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CHARACTERIZATION AND EVALUATION OF BIO-COMPATIBLE EUDRAGIT-S-100 NANOPARTICLE LOADED WITH CLOFARABINE AS POTENTIAL ANTI-TUMOR DRUG DELIVERY SYSTEM

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Abstract:

Clofarabine is a second-generation purine nucleoside analog that has been synthesized to overcome the limitations and incorporate the best qualities of fludarabine and cladribine. Clofarabine acts by inhibiting ribonucleotide reductase and DNA polymerase, thereby depleting the amount of intracellular deoxynucleoside triphosphates available for DNA replication. Compared to its precursors, clofarabine has an increased resistance to deamination and phosphorolysis, and hence better stability as well as higher affinity to deoxycytidine kinase (dCyd), the rate-limiting step in nucleoside phosphorylation. Since the initiation of the first phase I study of clofarabine in 1993 in patients with hematologic and solid malignancies, clofarabine has demonstrated single-agent antitumor activity in adult acute leukemia, including acute myeloid leukemia (AML). Due to its unique properties of biochemical modulation when used in combination with other chemotherapy drugs, mainly cytarabine, combination regimens containing clofarabine have been evaluated. The treatment of acute leukaemias, which are the most common paediatric cancers, has improved considerably in recent decades, with complete response rates approaching ~90% in some cases. However, there remains a major need for treatments for patients who do not achieve or maintain complete remission, for whom the prognosis is very poor. In this article, we describe the challenges involved in the discovery and development of clofarabine, a second-generation nucleoside analogue that received accelerated approval from the US FDA at the end of 2004 for the treatment of paediatric patients 1–21 years old with relapsed or refractory acute lymphoblastic leukaemia after at least two prior regimens. It is the first such drug to be approved for paediatric leukaemia in more than a decade, and the first to receive approval for paediatric use before adult use.

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INTRODUCTION:

Nanoparticles as cancer-drug carrying systems: Nanoparticles are usually defined as particles with sizes smaller than 100 nm in one dimension at least. They can range from 1 to 100 nm in size. Nanoparticles in this size range have been prepared for drug formulation and delivery. The US National nanotechnology initiative refers to nanotechnology as the study of structures that are about 100 nm or less. Actually, the marketed liposomal and albumin-bound nanoparticles anticancer drug products are within the submicron size of 100–1000 nm. Based on morphological propertites, partilce-size, physico-chemical properties, NPs, which are used in many fields , can be classified into two main types: organic and inorganic. However, there is also a different classification where, in addition to these two, carbonbased NPs are considered as a separate type of NPs. Inorganic NPs involve gold and silicaNPs, fullerenes and quantum dots while organic NPs are liposomes, micelles, dendrimers, hybrid and compact polymeric NPs (nanospheres and nanocapsules). Thereby, micelles, liposomes, dendrimers, polymer conjugates and nanotubes are mostly used as drug delivery systems. Taking into account that their structure and composition differ, the appropriate system is chosen depending on the drug type used in the treatment and on the way it is attached to the carrier.

RESULTS AND DISCUSSIONS:

Characterization of Nano-Suspension Particles:

Particle size determination and distribution. Preliminary Particle Size Determination was performed by using Master-Sizer. The nano-suspension size was determined by photon correlation spectroscopy by master sizer (Master-sizer 2000; Malvern instruments Corp, U.K.). The particle size analysis was conducted at scattering-angle of 90° at ambient temperature (25°C) using suitably diluted sample solution with distilled & deionized (DD) water. Briefly, nano-suspension testing sample was added to the DD water operated with a low speed shaft-pump and swirled to distribute the nanoparticles uniformly within the zone-of-measurement.

Nanoparticles were characterized for three distribution widths namely-Dv-10, Dv-50 and Dv-90 (Dv-50, Dv-10 and Dv-90 are standard percentile readings from the analysis.), mean particle size, span (is defined as measurement of width of distribution-i.e, smaller span indicates the narrow particle size distribution of nanoparticles in sample) and uniformity. The measurements are done in three times as per SOP and the values were depicted in triplicate. The values reported are average diameter ± standard deviation for 3 replicate samples.The final optimized formulas prepared after trials were analyzed under Particle Size Analysis by master sizer for distribution width, Mean-Particle-Size (MPS), Surface Area (SA) (m2/g), Span and Uniformity. The results were presented in
the Table.

Among all the formulas (CLFRBN1-to-CLFRBN5) CLFRBN1, CLFRBN2 and CLFRBN3 were found to give better results in the above listed parameters. The particle size analysis report of Master sizer was found to be varying with the formulations (CLFRBN1-CLFRBN5). The formulation CLFRBN1 demonstrated the lowest distribution width (Dv-10, Dv-50 and Dv-90) 10, 23.2 and 2233.9 nm respectively among all the other formulations.The surface area of CLFRBN1 formulation was found to be 26.3 m2/g and it was largest among all the formulations. The surface area is large due to small particle size. The overall report of mean particle size analysis was found to be better in CLFRBN1 formulation which was recorded as 755.7 nm (MPS). The CLFRBN2 mean particle size (MPS) was reported with 2179.5 along with values (D10, D50 and D90) 10.8456.2 and 6071.5 nm respectively.

The mean particle size values (MPS) of CLFRBN3 , CLFRBN4 and CLFRBN5 was 1922.7, 1165.8 and 1000.9 nm respectively and the surface area values were recorded 23, 0.739 and 0.859 respectively.

The formulations CLFRBN1, CLFRBN2, CLFRBN3 which demonstrated the nano range in the master sizer reports were further subjected to analysis by Delsa NanoTMC. Before going to analysis by the TM zeta sizer (Delsa NanoTM C) the formulations CLFRBN1, CLFRBN2 and CLFRBN3 were subjected to probe sonication for 3 minutes with an impulse of 5 seconds.

The probe sonication leads to the reduction of particle size as well as improving the stability of the formulation. The results after the analysis by Delsa NanoTMC revealed that the formulation CLFRBN1 can be declared as best formulation in terms of size with a value of 832.2±806.5 and the detailed zeta sizer analysis is discussed below.Particle Size Analysis by Delsa Nano TM Common: The best formulations of Eudragit S 100 group (CLFRBN1-CLFRBN5) were subjected to particle size analysis by Beckman Coulter. The results were presented in Table. The formulation CLFRBN1, CLFRBN2 and CLFRBN3 were found to be better in achieving the nano range in master sizer report in the below figure.

Among the three Eudragit S 100 formulations (CLFRBN1, CLFRBN2 and CLFRBN3) the formulation CLFRBN1 was found to be the best in terms of size (416.4 nm) which was the lowest size of all the Eudragit formulations. The poly-dispersibility index of the formulation was found to be 0.329 for CLFRBN1 and it was 0.331 and 0.392 respectively for CLFRBN2 and CLFRBN3. The mean particle size of the CLFRBN1 was 832.2±806.5 and it was 1162.3±821.6 and 2688.3±4612.3 for CLFRBN2 and CLFRBN3 respectively.

Among these lowest Stander Deviation (S) was observed in CLFRBN1 which is ± 806.5 and this indicated the uniform particle size and distribution of the formulation as compared to CLFRBN2 and CLFRBN3.

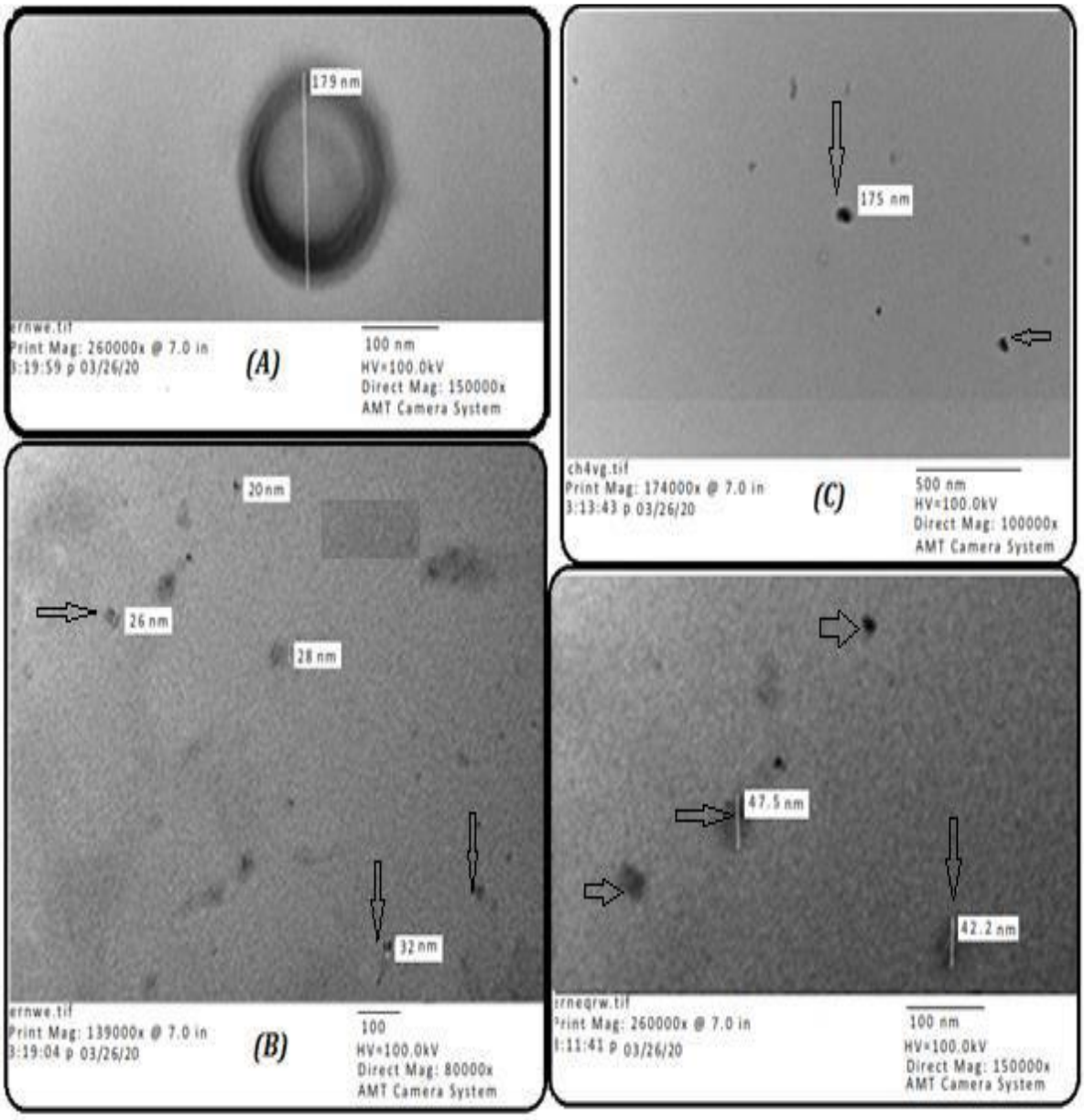
Likewise, Dv-10, Dv-50 and Dv-90 also established as 154.9, 524.40 and 1829.3 respectively whereas CLFRBN2 and CLFRBN3 represented as the highest indices which established that CLFRBN1 can be declared as optimized formula in the Eudragit-S-100 nanoparticle formulation design which contains Eudragit-S- 100 (100mg) and Pluronic F- 68 (0.5%) and prepared at 1000 rpm (Remi motors, RQT-127A; Vasai, India).

Table 1: Optimizing the composition of ingredients in Clofarabine nanosuspension.

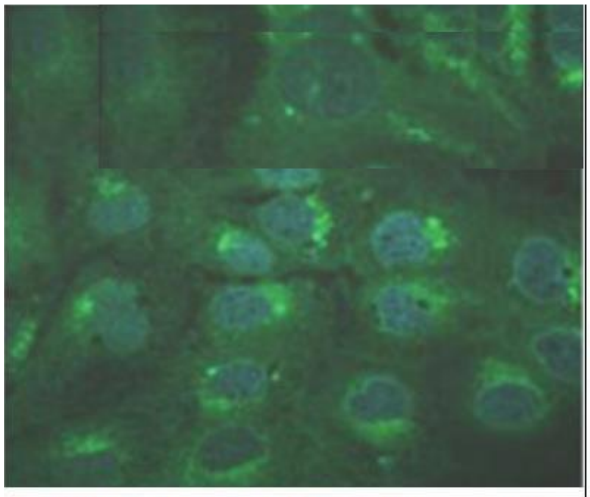
Components	CLFRBN-1	CLFRBN-2	CLFRBN-3	CLFRBN-4	CLFRBN-5
Clofarabine	10 mg	20 mg	30 mg	40 mg	50 mg
Eudragit-S-100	100 mg	200 mg	300 mg	400 mg	500 mg
Pluronic-F-68	0.5 % (w/v)	1 % (w/v)	1.5 % (w/v)	2 % (w/v)	2.5 % (w/v)
Tween 80 %	0.02 (w/v)	0.02 (w/v)	0.02 (w/v)	0.02 (w/v)	0.02 (w/v)
Acetone	5	8	10	10	12
Organic Phase	20 mL	25 mL	25 mL	30 mL	40 mL

The Particle Size and Zeta Potential Determination:
The mean diameter of CLOFARABINE nanoparticles in the dispersion was determined by photon correlation spectroscopy (PCS) using a laser light scattering instrument (Delsa Nano TM Common; Beckman Coulter) at fixed angle of 90°. The particle size analysis data was further examined based on the volume distribution. Zeta potential was also measured by using (Delsa Nano TM Common; Beckman Coulter) under the same conditions. Previously the samples were kept in low conductivity zeta cell in order to meet the instrumental conditions and then the readings were recorded.

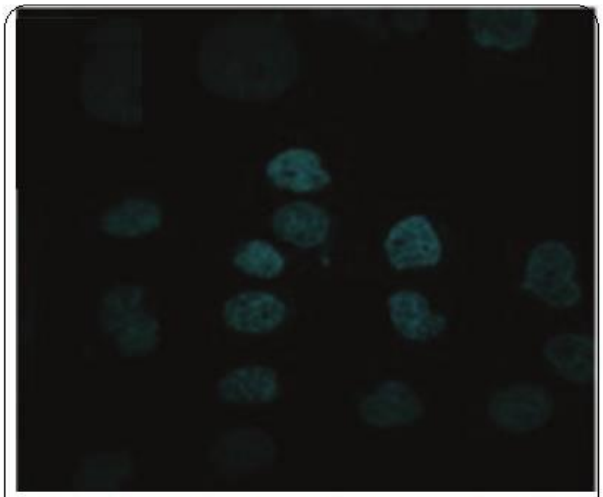
Particle Size Analysis of CFRBN-EUD-S-100-P.L.F-68-NPs by Malvern Master Sizer							
Formulation Code Number	Distribution width in (nm)			MPS	SA (m2 /g)	Span	Uniformity
	Dv-10	Dv-50	Dv-90				
CFRBN-1	11.1	24.5	2435.7	823.77	27.7	98.963	31.7
CFRBN-2	12.8	482.2	6198.3	2231.1	22.5	12.827	4.31
CFRBN-3	11.8	46.3	5624.4	1894.16	24.2	120.96	32.5
CFRBN-4	444.7	985.4	2191.8	1207.3	0.865	1.772	0.84
CFRBN-5	379.2	847.5	1821	1015.9	0.982	1.701	2.02



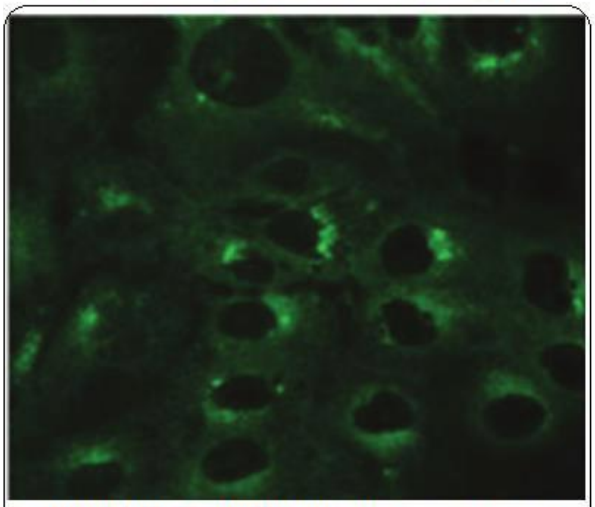
The particle size determination in the TEM images.



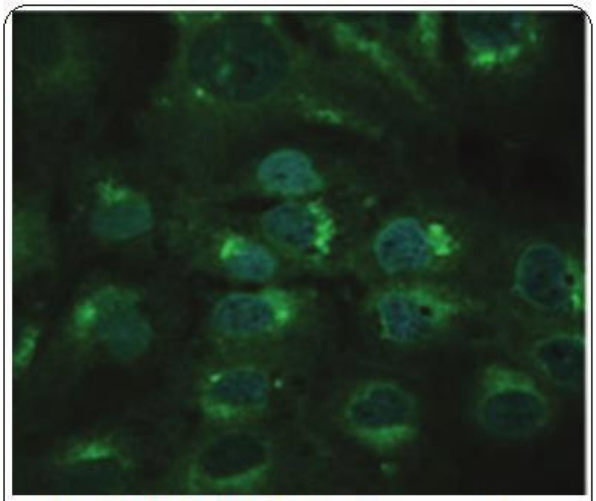
CLFRBN-EDU-S-100-P.L-F-68- formulation-1 -Nps uptake in to Hs-578-BST-Cell linings



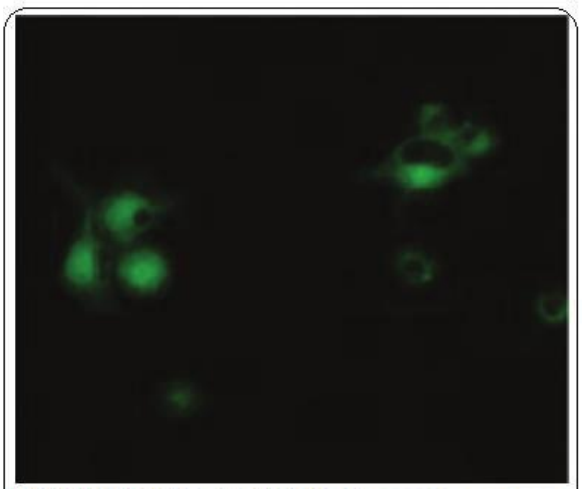
CLFRBN-EDU-S-100-P.L-F-68- formulation-1 -Nps uptake in to MCF-7-Cell linings



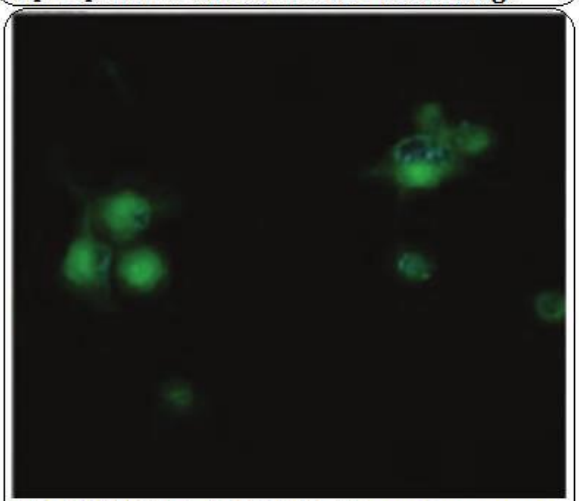
CLFRBN-EDU-S-100-P.L-F-68- formulation-1 -Nps uptake in to HS-39-T-Cell linings



CLFRBN-EDU-S-100-P.L-F-68- formulation-1 -Nps uptake in to UACC-3133-Cell linings



CLFRBN-EDU-S-100-P.L-F-68- formulation-1 -Nps uptake in to MDA-MB-453-Cell linings



CLFRBN-EDU-S-100-P.L-F-68- formulation-1 -Nps uptake in to UACC-732-Cell linings

Determination of CLOFARABINE Solubility CLOFARABINE solubility experiments were done utilizing PEG 400. The abundance quantity of API was included various dilutions of PEG 400. The PEG 400 and water were blended in various fixations (0.5:4.5), (1:4), (1.5:3.5), (2:3) and (3:2). The medication CLOFARABINE exhibited the better dissolution pattern in 2:3 proportion of PEG and water. Already five test tubes were taken containing various proportions of PEG and water as portrayed before. The 1mg medication was weighed precisely and brought into each test tube and was blended for few moments. After the total dissolution of CLOFARABINE again 1mg of CLOFARABINE was added to the above test tubes and again the test tubes were shaken for few moments.

These operations were replicated for multiple times until the measure of the medication became 5 mg in all the test tubes. At that point these test tubes were again shaken for 1 h persistently so as to accomplish greatest dissolvability.

At that point the test tubes were placed aside as such for 24 h and after 24 h the medication solvency in the test tubes were outwardly assessed. The test tube with no medication appears at the base was in the 2:3 proportion of solvent mixture and the other test tubes were containing the medication at the base which was undissolved.

The medication solvency with 1mg/ml without any traces at the base was exposed to the UV examination by planning working standard dilution of 5, 10, 15, 20, 25, 30, 35, 40, 45, and 50 µg/ml and were checked in UV-Spectrophotometer between the range 200-600 nm. The maximum absorbance was seen at 295 nm and 448 nm which were in acceptable consent to European Pharmacopeia.As the working dilutions of 5, 10, 15, 20, 25, 30, 35, 40, 45, and 50 µg/ml were seen as straight line relations ship, hence these range of concentrations were chosen and calibration curve was set up in triplicate and the mean was taken.This technique was additionally utilized in the readiness of standard curve of CLOFARABINE in 0.1 N HCl, 6.8 pH and 7.4 pH buffers.

UV Analysis of Clofarabine In the UV analysis a primary standard solution of CLOFARABINE (1mg/1ml) was set up by dissolving precisely measured amount of CLOFARABINE in water and PEG 400 (3:2) and the working standard solutions were set up by further diluting the standard solution with PEG 400 and water to make the final working standard solutions. The absorbance of resulting testing solutions was recorded in a 10mm quartz cell of an UV- Visible Spectrophotometer (Systronics, India).

The calibration plot of the medication and the absorbance maxima, as per Beer-lamberts' law were recorded. A similar methodology was completed utilizing the 0.1 N HCl, 6.8pH phosphate and 7.4 pH buffer solutions as the definition of nanoparticles implied for sustained and controlled release at specified site. The absorbance maxima was chosen at 295 nm.

Drug-Release studies of Clofarabine from Nanosuspended particles:

The medication release profile from arranged nano-suspension was controlled by utilizing dialysis pack (Himedia labs, cut-off weight 12000-11000 Da) strategy for more than 24 h.

In this technique, the dialysis packs were soaked in two-fold refined water for 12 hours before use. At that point the dialysis sacks were removed from the water and attached with the assistance of a string from one end. Clofarabine nano-suspended particles were then brought into the dialysis sack by methods for a pipette. Dialysis pack held the nanoparticles and permitted the free medication in dissolution media with a cut-off of 12000 Dalton.

The 10mg equivalent quantity of Clofarabine containing nano-suspension particles was filled in the dialysis sack and the opposite end was fixed solidly by methods for strings. The sack was set in the measuring glass containing 100ml of receiving medium as a media. The framework was held under attractive mixing conditions at 100 rpm.

The samples were collected at predetermined time frames, 1, 1.5, 2, 3, 4, 5, 6, 7, 8, 12, and 24 hrs. Testing solutions were collected from the dissolution media and were renewed with the same quantity of fresh new dissolution media so as to keep up the sink conditions. The testing dilutions were investigated spectrophotometrically utilizing UV spectrophotometer against the blank or placebo. All the perceptions were completed in triplicate.

***In Vitro* Drug Release Study**.

In Vitro drug release pattern was performed by utilizing the various buffer solutions, for example, 0.1N HCl (1.2 pH), 6.8 pH potassium dihydrogen phosphate buffer system and 7.4 pH phosphate solution to simulate the Gasto Intestinal tract situation of stomach, digestive system and colon separately. All the prepared formulation samples

(CLFRBN1-CLFRBN5) were studied to find out the drug-release fashion with reference to time profile analysis.

The medication release studies were performed up to 24 h. The testing dilutions were taken in different time spans. During the acidic conditions (1.2pH) the sample dilutions were collected for each thirty minutes up to 1.5 h. At that point during the intestinal conditions 6.8 pH the testing dilutions were withdrawn for each 1 h up to 4 h followed by the colonic conditions 7.4 pH the testing dilutions were collected for 1 h up to 8 h and then final two dilutions were taken in 12 h and 24 h. The examples pulled back from the framework were renewed with new media. The plan CLFRBN1 was seen as best as it discharged the medication up to 24 h. During the acidic condition the detailing CLFRBN1-formulaiton was released around 9% of the medication only at 1.5 h. Being a sustained released dosage form containing the nanoparticles, it is suggested that the medication release should be minimum as per the protocol to be least in the upper GI tract and it was acquired in the prepared Eudragit
nanoparticles of CLFRBN1.

After the acidic condition the medication release profile was gradually extended up to 12 hrs and it was found that as 52.928±1.22. The further sample was collected at 24 h and the medication release was seen as 85.506±2.13. The other formulation is also showed the controlled release profile however the medication release pattern was highly variable may be due to different proposition of polymer in each developed formulation.

The CLFRBN2-formulaiton and CLFRBN3-formulation discharged the medication measure of 44.092±1.26 and 46.122±1.19 individually toward the end of 12 hrs. The medication discharge from these details was 79.126 ±2.15 and 73.225±2.931respectively for complete 24 hrs. The rest of the details CLFRBN4 and CLFRBN5 showed the futile drug release patterns of 60.017±3.33 and 64.165±3.11 individually. In any case, these dosage forms were at that point discarded from the class of best formulation dependent on the results of study. Hence it was demonstrated that the best detailing CLFRBN1 discharged the medication in a deferred discharge design with a foreordained medication discharge profile. Relative measure of medication released from polymeric nanoparticles of Eudragit S 100 (CLFRBN1-CLFRBN5) .

Drug Release Kinetics:

The manufactured Eudragit S 100 nanoparticles were subjected to the investigation of medication release kinetics and mode of drug release. The prepared formulations were evaluated by fitting the medication discharge time profile with the different pharmacokinetic models, for example, Zero-order rate of release, First- order rate of release, Higuchi, Korsmeyer Peppas and Hixon-Crowell. All the five optimized nanoparticle dosage forms, CLFRBN1-to-CLFRBN5 were studied for the mode of drug release.

All the formulations from CLFRBN1-to-CLFRBN5 exhibited the best fit with Korsmeyer-Peppas model. The best formulation CLFRBN1 exhibited the R2estimation of 0.990. The 'n' estimation of CLFRBN1 nanoparticle suspension was 0.807 demonstrated non-fickian patterns of dispersion.

The 'n' is an exponent used to portray distinctive drug release systems. The 'n' estimate in the event if 'n'> 0.89 demonstrates super case transport and on the off chance that $0.45<n<0.89$, at that point it is a non fickian-mode of drug release by diffusion. In the other nanoparticle formulations, for example, CLFRBN2, CLFRBN3, CLFRBN4 and CLFRBN5 likewise showed the Korsmeyer-Peppas energy.

The 'n' estimation of details showed to be around 0.994, 0.891, 0.873 and 0.955 separately for CLFRBN2, CLFRBN3, CLFRBN4 and CLFRBN5. Among these nano- suspensions, CLFRBN2 and CLFRBN5 exhibited the super simplicity transport with the exception of CLFRBN1, CLFRBN3 and CLFRBN4 which followed non fickian atypical mode of drug release pattern.

Stability Studies:

The Optimized formulation of the overall formulation design was injected into 10ml ampoules and sealed for storage at 2-8°C for short term stability study. The parameters such as clarity, appearance, and drug release pattern and entrapment efficiency were determined. The best formulation CLFRBN1 was subjected to stability studies in terms of short term and it was found to be stable during 30-day study period. The parameters such as physical appearance, % Entrapment efficiency and drug release profiles were evaluated after 24hrs, 7-days and 30-days. The results were fulfilling the acceptance criterion and there was a negligible deviation in these results indicated that the optimized formulation is anticipated to be stable for long term stability studies. However, the long term stability studies are recommended for data analysis in further formulation development process.

Anti-breast Cancer /Cytotoxic activity Procedure (in-brief) Cell line Culture Cell lines were obtained from National centre for cell sciences Pune (NCCS). The medium and Trypsin-Phosphate-Versene-Glucose (TPVG) was brought to room temperature by thawing. The tissue culture bottles were observed for growth, cell degeneration, pH and turbidity in inverted microscope.

After the cells became 80% confluent and free-flowing then sub culturing was done. The mouth of the bottle was wiped off by using spirit soaked cotton to remove the adhering particles.The growth medium was discarded 4-5 ml of Minimum-Essential-Media (MEM) was added without Foetal-Calf-Serum (FCS) and rinsed gently by tilting. The non-viable cells and excess FCS were washed out, and the medium was discarded. TPVG was added over the cells, incubated at 37o C for 5 minutes to develop the cell as individual and discrete entities. The cells become individual and were present as suspension. 5ml of 10% MEM was added to FCS by using serological pipette. Normal breast (MCF-7) cells were cultured in 1:1 mixture of DMEM and Ham's F12 medium with 20 mg/ml of epidermal growth factor (EGF), 100 µg/ml cholera toxins, 0.01 mg/ml insulin and 500 µg/ml Cortisol, and 5% chelex treated horse serum. Purified berberine and tamoxifen were dissolved in dimethyl sulfoxide (DMSO) and used for the bioassays.

$$\text{Cell viability}(\%) = \frac{\text{Mean optical density}}{\text{Control optical density}} \times 100$$

MTT-(3-(4,5-dimethyl thiazol-2-yl) -2,5-diphenyl tetrazolium bromide, a tetrazole) Assay:

Principle:

Measurement of cell viability and proliferation forms the basis for numerous in vitro assays of a cell population's response to external factors. The reduction of tetrazolium salts is now widely accepted as a reliable way to examine cell proliferation. The yellow tetrazolium MTT (3-(4, 5-dimethylthiazolyl-2)-2, 5-diphenyltetrazolium bromide) is reduced by metabolically active cells, in part by the action of dehydrogenase enzymes, to generate reducing equivalents such as NADH and NADPH.

The assay relies on the reduction of MTT, a yellow water-soluble tetrazolium dye, primarily by the mitochondrial dehydrogenases, to purple colored formazan crystals.The formazan product is analyzed spectrophotometrically (550-570 nm) after dissolution in DMSO, the spectra of nanoparticle-treated and untreated cells giving an estimate of the extent of cytotoxicity. The resulting intracellular purple formazan can be solubilized and quantified by pectrophotometric means.

The MTT Cell Proliferation Assay measures the cell proliferation rate and conversely, when metabolic events lead to apoptosis or necrosis, the reduction in cell viability. The MTT Reagent yields low background absorbance values in the absence of cells. For each cell type the linear relationship between cell number and signal produced is established, thus allowing an accurate quantification of changes in the rate of cell proliferation.

MTT Assay The anticancer activity of samples on breast cancer cells were determined by the MTT (3-(4, 5-dimethyl thiazol-2yl)-2, 5-diphenyl tetrazolium bromide) assay was used to assess the cytotoxicity Horiuchi et al (1988). Cells (1 × 105/well) were plated in 0.2 ml of medium/well in 96-well plates. For MTT assay the medium from the wells was removed carefully after incubation. Each well was washed with MEM (w/o) FCS for 2-3 times and 200µl of MTT (5mg/ml) was added. The plates were incubated for 6-7 hrs in 5% CO2 incubator for cytotoxicity. After incubation, 1ml of DMSO (solublizing reagent) was added to each well and mixed well by micropipette and left for 45sec. Presence of viable cells was visualized by the development of purple color due to formation of formazan crystals. The suspension was transferred to the cuvette of a spectrophotometer and the OD (optical density) values were read at 595nm by using DMSO as a blank. Measurements were performed and the concentration required for a 50% inhibition of viability (IC50) was determined graphically Standard Graph was plotted by taking concentration of the drug in X axis and relative cell viability in Y axis.

CONCLUSION:

Clofarabine can be successfully loaded into the Eudragit nanoparticles with high encapsulation efficiency. This study explored the use of Clofarbine loaded Eudragit nanoparticles as a biodegradable drug delivery system for treatment of cancer. The release profile of CLFRBN-NP showed good cellular adhesion and controlled release properties compared to conventional dosage form. The Eudragit® S 100 coating over pluronic layer helps the nanoparticle to release clofarabine only at the tumour area which shows a sustained release.

The optimized formulation was effective against cancer cell lines In Vitro. Thus, current study clearly indicates the promising potential of Eudragit® S 100 encapsulated pluronic-F nanoparticles of clofarabine

for tumour suppression with better patient compliance. Future studies using animal models will provide more light on the effectiveness of the nano-formulation In Vivo. Thus, Pluronic-F-Eudragit S100 nanoparticles are promising drug carriers that can lead to effective cancer treatment.

REFERENCES:

1. Schiffer, C. A. Acute myeloid leukemia in adults: where do we go from here? Cancer Chemother. Pharmacol. 48 (Suppl.): S45–S52, 2001.
2. Plunkett, W., and Gandhi, V. Purine and pyrimidine nucleoside analogs. In: G.Giaccone, R. Schilsky, and P. Sondel (eds.), Cancer Chemotherapy and Biological Response Modifiers, Annual, Vol. 19, pp. 21–45. Amsterdam: Elsevier Science Publishers, B. V., 2001.
3. Gandhi, V., Plunkett, W., Du, M., Ayres, M., and Estey, E. Prolonged infusion of gemcitabine: clinical and pharmacodynamic studies during a Phase I trial in relapsed acute myelogenous leukemia. J. Clin. Oncol., 20: 665–673, 2002.
4. Silverman, L. R., Demakos, E. P., Bercedis, L., Kornblith, A. B., Holland, J. C., Odchimar-Reissig, R., Stone, R. M., Nelson, D., Powell, B. L., DeCastro, C. M.,et al. A randomized controlled trial of azacytidine in patients with myelodysplastic syndrome: a study of the cancer and leukemia group B. J. Clin. Oncol., 20: 2429–2440, 2002.
5. Wijermans, P., Lu¨bbert, M., Verhoef, G., Bosly, A., Ravoet, C., Andre, M., Ferrant, A. Low-dose 5-aza-2-deoxycytidine, a DNA hypomethylating agent, for the treatment of high-risk myelodysplastic syndrome: a multicenter Phase II study in elderly patients. J. Clin. Oncol., 18: 956–962, 2000.
6. Piro, L. D., Carrera, C. J., Carson, D. A., and Beutler, E. Lasting remissions in hairy-cell leukemia induced by a single infusion of 2-chlorodeoxyadenosine. N.Engl. J. Med., 322: 1117–1121, 1990.
7. Montgomery, J. A., Shortnacy-Fowler, A. T., Clayton, S. D., Riordan, J. M., and Secrist, J. A., III. Synthesis and biologic activity of 2-fluoro-2-halo derivatives of
9. D-arabinofuranosyladenine. J. Med. Chem., 35: 397–401, 1992.
8. Parker, W. B., Allan, P. W., Hassan, A. E., Secrist, J. A., III, Sorscher, E. J., and Waud, W. R. Antitumor activity of 2-fluoro-2-deoxyadenosine against tumors that express Escherichia coli purine nucleoside phosphorylase. Cancer Gene Ther., 10: 23–29, 2003.
9. Carson, D. A., Wasson, D. B., Esparza, L. M., Carrera, C. J., Kipps, T. J., and Cottam, H. B. Oral antilymphocyte activity and induction of apoptosis by 2-chloro-2-arabino-fluoro-2deoxyadenosine. Proc. Natl. Acad. Sci. USA, 89: 2970–2974, 1992.
10. Parker, W. B., Shaddix, S. C., Rose, L. M., Shewach, D. S., Hertel, L. W., Secrist, J. A., III, Montgomery, J. A., and Bennett, L. L., Jr. Comparison of the mechanism of cytotoxicity of 2-chloro-9-(2-deoxy2-fluoro--D-arabinofuranosyl) adenine, 2-chloro-9-(2-deoxy-2-fluoro- -D-ribofuranosyl)adenine 2-chloro-9-(2-deoxy-2, 2-difluoro--D-ribofuranosyl)adenine in CEM cells. Mol. Pharmacol., 55:515–520, 1999.