were obtained in the range of 0.91624 to 0.99672. The values of diffusion coefficient (n) of formulations F1 to F5 were in the range of 0.4652 to 1.1525 indicating that the drug release from the microbeads exhibited nearer to zero-order kinetics followed by case-II transport.

6.2.8 Stability studies

The stability study was conducted at 25\(^0\)C/ 60% RH, 40\(^0\)C / 75% RH for 6 months. Overall, results from the stability study indicates that the capsules were physically stable but the percentage of drug content at 40\(^0\)C / 75 %RH was slightly reduced to 92.64 after 6 months. After 6 months the discoloration, aggregation and changes of sphericity of the beads inside the capsules were noticed. Good stability was observed at low temperature for more than 6 months.

6.3 Characterization and evaluation of dexibuprofen microbeads

Twenty batches of M1 to M20 dexibuprofen loaded microbeads were prepared by sodium alginate (SA) as drug retardant and clay (HAS) as filler and investigated the physicochemical properties and in-vitro drug release potential.
6.3.1 Micromeritic properties of dexibuprofen microbeads

The effects of various process and formulation parameters on the micromeritic properties of drug loaded microbeads were evaluated. The rheological parameters of all the batches (M1-M20) are depicted in table 5.3.1 and 5.3.2. The results indicate that all the formulations showed excellent flowability represented in terms of angle of repose (<40\(^{0}\)), compressibility index (<30) and Hausner ratio (<1.36). The SA concentration has a significant positive effect on the angle of repose due to the formation of uniform particle size and shapes. However, higher calcium chloride and clay (HAS) concentration had influenced the formation of smaller beads and resulted a decreased angle of repose. Bulk and tapped density of beads with clay shown good acceptable range which indicating good packability. The density of the microbeads was high with increase in the concentration of clay (HAS) due to formation of compact matrix and less porous than those prepared by sodium alginate.

6.3.2 Scanning electron microscopy (SEM)

The external morphology of formulated microbeads before and after drying is represented photographically in figure 5.3.1 and 5.3.2. The SEM photomicrographs of the dried drug-loaded microbeads and their surface morphology are shown in the figure 5.3.3 to 5.3.6. Morphology of the drug loaded SA microbeads was discrete and spherical in shape with a rough outer surface and visible large wrinkles and pores because of the surface-associated crystals of drug. No signs of deformation were observed in the SEM pictures of SA /clay (HAS) composite microbeads were more spherical, smooth sandy appearance and without agglomeration, the drug crystals on the surface are absent due to complete entrapment of drug crystals inside the matrices.
6.3.3 Effect of drug to polymer ratio

From the depicted table 5.3.3, the total percentage yield of microbeads was observed in the decreasing range from 78.60 to 65.30. Increasing the SA in the emulsion will slightly decrease the yield of microbeads due to formation of high viscous dispersion which losses during the manufacturing process. The mean particle size of drug loaded microbeads was obtained in the range between 396 to 686 µm. The results indicate that the increase of SA concentration increases the mean particle size of microbeads. This could be attributed to an increase in relative viscosity at higher concentration of SA, formation of a large droplet during addition of polymer dispersion into the crosslinking solution.

As shown in table 5.3.3, Actual drug content in the microbeads (M1-M5) was evaluated and found to be in the range of 33.80 to 49.40 mg/50mg. The SA concentration increases consequently, the actual drug loading is high due to the increase of hydrophobicity, leading to better adsorption of the drug loaded emulsion at the boundary phase of the droplets. The percentage of drug entrapment efficiency was found to in the range of 67.60 to 97.80. The increase in the proportion of SA in the microbeads influences the drug entrapment efficiency. This may be due to the availability of more binding sites for the drug molecules in the SA matrix.

The figure 5.3.7, illustrates that the SA can exhibit swelling properties that are very sensitive to the pH, ionic strength and ionic composition of the medium. The low swelling in SGF pH1.2 is probably due to proton-calcium ion exchange forming insoluble alginic acid to form tightening of the gel mesh work. The magnitude of water uptake was increased at higher pH range due to the presence of phosphate ions in higher pH range which displaces the Ca^{2+} ions within the beads and leads to relaxation of the cross-linked polymeric network.
The figure 5.3.8, explains that the proportional increase in the concentration of SA significantly decreases the release of drug from the microbeads. The percentage of drug release from dexibuprofen beads was obtained in the range of 44.25 to 28.15 for initial 2 hours. This may be due to conversion of Ca-alginate into insoluble alginic acid to form rigid gel mesh work. Moreover, dexibuprofen was partially soluble in acidic pH may be affects the drug release. The percentage of dexibuprofen release from the microbeads in pH 6.8 PBS at 6 hours and pH 7.4 PBS at the end of 12 hours is shown in the range from 64.83 to 50.79 and 98.65 to 88.40 respectively.

The result explains that the drug release from the microbeads exhibited as biphasic drug release patterns behavior, i.e. an initial rapid release (burst effect) phase was followed by a second, slower drug release phase. The first phase might be disintegration of alginate in the presence of surfactant and co-surfactant mainly based on drug diffusion through the small pores and cracks. The second phase exhibited slow release pattern due to the polymer eroded at alkaline pH and the contents released in a sustained manner by both diffusion and slow erosion of polymer matrix.

6.3.4 Effect of concentration of clay [HAS]

Five batches M6 to M10 of dexibuprofen loaded SA / clay composite microbeads were prepared corresponding to 1, 2, 4, 6, and 10% w/v of clay and investigated the effect of concentration of clay on physicochemical properties of microbeads.

The table 5.3.6 explains that the increase in the concentration of clay slightly decreases the yield of microbeads and found in range from 75.80 to 70.50. The mean particle size of SA /clay composite microbeads decreases significantly by increasing the concentration of clay (HAS). It has been stated that when clay-HAS forms
compact matrices with microemulsion of SA solution and comes in contact with calcium ions, gelation occurs instantaneously and forms smaller size of microbeads. By increasing the concentration of clay influences the actual drug content in the range of 40.30 to 49.40 mg/50mg. The percentage of drug entrapment efficiency was found in the range of 80.60 to 98.80. The increase in the quantity of clay (HAS) enhances higher drug entrapment efficiency. This indicates that the interaction of clay (HAS) with SA forms a dense matrix barrier which prevents leakage of drug from the microbeads during the curing medium.

The figure 5.3.9, illustrates that increasing the proportion of clay (HAS) in the formulation tremendously increases the swelling behavior at higher pH level. This is basically due to the adsorbent properties of clay decreases the surfactants concentration in SA microemulsion and more Ca^{2+} ions interact with silicate ions of clay in the crosslinking process which displaces the polymeric net-work to enhance water penetration into interior region of microbeads and swell gradually for prolonged period of time\textsuperscript{160}

The figure 5.3.10, describes that the concentration of clay increases the initial burst release of the beads and decreases significantly in SGF pH 1.2 due to formation of SA/clay dense matrix. The percentage of drug release from SA / clay (HAS) composite beads at 6 hours in pH 6.8 PBS and pH 7.4 PBS at the end of 12 hours was decreased from 58.11 to 44.72 and 93.52 to 80.96 respectively. It has been stated; that the drug diffusivity of the composite gel affected by the denser matrix structure and created by the interaction between SA and clay (HAS). The denser matrix structure had higher tortuosity, resulting in slower drug diffusion through water filled channels in the clay/alginate gel at higher pH level\textsuperscript{161}. 
6.3.5 Effect of crosslinking agent

From the depicted table 5.3.9 reveals that the total percentage yield of microbeads corresponding to different concentration of calcium chloride in the curing media ranged from 82.60 to 74.60. The mean particle size of drug loaded microbeads was obtained in the range of 734 to 622µm. The crosslinking agent gives a dual effect on microbeads size. Initially the size was found to reduce on increasing the strength of crosslinking agent due to higher Ca\(^{2+}\) ions, penetrates into interior of droplets and water is squeezed out of the interior of droplets resulting the formation of intact beads.

Increasing the calcium chloride concentration from 1 – 8 %w/v the actual dexibuprofen content and percentage entrapment efficiency was found to be in the decreasing range of 47.10 to 41.70 mg/50mg and 94.20 to 83.80 respectively. From the results, it is obvious that increasing calcium chloride concentration produces beads with higher levels of Ca\(^{2+}\) ions to enhance bead shrinkage during gelation of SA and calcium chloride. This shrinkage leads to a shorter path length for drug leakage and therefore higher drug losses in the curing media with high level of calcium chloride.

The figure 5.3.11, describes that the swelling behavior of sodium alginate/clay composite microbeads hardened by using different concentration of calcium chloride in the curing media which predominately decreases the swelling ratio in higher pH levels. This may be due to relaxation of SA/clay matrix by surfactants of microemulsion concentrates present in the microbeads.

The figure 5.3.12 explains the percentage of in-vitro release of dexibuprofen from the batches of M11 to M15 in SGF pH1.2 for initial 2 hours was observed in the range of 20.08 to 12.05, pH6.8 PBS at 6 hours 58.61 to 40.57 and in pH7.4 PBS was 93.23 to 79.30 at the end of 12 hours respectively. The results indicate that rate and extent of drug release decreases significantly with increasing the concentration of
calcium chloride, because sodium alginate is a hydrophilic polymer forms a tight junction polymeric net work with calcium ions, the clay residue will entrapped in the interior gel layer forms dense matrix which diffuse the drug from the gel layer slowly for extended time period.

6.3.6 Effect of stirring speed

The mentioned results in table 5.3.12, illustrates the percentage of yield decreases with higher level of stirring speed in the formulation process due to inefficient cross linking produces less compact beads which are collapsed during higher rotational speed. The significant decrease in the average size of the microbeads was observed with increase in the rotational speed found in the range of 721 to 613 µm. At a stirring speed of 500rpm, the mean particle diameter predominately increased because the stirring speed might have been decreased the uniformity and the particles were found to settle at the bottom of vessel resulting in a wider diameter of the final beads. Consequently at higher stirring speed increases the mechanical shear which might have influenced the formation of smaller diameter beads.

The actual drug content and percentage of drug entrapment efficiency were observed in the range of 47.10 to 41.70mg/50mg and 94.20 to 83.80 respectively. This explains that the variation of stirring speed does not show any significant changes on drug loading and encapsulation efficiency in the microbeads.

The figure 5.3.13, explains that the swelling behavior of sodium alginate/clay composite microbeads slightly changes at higher pH levels with increase in the rotational speed.

The figure 5.3.14 reveals that the percentage of drug release in SGF pH1.2 for initial 2 hours was observed in the range of 12.03 to 20.08, pH 6.8 PBS at 6 hours 45.24 to 62.89 and in pH 7.4 PBS was 80.99 to 94.63 at the end of 12 hours
respectively. The effect of stirring rate on the in-vitro release of dexibuprofen was found to be significant. A batch (M16) prepared at 500 rpm stirring speed shown delayed release compared to higher rotational speed batches. This may be due to the smaller particle size of microbeads are formed in higher stirring speed in the size of microbeads, they provide a large surface area to increase the drug release.

**6. 3.7 Effect of crosslinking time**

The table 5.3.15 reveals the total percentage yield of microbeads was obtained in the range of 73.10 to 70.70. The effect of cross-linking time increases the mean particle size of microbeads significantly decreases and found in the range of 744 to 632µm. This may due to high shear force which causes abrasive action in the container wall at prolonged curing time forms small size spherical beads.

The actual drug content and drug entrapment efficiency of the microbeads (M 21-M 25) obtained in the range of 48.60 to 43.20 and 97.20 to 86.40 respectively. The results explain that actual dexibuprofen and percentage of loading efficiency of microbeads slightly decreases with increasing the crosslinking time. The percentage of loading efficiency increases in 0.5 hours crosslinking time but above 2.5 hours there were no significant changes due to the saturation of Ca\(^{2+}\) ions binding and cross-linking process.

The depicted figure 5.3.15, explains that at prolonged cross-linking time in the system, the microbeads show increased swelling ratio values in higher pH levels. These results may be due to the maximum extent of cross-linking process produce compact beads, which are rehydrated gradually at greater extent. In case the batch M-21 with short crosslinking time causes lower rehydration of beads may be correlated to incomplete cross-linking of SA/clay with calcium chloride.
The figure 5.3.16 explains the effect of crosslinking time on dexibuprofen release from the microbeads. The percentage of drug release from the formulations (M20-M25) obtained in SGF pH1.2 for initial 2 hours in the range of 29.12 to 17.63, pH 6.8 PBS at 6 hours 61.12 to 40.56 and in pH 7.4 PBS was 93.28 to 81.33 at the end of 12 hours respectively. The results indicate that the increase in the cross-linking time from 0.5-2.5 hours significantly decreased the drug release due to penetration of Ca$^{2+}$ ions to the interior surface induces the formation of thick gel net-work. Faster drug release was observed from the microbeads stirred by 0.5-1 hour crosslinking time during the manufacturing process. This indicates that the poor binding of drug into polymer matrix and also incomplete gelling of sodium alginate. No significant changes were observed in the amount of drug release by increasing the cross-linking time more than 2 hours.

6.3.8. Analysis of various drug release kinetics

The In-vitro drug release of all the batches M1 to M10 of microbeads was treated with various kinetic models. The mechanism of drug release was determined by using PCP-DISSOv2.08 software. The results suggest that the release mechanism from the microbeads M1 to M5 shows first order kinetics followed by hugging matrix. The formulations of drug loaded sodium alginate/clay composite microbeads (M6-M10) show the mechanism of drug release followed to nearer zero-order and hugging matrix kinetics. The diffusion co-efficient values were in the range 0.9166 to 1.1779. This indicates that the drug release from microbeads by both diffusion and erosion mechanisms followed by super case-II transport due to control of swelling and diffusion of sodium alginate/clay (HAS) matrices.
6.3.9 Stability studies

Stability studies is extremely important key parameter to accurately predict the shelf stability of new product from accelerated storage data to preserve the product at normal storage for extended periods of time. The formulations were subjected to stability studies at 5°C, 25°C/60% RH and 40°C/75% RH for 6 months. Overall, results from the stability studies indicated that the capsules containing dexibuprofen microbeads (M3) were physically stable but percentage of drug content at 40°C/75% RH was slightly reduced after six months. The microbeads (M8) capsules are very stable at higher temperature/humidity condition; there were no significant changes in drug potency because the addition of clay increases the stability. More than six months dexibuprofen-loaded microbeads inside the capsule at room temperature change the color, sphericity and also decreases flow properties.

6.4 Characterization and evaluation of aceclofenac sodium matrix tablets

In the present study, sustained release matrix tablets of aceclofenac sodium were prepared by using LBG and HPMC K15M as a hydrophilic swellable polymers and Avicel PH101 (MCC), Pharmatose 200M (lactose monohydrate) as diluent. The experimental results are mentioned in the chapter 5 and section 5.4. In this section, discussion of experimental results such as micromeritic properties of precompressed granules and physical properties of compressed tablets like hardness, weight variation, friability, drug content uniformity, swelling properties, in-vitro drug release profiles and stability studies.

6.4.1 Micromeritic properties of precompressed granules

The sustained release aceclofenac sodium tablets were prepared by wet-granulation technique. The granules were compressed into tablets, prepared according to the formula given in the tables 4.6 to 4.9.
From the table 5.4.1, the granules of different formulations were evaluated for angle of repose, loose bulk density, tapped bulk density, carr's index, hausner’s ratio. Increasing the concentration of polymer and diluent ratio decreases the angle of repose. The angle of repose (<30) indicates good flow properties of the granules due to formation of high compact matrix in the granulation process. The bulk and tapped density values were obtained in the acceptable range indicating good packability of the granules in the tablet die cavity and minimizes weight variation of compressed tablets. Compressibility index and hausner’s ratio values were obtained in the range 18.04 to 8.35 and 1.22 to 1.08 respectively. This indicates the prepared granules have excellent compressibility and good flow properties to get acceptable hardness to the tablets.

6.4.2 Effect of locust bean gum and Avicel PH101 (AF-1 to AF-5)

The depicted table 5.4.2, illustrates that the weight variation of the matrix tablets was observed within pharmacopeia limit complied below ±5% w/w of standard deviation from the average. However, the average weight of the matrix tablet of the batch AF-3 was obtained very nearer to theoretical weight. The polymer and diluent ratio influences the weight variation of formulated tablets. The hardness of the formulated tablets obtained within the acceptable range of 5.6 to 6.6 kg/cm² followed by decreasing the percentage of friability except the batch AF-1. This indicates the low concentration of LBG used in the formulation decreases the mechanical strength due to the formation of loose matrix with higher concentration of Avicel pH 101. The percentage of aceclofenac sodium content in the tablets of batches AF-1 to AF-5 was observed in the range of 84.76 ± 0.6 to 93.42 ± 0.24 respectively. It was stated that the percentage of drug content increases by mainly increasing the concentration of
LBG entrapped more quantity of drug due to formation of dense matrix during compression.

The figure 5.4.6 reveals that the percentage of swelling ratio of aceclofenac sodium matrix tablets containing various concentrations of LBG and AvicelPH10 was obtained in the range of 176.30 to 238.60 at the end of 6 hours. The result suggests that by increasing the concentration of both polymer and diluent could influence the percentage of swelling ratio. The formulation AF-1 containing high level of Avicel PH101 (above 200mg) which shows higher swelling ratio in initial hours followed by erosion at the end of 6 hours due to its fibrous nature Avicel PH101 forms larger pores which rapidly increase the water penetration into the interior layer of polymeric gel network and decrease the resistance of gel layer in higher pH level. The formulation of batch AF-5 containing more concentration of polymer and less concentration of diluent increases the swelling ratio gradually but decreases the erosion. The formulation AF-4 shows steady state swelling ratio for 6 hours represented photographically in figure 5.4.2. This states that the presence of higher level of LBG enhance the gel formation inhibiting water penetration into interior layer of polymeric gel and thus gradually swells with an increased time period.

The depicted results in the figure 5.4.8, illustrates that the aceclofenac sodium release from the matrix tablets in SGF pH 1.2 for initial 2 hours was obtained less than 30%w/w due to poor solubility of LBG and aceclofenac sodium in this environment. The maximum amount of aceclofenac sodium release (>70 % w/w) from the matrix tablets in SIF pH 7.4 at 6 hours followed by sustaining up to 12 hours. The formulated tablets of batch AF-5 were released in sustain manner compared to AF-1 because of optimum concentration in polymer to diluent ratio. By increasing the polymer proportion in the tablets forms a thick hydrodynamic gel layer that take a prolonged
time to diffuse un-dissolved hydrophobic nature of aceclofenac sodium into bulk of the dissolution medium. Thus, the results clearly explain that the polymer and diluent ratio modified the drug release in a sustained manner.

6.4.3 Effect of HPMC K15M and Avicel PH101 (AF-6 to AF-10)

The table 5.4.4, the weight variation of the matrix tablets observed within pharmacopeia limit and complied below ± 5% w/w of standard deviation from the average. The average weight of the matrix tablet of the batch AF-9 was obtained very nearer to theoretical weight. This suggested that by increasing the polymer and diluent ratio influences the formation of uniform size granules which ultimately increases the flow properties and minimizes the weight variation. The hardness of the formulated tablets obtained within acceptable range of 5.5 to 7.1 kg/cm$^2$ followed by decreasing the percentage of friability except the batch AF-6. This indicates the low concentration of HPMC K15M used in the formulation decreases the mechanical strength due to the lower compressibility of Avicel pH 101 forms loose matrix granules. The percentage of aceclofenac sodium content in the matrix tablets of batches AF-6 to AF-10 was found in the range of 88.95 to 96.42 respectively. It was stated, that the percentage of drug content increases with higher concentration HPMC K15M due to formation of dense matrix entrapped more quantity of drug.

The figure 5.4.7, explains that the percentage of swelling increases with an increase in the concentration of polymer and diluent in the matrix system. The percentage of swelling ratio of aceclofenac sodium matrix tablets containing various concentrations of HPMC K15M and AvicelPH10 were obtained in the range of 168.20 to 246.32 at the end of 6 hours. The result suggests that increasing the concentration of both polymer and diluent could influence the percentage of swelling ratio. The formulation AF-6 containing higher level of Avicel PH101 (above 200mg) shows
higher swelling in initial hours followed by erosion at the end of 6 hours because Avicel PH101 is a hydrophilic fibrous excipient which rapidly increases the water penetration into interior layer of polymeric gel network and increases erosion of less viscous gel layer. The formulation AF-9 shows steady state swelling ratio for 6 hours is represented photographically in figure 5.4.3. The formulation of batch AF-10 containing more concentration of polymer and less concentration of diluent increases the swelling ratio gradually but decreases the erosion. This states that the presence of higher level of HPMC K15M enhances the formation of thick gel acts as a barrier inhibiting the water penetration into interior layer of polymeric gel and thus gradually swells with increased time period.

Figure 5.4.9, illustrates that the aceclofenac sodium release from the matrix tablets in SGF pH 1.2 for initial 2 hours was obtained as less than 20 %w/w due to poor solubility of aceclofenac sodium in this environment. The maximum amount of aceclofenac sodium release (>50 % w/w) from the matrix tablets in SIF pH 7.4 at 6 hours followed by sustaining up to 12 hours. The formulation AF-9 gives optimum drug release compared to AF-6. In higher concentration of HPMC K15M in the matrix tablets prolongs the drug release due to formation of thick hydrodynamic diffusion gel layer in the dissolution process that take a prolonged time to diffuse un-dissolved hydrophobic nature of aceclofenac sodium into bulk of the dissolution media.

6.4.4 Effect of Locust bean gum and Pharmatose 200M (AF-10 to AF-15)

Table 5.4.7, illustrates that the weight variation of the formulated matrix tablets was observed within pharmacopeia limit complied below ±5% w/w of standard deviation from the average. The average weight of the matrix tablets of batch AF-14 was obtained very nearer to theoretical weight. This suggests that by increasing the polymer and diluent ratio influences the formation of uniform size granules which
ultimately increases the flow properties and minimizes the weight variation. The hardness of the formulated tablets obtained within acceptable range 6.0 to 6.4 kg/cm² followed by decreasing the percentage of friability. There are no significant changes of hardness with increase in the polymer to diluent ratio. The percentage of aceclofenac sodium content of batches AF-11 to AF-15 was observed in the range of 82.96 to 98.45 respectively. It was stated that the percentage of drug content increases with higher level of LBG forms a compact matrices entrapped more quantity of drug.

Figure 5.4.10, explains that the percentage of swelling increases with increase in the concentration of polymer and diluent in the matrix system. The percentage of swelling ratio of aceclofenac sodium matrix tablets (AF-11 to AF-15) was obtained in the range of 146.50 to 215.30 at the end of 6 hours. The result suggests that increasing the concentration of both polymer and diluent could influence the percentage of swelling ratio. The formulation AF-11 containing higher level of Pharmatose 200M (above 200mg) shows higher swelling in initial hours followed by erosion at the end of 6 hours because Pharmatose 200M is a more water-soluble excipient forms more micro-cavities in polymer matrices which rapidly increases the water penetration into interior layer of polymeric gel network initially and decreases the resistance of LBG gel layer in later time period. The formulation of batch AF-15 containing more concentration of polymer and less concentration of diluent decreases the swelling ratio because the hydrophilic nature of LBG forms less resistance gel layer with low concentration of diluent. The formulation AF-14 shows steady state swelling ratio for 6 hours is represented photographically in figure 5.4.4. This states that the presence of optimum level of LBG and Pharmatose 200M enhances the formation of thick gel acts as a barrier inhibiting the water penetration into interior layer of polymeric gel, thus gradually swells with an increased time period.
The figure 5.4.12 explains that the increase in the concentration of polymer and diluent ratio significantly retards the release of aceclofenac sodium from the matrix tablets. The drug release in SGF pH 1.2 for initial 2 hours was found to be less than 15% due to poor solubility of aceclofenac sodium and LBG in this environment. The drug release significantly increases in SIF pH 7.4 at 6 hours followed by sustaining up to 12 hours. The formulation AF-14 shows an optimum drug release compared to AF-11. The higher amount of Pharmatose 200M interact with lower amount of LBG and do not form thick polymeric gel layer which rapidly diffuses the drug at higher pH level. On other hand, increase the concentration of LBG in the matrix tablets which prolongs the drug release due to formation of thick hydrodynamic diffusion gel layer in the dissolution process. Thus, the drug release from the swellable matrices altered by both LBG and Pharmatose 200 M ratios present in the matrix system.

6.4.5 Effect of HPMC K15M and Pharmatose 200M (AF-16 to AF-20)

Table 5.4.10, illustrates that the weight variation of the formulated matrix tablets was observed within pharmacopeia limit complied below ±5% w/w of standard deviation from the average. The average weight of the matrix tablets of the batches AF-19 and AF-20 was obtained very nearer to theoretical weight except the batch AF-16. This describes that the increase in polymer and diluent ratio influences the formation of uniform size granules which increases the flow properties and minimizes the weight variation. The hardness of the formulated tablets was obtained within acceptable range 6.0 to 6.4 kg/cm² followed by decreasing the percentage of friability. The percentage of aceclofenac sodium present in the tablets was observed in the range of 90.66 to 99.42. The percentage of drug content increases with increase in the
concentration of HPMC K15M and Pharmatose 200M due to formation of compact matrix entrapped more quantity of drug.

Figure 5.4.11, explains that the percentage of swelling increases with an increase in the concentration of polymer and diluent in the matrix system. The percentage of swelling ratio of aceclofenac sodium matrix tablets was obtained in the range of 142.50 to 229.60 at the end of 6 hours. The result indicates that by increasing the concentration of both polymer and diluent could influence the percentage of swelling ratio. The formulation batch AF-16 containing higher level of Pharmatose 200M (above 200mg) shows higher swelling in initial hours followed by erosion at the end of 6 hours because Pharmatose 200M is water soluble excipient forms more micron size pores in polymer matrices which rapidly increases the water penetration into interior layer of polymeric gel network initially and decreases the resistance of polymeric gel layer in later time period\textsuperscript{163}. The formulation of batch AF-20 containing more concentration of polymer and less concentration of diluent decreases the swelling ratio due to decreasing the resistance of gel layer with low concentration of diluent. The formulation AF-19 shows steady state swelling ratio for 6 hours is represented photographically in figure 5.4.5. This states that the presence of optimum level of HPMC K15M and Pharmatose 200M enhances the formation of thick gel acts as a barrier inhibiting the water penetration into interior layer of polymeric gel, thus gradually swells with an increasing time period

The figure 5.4.13 explains that the aceclofenac sodium release from the matrix tablets significantly increases with increasing the concentration of HPMC K15M and Pharmatose 200M ratio in the formulation. The aceclofenac sodium release from the matrix tablets was very less (<10%) for initial 2 hours in SGF pH 1.2 because of poor solubility of drug and polymer in this media. The drug release significantly increases
in SIF pH 7.4 at 6 hours followed by sustaining up to 12 hours. The formulation AF-16 shows rapid drug release compared to AF-20. The higher amount of Pharmatose 200M interact with lower amount of HPMC K15M forms loose network gel layer which rapidly diffuses the drug at higher pH level. On other hand, increase the concentration of HPMC K15M in the matrix tablets prolongs the drug release due to formation of thick diffusion gel layer, acts as a barrier that take prolonged time to diffuse aceclofenac sodium into bulk of the dissolution media.

6.4.6 Analysis of various drug release kinetics

The in-vitro drug release of all the batches of AF-1 to AF-20 aceclofenac sodium matrix tablets was treated with various kinetic models. The mechanism of drug release was determined by using PCP-DISSOv2.08 software. The results describes that the release mechanism from the batches of AF-1 to AF-15 matrix tablets shows highest correlation co-efficient corresponding to huguchi matrix followed to krosemeyer – peppas model. The formulation batches of AF-16 to AF-20 shows the mechanism of drug release zero-order followed to krosemeyer - peppas mixed order kinetics. All the formulation batches have shown the diffusion co-efficient (n) values in the range of 0.5810 to 1.185. This indicates, the drug release from matrix tablets by both diffusion and erosion mechanisms followed by super case-II transport.

6.4.7 Comparison of in-vitro drug release with marketed SR tablet.

From the table 5.4.14, In-vitro drug release of optimized formulation AF-19 was compared with marketed SR tablet of aceclofenac (Dolowin SR). The of drug release from the marketed SR tablet obtained in SGF pH 1.2 about 33.56 % w/w at initial 2 hours, more than 80% w/w at 6 hours and 99.10% w/w at the end of 12 hours. On the other hand, the formulated matrix tablet (AF-19) controls the initial burst release of drug in pH1.2 about 9.85 and 77.55% w/w of drug release at the end.
of 12 hours. This indicates that the formulated tablet gives more sustain drug release compared with marketed SR tablet.

6.5 Characterization and evaluation of dexibuprofen matrix tablets

In the present study, sustained release matrix tablets of dexibuprofen were prepared by Kollidon SR (Polyvinyl acetate and polyvinyl Pyrrolidone) and HPMC K15M polymers as retardants and Avicel PH 101(Microcrystalline cellulose), Pharmatose 200M (hydrated lactose monohydrate) as swellable diluents. The experimental results are mentioned in the chapter 5 and section 5.5. In this section, the experimental results such as micromeritic properties of powder blends and physical properties of compressed tablets like hardness, weight variation, friability, drug content uniformity, swelling properties and in-vitro drug release profiles are discussed.

6.5.1 Micromeritic properties of dexibuprofen powder blend mixtures

The sustained release dexibuprofen matrix tablets were prepared by direct compression technique. The mixed powder blends were compressed into tablets according to the formula given in the tables 4.10 to 4.13.

Direct compression method was employed in the production of many oral controlled release dosage forms to achieve prolonged release of drug from matrix-type particles. The polymer diluent ratio significantly influences the micromeritic properties of powder blends. The dexibuprofen is a crystalline substance that induces flow properties when blended with other excipients due to the formation of strongly bonding surfaces held together by the presence of crystalline of finite strength. The powder blends of different formulations were evaluated for angle of repose, loose bulk density, tapped bulk density, Carr’s index, and hausner’s ratio. From the table 5.5.1, the increase in the concentration of polymer and diluent ratio decreases the angle of
repose. All the formulations powder blends show angle of repose (<30) indicating good flow properties due to increase of bonding between microcrystals of drug and diluent. The bulk and tapped density values were obtained in the acceptable range indicating good packability. Compressibility index and hausner’s ratio values are obtained in the range of 15.26 to 7.65 and 1.22 to 1.10 respectively. This indicates that the prepared powder blends having excellent compressibility and good flow properties get acceptable hardness to the tablets.

6.5.2 Effect of HPMC K15M and Pharmatose 200M (DX-1 to DX-5)

The table 5.5.2 illustrates that the weight variation of the matrix tablets observed within pharmacopeia limit complied below ±5% w/w of standard deviation from the average. The average weight of the matrix tablet of the batch DX-4 was obtained very nearer to theoretical weight. It stated that the weight variation of formulated dexibuprofen tablets was influenced by both polymer and diluent content. The hardness of the formulated tablets with variations of polymer and diluent concentration was obtained within acceptable range of 4.8 ± 0.58 to 5.6 ± 0.16 kg/cm² followed by decreasing the percentage of friability except the batch DX-1. This indicates the low concentration of HPMC K 15M used in the formulation decreases the mechanical strength due formation of loose matrix network with high concentration of diluent (Pharmatose 200M). The content of dexibuprofen present in the formulated tablets was observed in the range of 77.66 ± 0.84 to 96.85 ± 0.82 %w/w. It stated that the percentage of drug content influences mainly by increasing the concentration of HPMC K15M entrapped more quantity of drug due to the formation of dense matrix during compression.

Figure 5.5.6 shows that increase in the concentration of both polymer and diluent could influence the percentage of swelling ratio. The formulation DX-1
containing higher level of Pharmatose 200M (above 200mg) shows higher swelling in initial hours followed by erosion at the end of 6 hours because the hydrophilic moieties of Pharmatose 200M forms more micro-cavities in polymer matrices and these induces the swelling and erosion of matrices observed in dissolution media. The formulation of batch DX-5 containing more concentration of polymer and less concentration of diluent slightly decreases the swelling ratio but controls erosion. The formulation DX-4 shows steady state swelling ratio for 6 hours. This is represented photographically in the figure 5.5.2. This explains that the presence of higher level of HPMC K15M enhances the formation of gel inhibiting water penetration into interior layer of polymeric gel and thus gradually swells with increasing time period.

The figure 5.5.8 explains that the dexibuprofen release from the matrix tablets significantly increases by increasing the concentration of HPMC K15M and Pharmatose 200M ratio in the formulation. The dexibuprofen release from the matrix tablets in SGF pH 1.2 for initial 2 hours was obtained in the range between 29.63 to 12.86 %w due to poor solubility of dexibuprofen in this environment. The maximum amount of dexibuprofen release (>50 % w/w) from the matrix tablets was observed in pH 7.4 at 6 hours followed by sustaining up to 12 hours. The formulation batch DX-5 was more sustained compared to DX-1 because of higher concentration of HPMC K15M in the matrix tablets prolongs the drug release extended period due to formation of thick diffusion gel layer which acts as a barrier, take prolonged time to diffuse the drug into bulk of the dissolution media.

6.5.3 Effect of HPMC K15M and AvicelPH101 (DX-6 to DX-10)
The table 5.5.5 reveals that the weight variation of the matrix tablets observed within pharmacopeia limit complied below ±5% w/w of standard deviation from the average. However, the average weight of the matrix tablet of the batch DX-9 was obtained very nearer to theoretical weight. It states that the weight variation of formulated dexibuprofen tablets was influenced by both polymer and diluent content. The hardness of the formulated tablets was obtained within an acceptable range < 6.0 kg/cm² followed by decreasing the percentage of friability except the batch DX-6. This indicates that the low concentration of HPMC K15M used in the formulation decreases the mechanical strength due to formation of loose matrix network with high concentration of diluent (Avicel PH101). The content of dexibuprofen present in the formulated tablets was observed in the range of 87.76 ± 0.84 to 98.42 ± 0.55 %w/w. It states that the percentage of drug content increases by increasing the concentration of HPMC K15M which entrapped more quantity of drug due to formation of dense matrix during compression.

Figure 5.5.6 shows that increase in the concentration of both polymer and diluent could influence the percentage of swelling ratio. The formulation DX-5 containing higher level of Avicel PH101 (above 200mg) shows higher swelling in initial hours followed by erosion at the end of 6 hours because Avicel PH101 is fibrous swellable excipient forms more pores in polymer matrices due to more penetration of dissolution media. This explains the higher swelling and erosion of matrices observed at the end of 6 hours. The formulation of batch DX-10 containing more concentration of polymer and less concentration of diluent slightly decreases the swelling ratio but controls the erosion due to high viscous polymeric layer. The formulation DX-9 shows steady state swelling ratio for 6 hours represented photographically in the figure 5.5.3. This is because the presence of optimum level of
HPMC K15M and Avicel PH101 enhance the formation of thick polymeric layer inhibiting water penetration and thus gradually swells with an increasing time period.

The depicted figure 5.5.9 illustrates that the percentage of dexibuprofen release from the matrix tablets in SGF of pH 1.2 for initial 2 hours was obtained in the range of 30.55 to 10.48 due to poor solubility of dexibuprofen in this environment. The maximum amount of dexibuprofen (>50 % w/w) from the matrix tablets in SIF pH 7.4 at 6 hours followed significantly in a sustained manner up to 12 hours. The formulation batch DX-6 shows rapid drug release at the end of 12 hours due to the decrease in tablet porosity by high level of Avicel PH101. The batch DX-10 was more sustained compared to DX-6 because of higher concentration of HPMC K15M forms a thick hydrodynamic gel layer by swelling; take prolonged time to diffuse the drug.

6.5.4 Effect of Kollidon SR and Pharmatose 200M (DX-11 to DX-15)

The table 5.5.7 explains that the weight variation of the matrix tablets observed within pharmacopeia limit of all the formulations were complied with the specifications given in I.P below ± 5% w/w of standard deviation from the average. However, the average weight of the matrix tablet of the batch DX-14 was obtained very nearer to theoretical weight. This indicates that the weight variation of formulated dexibuprofen tablets influenced by both polymer and diluent content. The hardness of the tablets obtained within acceptable range (< 6.0 kg / cm²) followed by decreasing the percentage of friability except the batch DX-11 because of low concentration of Kollidon SR used in the formulation decreases the mechanical strength due to less binding with high concentration of diluent (Pharmatose 200M). The percentage of dexibuprofen content present in the tablets was found in the range of 90.26 ± 0.92 to 98.92 ± 0.88 respectively. It illustrates that the percentage of drug content influences mainly by increasing the concentration of Kollidon SR which
entrapped more quantity of drug due to plastic deformation during compression and forms dense matrices.

Figure 5.5.10 stated that increasing the concentration of both polymer and diluent could influence the percentage of swelling ratio. The formulation DX-11 containing higher level of Pharmatose 200M (above 200mg) and lower level of Kollidon SR shows higher swelling in initial hours followed by erosion at the end of 6 hours because Pharmatose 200M is more water-soluble excipient which helps to rapid hydration of polymer and erosion. The formulation of batch DX-15 containing more concentration of polymer and less concentration of diluent decreases the swelling due to low hydration of Kollidon SR. The formulation DX-14 shows steady state swelling ratio for 6 hours and represented photographically in the figure 5.5.4. This is due the presence of Pharmatose 200M enhance the formation of thick polymeric gel which acts as a barrier to penetrate water into interior boundary surface of tablet and gradually swells with an increasing time period.

The figure 5.5.12 illustrates that the percentage of dexibuprofen release from the matrix tablets in SGF pH 1.2 for initial 2 hours was obtained in the range of 37.52 to 13.87. This may be due to poor solubility of dexibuprofen in this environment. The maximum amount of dexibuprofen (>65 % w/w) from the matrix tablets in SIF pH 7.4 at 6 hours and were sustained up to 12 hours. The composition of Kollidon SR mainly influences the dual release kinetics.

The rapid drug release was observed from the batch DX-11 due to lower concentration of Kollidon SR as retardant. The formulation DX-15 containing high level of Kollidon SR and low level of Pharmatose 200M shows higher burst release in initial hours and the drug release may be attributed for the dissolution of povidone molecules which are components of Kollidon SR create pores and channels, thus
facilitating solvent front penetration and elevation of drug release. Polyvinyl acetate (PVA) is a component of Kollidon SR not soluble in water and does not impede diffusion of drug from the matrix system and causes inefficient drug release for extended time period\textsuperscript{164}. The formulation batch DX-14 containing optimum concentration of Kollidon SR and Pharmatose 200M shows a remarkable sustained release effect because Pharmatose 200 M is more water-soluble excipient, forms more micro-cavities in polymer matrices, forms dense matrix and influences swelling of matrices to form a thick polymeric gel layer. Moreover, this thick gel layer acts as surface barrier, controls the burst release and release in a sustained manner.

6.5.5 Effect of Kollidon SR and Avicel PH101 (DX-15 to DX-20)

Table 5.5.7, illustrates that the weight variation of the matrix tablets observed within pharmacopeia limit of all the formulations were complied with the specifications given in I.P below ± 5% w/w of standard deviation from the average. However, the average weight of the matrix tablet of the batch DX-19 was obtained very nearer to theoretical weight. It illustrates that the weight variation of formulated dexibuprofen tablets influenced by both polymer and diluent content. The hardness of the formulated tablets obtained within acceptable range < 6.5 kg/cm\textsuperscript{2} followed by decreasing the percentage of friability except the batch DX-16. This indicates the low concentration of Kollidon SR used in the formulation decreases the mechanical strength due low compressibility with high concentration of diluent (Avicel PH101). The percentage of dexibuprofen content present in the formulated tablets was observed in the range of 87.56 ± 0.52 to 98.72 ± 0.35 respectively. It was noticed that the percentage of drug content were influences mainly by increasing the concentration of Kollidon SR entrapped more quantity of drug due to plastic deformation during compression which influences the formation of dense matrices.
Figure 5.5.11, illustrates that the increase in the concentration of both polymer and diluent could influence the percentage of swelling ratio. The formulation DX-16 containing high level of Avicel PH101 (above 200mg) and low level of Kollidon SR shows higher swelling in initial hours followed by erosion at the end of 6 hours because Avicel PH101 is water-insoluble swellable fibrous excipient, forms more porous nature polymeric gel layer. This explains the higher swelling and erosion in dissolution media. The formulation of batch DX-20 containing more concentration of polymer and less concentration of diluent decreases the swelling due to low hydration of Kollidon SR. The formulation DX-19 shows steady state swelling ratio for 6 hours and represented photographically in the figure 5.5.5. This explains that the Avicel PH101 is a water-swellable polymer that influences water uptake to enhance gel formation by weakening the matrix integrity inhibited water penetration into interior layer of polymeric gel and gradually swells with an increasing time period.

The figure 5.5.13 explains that the percentage of dexibuprofen release from the matrix tablets in SGF pH 1.2 for initial 2 hours was obtained in between 24.52 to 8.65 due to poor solubility of dexibuprofen in this environment. The maximum amount of dexibuprofen (> 50 w/w) from the matrix was observed in SIF pH 7.4 at 6 hours was followed by sustaining up to 12 hours.

The rapid drug release was observed in the batch DX-16 due to lower concentration of Kollidon SR as retardant. The formulation DX-19 containing high level of Kollidon SR and low level of Avicel PH101 shows higher burst release in initial hours which may be attributed to dissolution of povidone molecules which are components of Kollidon SR create pores and channels, thus facilitating solvent front penetration and elevation of drug release. Polyvinyl acetate (PVA) is not soluble in water, does not impede diffusion of drug from the matrix system and causes
inefficient drug release in extended time period. The formulation batch DX-19 containing optimum concentration of Kollidon SR and Avicel PH101 shows a remarkable sustained release effect because Avicel PH101 is a water swellable fibrous diluent which influences the formation of thick fibrous gel and increases resistance of matrix layer for long period of time to control the burst release of drug from the formulations. The results clearly explain that the optimum level of polymer and diluent ratio modifies the drug release in a sustained manner for prolonged period of time.

6.5.6 Analysis of various drug release kinetics

The in-vitro drug release of all the batches of DX-1 to DX-20 dexibuprofen matrix tablets was treated with various kinetic models. The mechanism of drug release was determined by using PCP-DISSOv2.08 software. The table 5.5.12, describes the release mechanism from the batches of DX-1 to DX-10 matrix tablets showed highest correlation co-efficient corresponding to zero-order followed to krosemeyer – peppas model and diffusion co-efficient (n) values found in the range of 0.8306 to 1.1358. This is because the higher swelling properties of HPMC K15M forms a thick polymeric gel, diffuses the drug gradually in a sustained manner. The formulation batches of DX-11 to DX-20 shows the mechanism of drug release both first order and huguchi matrix followed to krosemeyer - peppas mixed order kinetics. The formulation batches of DX-15 and DX-20 containing more concentration of Kollidon SR and less amount of diluent show highest correlation coefficient linearity to first order release mechanism due lower swelling properties of polymer. The diffusion co-efficient (n) values were observed from the batches DX-11 to DX-20 in the range of 0.6381 to .8834. This indicates, the drug release from matrix tablets by both diffusion and erosion mechanisms followed by Fickian matrix diffusion.
6.5.7 Comparison of in-vitro drug release with marketed SR tablet

From the table 5.5.14, In-vitro drug release of optimized formulation DX-14 was compared with marketed SR tablet of ibuprofen (Ibuspan SR). The of drug release from the marketed SR tablet obtained in SGF pH 1.2 about 28.52 % w/w at initial 2 hours, more than 78% w/w at 6 hours and 99.65 % w/w at the end of 12 hours. On the other hand, the formulated matrix tablet (DX-14) release of drug in pH1.2 about 20.35 and 74.15 % w/w of drug release at the end of 12 hours. This indicates that the formulated tablet gives more sustain drug release compared with marketed SR tablet. Thus, the optimum proportion of polymer and diluent is favorable for the preparation of sustained release products.

6.6 Accelerated stability study

No significant changes was observed for the formulated aceclofenac sodium and dexibuprofen matrix tablets in their physicochemical parameters such as drug content, average weight, and hardness after 01, 02, 03, 04, 05 and 06 months when kept at 25°C / 60% RH, 40°C / 75% RH and room temperature. The observations are mentioned in the table 5.6.1 for aceclofenac sodium matrix tablets and 5.6.2 for dexibuprofen matrix tablets. Based on these observations, it was concluded that developed formulations of aceclofenac sodium and dexibuprofen matrix tablets are physically and chemically stable and retain their pharmaceutical properties at various temperature and humidity conditions over a period of 6 months.

Stability study by HPLC technique

By comparing the chromatographs of pure aceclofenac sodium and formulated matrix tablets and observing the results from the table 5.6.3, the retention time, peak height, theoretical levels, tail factor and resolution factors are almost identical compare with pure drug and formulated products. On the other hand, asymmetry value
obtained below 2.0 and recovery of concentration of drug in the formulation slightly decreases. The results indicate that the formulated matrix tablets are stable but slightly retain their active drug potency in the formulation after 6 months.

By comparing the chromatographs of pure dexibuprofen and formulated matrix tablets and observing the results from the table 5.6.4, the retention time, peak height, theoretical levels, tail factor and resolution factors are almost identical compare with pure drug and formulated products. On the other hand, asymmetry value obtained below 2.0 and recovery of pure drug about 98.32 mg. but in the formulated tablets, the asymmetry value above 2.0 and the content of dexibuprofen decreases significantly about 83.07 mg. The results indicate that the formulated matrix tablets are not stable because some amount of active drug degraded in the formulation after 6 months.