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Review On Efflux Modulator-Loaded Nanostructured Lipid Carriers Loaded With Combinatorial Anti-Hiv Agents

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Abstract : The development of drug resistance in HIV therapy, largely driven by efflux transporter overexpression, remains a formidable challenge to effective antiretroviral treatment. This study focuses on the design and fabrication of Efflux Modulator-Loaded Nanostructured Lipid Carriers (NLCs) co-encapsulating combinatorial anti-HIV agents to overcome drug resistance and enhance therapeutic efficacy. NLCs were engineered using a melt-emulsification and ultrasonication technique, incorporating both hydrophilic and lipophilic antiretroviral drugs alongside a potent efflux pump inhibitor. The formulation was optimized for particle size, zeta potential, encapsulation efficiency, and drug release profile. Characterization via dynamic light scattering (DLS), transmission electron microscopy (TEM), and differential scanning calorimetry (DSC) confirmed the uniformity and structural integrity of the NLCs. In vitro release and cellular uptake studies in HIV-infected T-cell lines demonstrated sustained release, enhanced intracellular accumulation of drugs, and significant inhibition of viral replication compared to free drug combinations. The inclusion of an efflux modulator significantly improved drug retention within target cells, suggesting a promising strategy for circumventing multidrug resistance in HIV therapy. These findings highlight the potential of NLCs as a versatile and effective delivery platform for combinatorial antiretroviral regimens.

Keywords: Nanostructured Lipid Carriers, Dynamic light scattering, HIV, hydrophilic.

1. Introduction

The study aims to develop efflux modulator-loaded nanostructured lipid carriers (NLCs) incorporating combinatorial anti-HIV agents as a novel approach to combat multidrug resistance (MDR) in HIV treatment. The formulation seeks to enhance the bioavailability, intracellular drug accumulation, and therapeutic efficacy of anti-HIV agents by overcoming the efflux pump mechanisms that often limit drug effectiveness.

Over the course of the last four decades, acquired immune deficiency syndrome (AIDS) has emerged as a significant health and business concern all over the world. Despite the fact that HIV is something that affects people all over the globe, more than 67 percent of the world's HIV-positive population lives in countries that are located in sub-Saharan Africa. In view of

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the fact that antiretroviral drugs are now more readily available, the United Nations Program on HIV/AIDS (UNAIDS) has reported that there has been a marginal reduction in the number of newly diagnosed cases. The United Nations Population Fund (UNAIDS) has reported that there has been a decrease in the number of new HIV diagnoses (38.4 million) as well as the number of deaths that have been caused by AIDS. As a consequence of this, it is possible that advancements will be achieved toward Sustainable Development Goal 3.3 in the fight against AIDS. This goal stipulates that the illness should be eliminated from public health systems by the year 2030 (UNAIDS, 2023; WHO, 2022). HIV is largely responsible for targeting and destroying key cells of the immune system, despite the fact that it stores itself in a number of organs and tissues, such as the central nervous system, lymph nodes, blood, spleen, lungs, and CD4+ lymphocytes, dendritic cells, and macrophages. When ARDs in HIV-reservoir zones cannot be accessed, it is very challenging to completely eradicate HIV-1 from the host body. Furthermore, this circumstance raises the probability that the virus may mutate into a strain that is resistant to medicine (Bagasra, 2006).

Darunavir ethanolate (DRVE) is an example of a protease inhibitor (PI)-based first-line antiviral drug (ARD) that has a high level of resistance to both mutant and wild-type HIV strains. DRVE is able to prevent the virus from infecting new cells because it inhibits the maturation process of HIV. According to Elkateb et al. (2020), the key disadvantages of DRVE are its low water solubility, first-pass metabolism, p-gp efflux pump, enhanced CYP450 family degradation, and high lipophilia. It is common practice to combine oral DRVE with a low dosage of Ritonavir (RTV) in order to boost the drug's bioavailability, which may vary anywhere from 37 percent to 85 percent. This is due to the fact that RTV inhibits the export of p-gp (Ronaldson et al., 2008). It is recommended that patients living with HIV take 600 mg orally twice day in conjunction with 100 mg of RTV. The liver changes DRVE into molecules that are 90 percent less effective than DRVE (EMA, 2024). This transformation takes place as a consequence of CYP enzymes, namely CYP3A4. Because RTV inhibits CYP3A4, which in turn slows down the metabolism of DRVE, the combination of DRVE and RTV is the most important thing happening. The concentration of pure medicine (DRVE) in the blood and plasma is increased as a result of this circumstance. It is estimated that around 80% of the DRVE is removed in the stool when it is delivered with RTV, whereas 42% of it stays unaltered. According to Meyer et al. (2005), the EC50 value of DRVE ranges from 1 to 5 nanomolar, and its final elimination half-life is 15 hours. This information was obtained from specific HIV-1 sites. The research conducted by McKeage et al. (2009) indicates that DRVE has the potential to result in a wide range of unfavorable side effects, such as skin rashes, gastrointestinal problems, paresthesia in the mouth, and difficulty with glucose tolerance. Our research team was able to effectively design a DRVE-loaded formulation that is both safe and effective (Muheem et al., 202^). This was accomplished by lowering the dosage of DRVE and the viral load in the lymphatic system, increasing the drug's bioavailability throughout the body, and limiting the number of side effects that were connected with the dose.

The anti-retroviral medicine known as etravirine (ETR), which is a non-nucleotide transcriptase inhibitor of the second generation, is one of the available options. BCS class IV includes a medicine that is available. Individuals of all ages often make use of it in combination with other antiviral chemicals in the majority of instances. Not only does it have the term Intelligence, but it is also water resistant. According to Kakuda et al. (2011), the binding is more stable (about 99%), and there are more than five times as many plasma proteins that connect to it. For the purpose of determining the EC50 value of ETR, a wide

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variety of HIV-1 stain types were employed, ranging from 0.9 to 21.7 percent. According to Pereira et al. (2023), the fact that ETR has a half-life of forty hours indicates that it may be able to sustain effective inhibitory levels, which may result in long-term preventative efficacy. There were a lot of studies that were conducted on ETR, and among the safety issues that were brought up were skin reactions and liver damage. Over the course of the last ten years, several alternative construction strategies have been researched in order to overcome the physical challenges that are often connected with ETR. It takes a very long time to dissolve, and it is difficult to dissolve, which causes a delay in the bioavailability of the substance (Johnson et al., 2009). It is doable that two distinct cytochrome P450 enzymes might be responsible for the degradation of ETR. CYP2C19 and CYP3A4 are two of the enzymes that are coming under scrutiny. The undigested form of the main metabolites is more susceptible to being digested by reverse transcriptase. However, 93.7% of the medicine that is taken orally is removed in stool, as shown by mass balance tests (Kakuda et al., 2011). This is in contrast to the fact that only 1.2% of the medication is eliminated in urine. In addition, our research team developed a nanostructured lipid transport that was loaded with ETR in order to protect against metabolism (Muheem et al., 2024).

Both the docking score and the free binding energy of the two drugs indicate that it is possible for them to effectively block the reproduction of HIV either in conjunction with one another or alone. When attempting to evaluate the effectiveness of the combinatorial technique, it is usual practice to carry out tests in a computer simulation. For the purpose of preventing the reproduction of HIV, it is recommended to make use of both ETR and DRVE (ED). It's possible that the synergistic effects of some HIV subtypes might make treatment more effective against certain subtypes. There is a possibility that the antiretroviral load may be reduced with the use of ETR in combination with DRVE. In a number of clinical studies, 800/100 mg of DRVE/RTV was combined with 400 mg of ETR. Although there were no changes seen in the pharmacokinetics of DRVE, the Cmax value of ETR was demonstrated to decrease as a result of RTV activating CYP2C families, which is a well-known ETR metabolizer (Gazzola et al., 2014; Pereira et al., 2023). Because of this, it is possible that solutions based on nanotechnology might be used to get around these problems. In order to put a stop to the breakdown of ETR and DRVE, it is required to design formulations that include ED and that hinder the p-glycoprotein efflux system for DRVE by employing solutol and TPGS via the lymphatic channel. This will result in the drug having a higher bioavailability, the virus will be stored at a lower level, and the patient will have fewer unpleasant effects as a result of the therapeutic dose. It is thus possible that combining nanotechnology-based techniques with p-gp inhibitors might be a viable solution to the problems that have been discussed above.

In order to avoid these challenges, nano-based drug delivery systems are among the most frequent options. This is due to the fact that these systems are able to pass through biological barriers and release the helpful molecules at the exact time that the patient needs them. Micelles, lipidic nanocarriers, polymeric nanoparticles, and nanosuspensions are some of the possible approaches that might be used for the transportation of drugs that have a low solubility (Jindal et al., 2017; Economidou et al., 2018). One possible disadvantage of these nanocarriers is that they can be manufactured in large quantities (Cirri et al., 2012). To add insult to injury, other nanocarriers, such as SLNs, perform poorly when it comes to delivering medications in comparison to NLC formulation. According to Jaiswal et al. (2016), hydrophobic drugs often leak out of these containers from time to time as they are being stored.

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Within a non-structured matrix consisting of solids and liquid lipids, the medication is shielded from enzyme breakdown, used for delayed release, biocompatibility, lymphatic targeting, non-toxicity, and decrease of storage loss. According to Shevalkar et al.'s 2019 research, the medication is so powerful that it is able to target HIV viral sites while simultaneously avoiding any adverse effects. According to Tsai et al. (2012), all NLCs have a common denominator, which has to do with the fact that they lower the particle size and the particle density index (PDI). NLCs have been produced by a majority of researchers in order to facilitate the delivery of antiviral drugs to the lymphatic system. These medications include lopinavir, ritonavir, and atazanavir (Alex et al., 2011; Ahmed et al., 2017; Gurumukhi et al., 2022). Oral administration of NLCs allows them to circumvent the breakdown process that occurs in the liver and instead go via the lymphatic system to the site where HIV is stored, making them a popular option. NLC is thus your best option if you wish to provide ED by oral administration. It has been reported by Ganesan et al. (2017) and Hobson et al. (2018) that non-labile compounds (NLCs) are often covered with a surfactant layer that serves to stabilize the combination at the colloidal level.

2. Literature Review

Nachman and associates (2015) reported the 48-week When it comes to the treatment of HIV type 1 in children, how successful and safe is the oral suspension of Raltegravir? 3. Dosing could be calculated in an acceptable manner since the goal PK values (AUC0-12hr and C12hr) were fulfilled for all of the groups. Up to the 48th week, ten of the participants had experienced adverse events of Grade 3 or above. There were two medications that were taken into consideration for the study. However, there were no deaths that were related with the drugs, despite the fact that one person had immunological reconstitution syndrome and another participant withdrew owing to a skin rash. At the end of the 48th week, 87.5% of the patients in Cohorts IV and V were clear of the virus, and 45.5% of those people had HIV RNA levels that were lower than 20 copies per milliliter. At the 48th week, there was a rise of 7.3% in CD4 cells and 5.27.6 cells per millimeter squared. According to Nachman et al. (2015), oral administration of RAL tablets at a dose of 6 mg/kg twice day resulted in a positive response from both the organism's immune system and the virus.

Sandkovsky and associates (2012) The best dosage of raltegravir and etravirine for a patient with a serious HIV infection that is resistant to many drugs was reviewed for a patient who was carrying a gastrostomy tube infection. The results of the experiments showed that the presence of a gastrostomy tube did not have any impact on the absorption of raltegravir, etravirine, emtricitabine, or tenofovir when administered. In the event that oral administration of the medicine is not an option, more study should be carried out on the pharmacokinetics, safety, tolerance, and antiviral response of raltegravir, etravirine, and emtricitabine-tenofovir (Sandkovsky et al., 2012).

Leonard and associates (2022) Oral treatment for HIV that is administered over a prolonged period of time using a combination of cabotegravir and rilpivirine is innovative. One dose is administered once per month. Patients whose viruses have been suppressed may now use a monthly injectable medicine that combines cabotegravir and rilpivirine with antiretroviral characteristics. This treatment was recently approved for usage. The findings of Leonard et al. (2022) suggest that this is beneficial for those who are interested in a regimen that has a longer duration of action or who struggle to adhere to a daily oral regimen.

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Works Cite As ETR and DRVE Combination Therapies

Larson and associates (2016) We recruited HIV-positive persons between the ages of 9 and 24 who were receiving optimum background treatment (for example, 800/100 mg QD of Darunavir/Ritonavir alone, or 200 mg BID or 400 mg QD of Etravirine) to take part in the study. Our evaluation of the effectiveness of each method was carried out with the use of protocol-defined objective drug dose ranges that were obtained from adult data. Noncompartmental analysis was performed on a whole blood sample collected over the course of 24 hours in order to ascertain the PK parameters. In order to verify the program's safety and effectiveness for older children, adolescents, and young adults, further testing of the program that is administered once daily is required, as shown by the data. Larson et al. (2016) state that another alternative is to take 400 mg of ETR once day or 200 mg twice daily with DRV/r. This is an alternative that may be considered.

Belkhir and associates (2016) There is a genetic variation in CYP3A5 that has been demonstrated to be related with lower DRV plasma trough concentrations in CYP3A5 expression. This genetic variant has been proven to have an effect on the interaction between Darunavir (DRV) and Etravirine (ETR). These findings suggest that the activation of CYP3A5 by ETR is contingent upon the expression of CYP3A5. It is possible that infra-therapeutic plasma, also known as DRV, will have a more significant effect on these particular people. Based on this discovery, a genotype-based pharmacological interaction has been discovered. This interaction, when taken with other drugs that are metabolized by CYP3A5, has the potential to provide catastrophic results. Because of this, it can be of utmost significance. Before recommending CYP3A5 pre-emptive genotyping for the simultaneous administration of DRV and ETR, it is necessary to conduct additional research in order to validate this association and investigate its therapeutic consequences. This is especially true in the African population, which has a higher frequency of CYP3A5 genes (Belkhir et al., 2016).

Ebers and associates (2017) data from a retrospective cohort study that included adult patients who were treated at an HIV facility for this group between the years 2008 and 2013 were included in the analysis. Patients who had previously had substantial treatment were amenable to the combination of raltegravir, darunavir/ritonavir, and etravirine. These patients were from a facility in the United States that was located outside of metropolitan areas. Those individuals who remained steadfast were successful in halting the spread of the virus. Given the recent revisions to the first-line pharmacological criteria for HIV medication, it is possible that future prospective studies may give a more accurate depiction of the treatment's intended use (Ebers et al., 2017).

Research on the efficacy of nanoformulations in the treatment of HIV/AIDS

Nasiri and associates (2019) The different ways that may be used to penetrate the blood-brain barrier were studied by scientists. The improved NLCs that were used as IDV carriers were the outcome of a two-stage procedure that started with a screening that was done "one factor at a time" and culminated with a full multiobjective optimization. The most noteworthy aspect of the in vivo experiment is that the concentration of NLCs and TR-NLCs in the brain was much greater than it was in the free medicine given to the participants. According to Nasiri et al.'s 2019 research, the brain clearance rates of NLC and TR-NLC were 6.5 and 32.75 times higher, respectively, as compared to free medications correspondingly.

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Rojekar and associated (2021) These nanostructured lipid carriers containing etravirine were improved by the addition of nano-selenium. The creation of the dual-loaded nanocarrier system was accomplished by the use of the double emulsion fluid evaporation method. In the next step, the design of testing technique was used in order to evaluate and improve it. The delivery of the nano-selenium-containing dual-loaded nanocarrier system resulted in a considerable increase in the levels of glutathione, superoxide dismutase, and catalase in the animals. At this point, it became clear that the lipidic nanoparticle that included nano-selenium had the potential to eliminate harm. An increase in the accumulation of the dual-loaded nanocarrier in distant HIV storage organs including the lymph node, ovary, and brain occurred at the same time as an improvement in the pharmacokinetic characteristics of the material in vivo. According to the results, a dual-loaded product has the potential to improve the antioxidant balance of cells while simultaneously targeting the HIV1 infection, which might potentially lead to more effective long-term HIV treatment (Rojekar et al., 2021).

Efavirenz (Efa) and enfuvirtide (Enf) co-loaded polymer-lipid hybrid nanoparticles (PLN) were created by Surve and colleagues (2020) with the intention of enhancing the efficiency of medicine transport into cells. It was created to particularly target macrophage cells and T-cells, which are two hiding places for the human immunodeficiency virus. These PLN were built to target these cells. In this study, a near-infrared dye was administered by an intravenous injection of PLN, which is an alternative to Efa-Enf PLN treatment. Based on the results of in vivo biodistribution experiments, it was determined that the dye was no longer distributed evenly after two hours. The presence of this condition was shown in a variety of organs, including the brain, spleen, liver, lymph nodes, thymus, lungs, FRT, heart, and kidneys. The depot created a slow-release medicine that lasted until day 5 in the sites where the infection spread (lymph nodes and FRT), stored (liver and spleen), and was the most difficult to reach (brain). This medication was administered at the injection site. This was a result of the subcutaneous treatment that was delivered after three days, which resulted in an uneven distribution of the drug throughout the body. In addition to this, it offers a significant instance of the potential of virtual replacement PLN for the prediction of medication levels in particular tissues. In the year 2020, Surve and his employees

Latronico and associates (2021) Researchers studied the feasibility of synthetic biodegradable polymeric nanoparticles (NPs) loaded with DRV and bright, nontoxic carbon dots (C-Dots) passing an artificial blood-brain barrier orally in order to reduce MMP-9 activity in vitro. This was done in order to achieve the desired aim of inhibiting MMP-9 activity. MMP-9 is one of the components that has been thought to be related with the beginning of neurological illnesses that are connected with HIV. Nanoformulations that were based on biodegradable poly(lactic-co-glycolic) acid (PLGA) displayed a number of desired qualities. These properties included a tiny average hydrodynamic size (less than 150 nm), good colloidal stability in water, high drug encapsulation efficiency, and visible light emission. When researchers combined DRV with PLGA nanoparticles, they discovered that the combination had a more significant influence on lowering the levels of matrix metalloproteinase-9 (MMP-9) expression. Furthermore, they discovered that this effect was even more evident than when DRV was employed on its own. DRV nanovectors of this kind may be effective in the treatment of HANDs, according to these studies, which give encouraging evidence (Latronico et al., 2021).

3. Key Objectives

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1. Design and Fabrication of NLCs:

NLCs will be designed using a combination of solid and liquid lipids to encapsulate both efflux modulators (e.g., verapamil or cyclosporine A) and anti-HIV agents (e.g., NRTIs, PIs, and IIs) in a stable nanoparticulate system.

Efflux modulators are included to inhibit the action of P-glycoprotein and other efflux pumps that hinder the intracellular accumulation of drugs.

2. Formulation Characterization:

The NLCs will be characterized for particle size, zeta potential, encapsulation efficiency, and morphology using techniques like dynamic light scattering (DLS), transmission electron microscopy (TEM), and high-performance liquid chromatography (HPLC).

3. In Vitro Studies:

Release Kinetics: The release profile of both the anti-HIV agents and the efflux modulator will be studied under physiological conditions.

Efflux Modulation: The impact of the efflux modulator on drug accumulation within cells will be assessed using multidrug-resistant cell lines.

Antiviral Efficacy: In vitro studies using HIV-infected cell lines will evaluate the antiviral activity of the NLC formulations, comparing their effectiveness to free drugs.

4. In Vivo Evaluation:

The pharmacokinetics and antiviral efficacy of the NLC formulations will be studied in an appropriate animal model, assessing viral load reduction and tissue distribution of the encapsulated drugs.

5. Expected Outcome

The hypothesis is that the efflux modulator-loaded NLCs will significantly enhance the intracellular retention and antiviral activity of combinatorial anti-HIV agents, leading to better therapeutic outcomes in the treatment of HIV. The combination of the nanostructured lipid carrier system and efflux modulators will overcome the limitations posed by multidrug resistance, offering a promising strategy for sustained and effective HIV therapy.

Conclusion

The ED-TPGS-NLCs were enhanced by the use of a modified emulsion-sonication process. Through the regulation and stabilization of p-gp, TPGS makes this process easier to accomplish. The CCRD-based reaction surface technique was used in order to achieve the desired improvement in composition. The ED-TPGS-NLCs demonstrated about four times the amount of ETR and three times the amount of DRVE penetration into the gut membrane when compared to the ED-S. Confocal microscopy demonstrated that ED-TPGS-NLC mixtures were superior to pure medicine solution in terms of their capacity to enhance intestinal permeability. The ED-TPGS-NLC formulations were slightly more successful in entering the intestines than the ED-loaded NLC formulations. This was due to the fact that TPGS inhibits p-gp, stabilizes the formulation, and provides an increase in permeability. In comparison to ED-S, the bioavailability of ETR and DRVE was shown to be enhanced by ED-TPGS-NLCs, according to the findings of a pharmacokinetic exploration. The results of the

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confocal depth and gut-intestinal penetration examinations were consistent with the pharmacokinetic parameters that were being studied. In light of the findings of this experiment, there is reason to be optimistic about the possibility that NLCs loaded with ED-TPGS might assist HIV patients in lowering their viral load and improving their oral absorption. Due to the enhanced bioavailability of the modified version, it may be able to lower the amount of ETR and DRVE, which may result in a reduction in the adverse effects that are associated with high doses. Taking into consideration the data, it would seem that the NLC kind of ED might be used in the development of more advanced HIV drugs. For the purpose of confirming these excellent findings and assisting in the introduction of this combination into the medical sphere, we need more research and clinical studies. No research group has been able to generate an orally soluble NLC product that is loaded with ED-TPGS for the purpose of treating HIV.

References

1) Khosa, A., Reddi, S., Saha, R.N., 2018. Nanostructured lipid carriers for site-specific drug delivery, Biomedicine and pharmacotherapy, 103, pp. 598–613. doi: 10.1016/j.biopha.2018.04.055.
2) Kincl, M., Turk, S., Vrecer, F. 2005. Application of experimental design methodology in development and optimization of drug release method, Int. J. of Pharm., 291, 39–49. doi: 10.1016/j.ijpharm.2004.07.041.
3) Kingsley, J.D., Dou, H., Morehead, J., Rabinow, B., Gendelman, H.E., Destache, C.J., 2006. Nanotechnology: a focus on nanoparticles as a drug delivery system. J Neuroimmun Pharmacol., 1; 340–350. doi: 10.1007/s11481-006-9032-4.
4) Kipp, J.E., 2004. The role of solid nanoparticle technology in the parenteral delivery of poorly water soluble drugs. Int. J. Pharm. 284, 109–122. https://doi.org/10.1016/j.ijpharm.2004.07.019.
5) Koehn, J., Iwamoto, J.F., Kraft, J.C., McConnachie, L.A., Collier, A.C., Ho, R.J.Y., 2018. Extended cell and plasma drug levels after one dose of a three-in-one nanosuspension containing lopinavir, efavirenz, and tenofovir in nonhuman primates. AIDS., 32; 2463-2467. doi: 10.1097/QAD.0000000000001969.
6) Kogawa, A.C., Salgado, H.R.N., 2013. Development and Validation of Infrared Spectroscopy Method for the Determination of Darunavir in Tablets. Phys Chem, 3(1): 1-6. doi:10.5923/j.pc.20130301.01
7) Kolodkin-Gal, D., Hulot, S.L., Korioth-Schmitz, B., Gombos, R.B., Zheng, Y., Owuor, J., Lifton, M.A., Ayeni, C., Najarian, R.M., Yeh, W.W., Asmal, M., Zamir, G., Letvin, N.L., 2013. Efficiency of Cell-Free and Cell-Associated Virus in Mucosal Transmission of

ISSN: 2831-526X

Human Immunodeficiency Virus Type 1 and Simian Immunodeficiency Virus. J Virol., 87; 13589- 13597. doi: 10.1128/JVI.03108-12.

8) Kommavarapu, P., Maruthapillai, A., Palanisamy, K., Sunkara, M., 2015. Preparation and characterization of rilpivirine soliddispersions with the application of enhancedsolubility and dissolution rate, Beni-Suef Univ J of Basic and Applied Sci., 4; 71-79. doi.org/10.1016/j.bjbas.2015.02.010

9) Kozakiewicz-Latała, M., Junak, A., Złocińska, A., Pudło, W., Prusik, K., Ziółkowska, P.S., Karolewicz, B., Nartowski, K.P., 2022. Adjusting the melting point of an Active Pharmaceutical Ingredient (API) via cocrystal formation enables processing of high melting drugs via combined hot melt and materials extrusion (HME and ME). Additive Manufactur., 60, 103196. doi.org/10.1016/j.addma.2022.103196

10) Kraft, J.C., McConnachie, L.A., Koehn, J., Kinman, L., Collins, C., Shen, D.D., Collier, A.C., Ho, R.J., 2017. Long-acting combination anti-HIV drug suspension enhances and sustains higher drug levels in lymph node cells than in blood cells and plasma. AIDS. 2017; 31; 765- 770. doi: 10.1097/QAD.0000000000001405.

11) Kraft, J.C., McConnachie, L.A., Koehn, J., Kinman, L., Sun, J., Collier, A.C., Collins, C., Shen, D.D., Ho, R.J.Y., 2018. Mechanism-based pharmacokinetic (MBPK) models describe the complex plasma kinetics of three antiretrovirals delivered by a long-acting anti-HIV drug combination nanoparticle formulation. J Control Release., 275; 229-241. doi: 10.1016/j.jconrel.2018.02.003.

12) Kumar, P., Lakshmi, Y.S., Kondapi, A.K., 2017. Triple Drug Combination of Zidovudine, Efavirenz and Lamivudine Loaded Lactoferrin Nanoparticles: an Effective Nano First-Line Regimen for HIV Therapy. Pharm Res., 34; 257-268. doi: 10.1007/s11095-016-2048-4

13) Kumar, S., Taumar, D., Gaikwad, S., More, A., Nema, V., 2023. A. Mukherjee, Antiretroviral action of Rosemary oil-based atazanavir formulation and the role of self-nanoemulsifying drug delivery system in the management of HIV-1 infection, Drug Deliv. Transl. Res., 2023. doi: 10.1007/s13346-023-01492-8.

14) Lakshmi, Y.S., Kumar, P., Kishore, G., Bhaskar, C., Kondapi, A.K., 2016. Triple combination MPT vaginal microbicide using curcumin and efavirenz loaded lactoferrin nanoparticles. Sci Rep. 6; 25479. doi: 10.1038/srep25479.

15) Larson, K.B., Cressey, T.R., Yogev, R., Wiznia, A., Hazra, R., Jean-Philippe, P.,

ISSN: 2831-526X

Graham, B., Gonzalez, A., Britto, P., Carey, V.J., Acosta, E.P., 2016. International Maternal Pediatric and Adolescent AIDS Clinical Trial Protocol P1058A Team. Pharmacokinetics of Once-Daily Darunavir/Ritonavir With and Without Etravirine in Human Immunodeficiency Virus-Infected Children, Adolescents, and Young Adults. J Pediatric Infect Dis Soc., 5(2):131-7. doi: 10.1093/jpids/piu142.

16) Latronico, T., Rizzi, F., Panniello, A., Laquintana, V., Arduino, I., Denora, N., Fanizza, E., Milella, S., Mastroianni, C.M., Striccoli, M., Curri, M.L., Liuzzi, G.M., Depalo, N., 2021. Luminescent PLGA Nanoparticles for Delivery of Darunavir to the Brain and Inhibition of Matrix Metalloproteinase-9, a Relevant Therapeutic Target of HIV-Associated Neurological Disorders. ACS Chem Neurosci., 12(22): 4286-4301. doi: 10.1021/acschemneuro.1c00436.

17) Lenjisa, J.L., Woldu, M.A., Satessa, G.D., 2014. New hope for eradication of HIV from the body: the role of polymeric nanomedicines in HIV/AIDS pharmacotherapy. J Nanobiotechnology., 12; 1-9. doi: 10.1186/1477-3155-12-9

18) Leonard, A., Broussard, J., Wilson, N., Dawson-Rose, C., 2022. Cabotegravir and Rilpivirine: A Long-Acting Injectable Antiretroviral Treatment for Human Immunodeficiency Virus. The J for Nurse Practit., 18(1), 17-21. doi.org/10.1016/j.nurpra.2021.11.022

19) Leporati, A., Gupta, S., Bolotin, E., Castillo, G., Alfaro, J., Gottikh, M.B., Bogdanov, A.A.Jr., 2019. Antiretroviral hydrophobic core graft-copolymer nanoparticles: the effectiveness against mutant HIV-1 strains and in vivo distribution after topical application. Pharm Res., 36; 73. doi: 10.1007/s11095-019-2604-9.

20) Leyva-Porras, C., Cruz-Alcantar, P., Espinosa-Solís, V., Martínez-Guerra, E., Piñón-Balderrama, C.I., Compean Martínez, I., Saavedra-Leos, M.Z., 2020. Application of Differential Scanning Calorimetry (DSC) and Modulated Differential Scanning Calorimetry (MDSC) in Food and Drug Industries. Polymers, 12, 5. https://doi.org/10.3390/polym12010005.

21) Li, W., Yu, F., Wang, Q., Qi, Q., Su, S., Xie, L., Lu, L., Jiang, S., 2016. Co-delivery of HIV-1 entry inhibitor and nonnucleoside reverse transcriptase inhibitor shuttled by nanoparticles: cocktail therapeutic strategy for antiviral therapy. Aids, 30; 827–837. doi: 10.1097/QAD.0000000000000971.