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Pharmacokinetics and biodistribution of zidovudine loaded in a solidified reverse micellar delivery system.

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Abstract

The aim of the research was to study the stability, release profile, pharmacokinetic and biodistribution properties of zidovudine (AZT)-solidified reverse micellar microparticulate. Lipid matrices formulated with Phospholipon® 90H and goat fat at ratios of 1:1, 2:1, 3:1 and 2:3 were used to prepare AZT-loaded SLM by melt dispersion followed by lyophilization. *In vitro* release studies of the drug were carried out using a sequential drug release method in both SGF (pH 1.2) and SIF (pH 7.2) while the in vivo drug release studies were carried out using Wistar albino rats. The result of our findings showed that the drug is compatibility with the lipid matrix with the 1:1 showing the most stable microparticle preparation which was then optimized. The formulations showed a concentration dependent increase in their concentration maximum (Cmax) with values of 116.05 µg/ml, 124.21 µg/ml, 128.95 µg/ml, 138.95 µg/ml and time to reach maximum concentration (Tmax) values of 5h, 8 h, 8 h, and 5 h for batches B1, B2, B3 and B4 containing 1 %, 2 %, 3 % and 5 % of AZT respectively. The area under curves (AUCs) of the microparticles formulated showed that the bioavailabilities of the microparticles were comparable to that of the conventional release tablet. The biodistribution studies of the microparticles in rats showed highest concentration of the drug in the liver with the least in the brain and higher biodistribution in various organs than pure AZT. The data suggested that SLM could be a promising drug delivery system to improve on the shortcomings of pharmacokinetics and bio-distribution properties of conventional AZT tablets like fluctuation in blood levels of the drug.

Keywords: solidified lipid microparticle (SLM), lipid matrix, Phospholipon® 90H, human immunodeficiency virus, highly active antiretroviral therapy.

Introduction

Tens of millions of people are currently infected by human immunodeficiency virus-1 (HIV-1) and millions of new infections occur each year [1-6]. HIV-1 infection leads to acquired immunodeficiency syndrome (AIDS) as a result of low levels of $CD4^+$ T cells that are directly killed by human immunodeficiency virus (HIV) and/or by apoptosis of infected $CD4^+$ T cells and indirect killing by CD8 cytotoxic T-lymphocytes. Zidovudine (AZT), a nucleoside reverse transcriptase inhibitor (NRTI), is the first antiretroviral drug that has proven effective in preventing vertical transmission of HIV, thus reducing mother-to-child HIV transmission (MTCT) from 25 % to 8 % in the absence of breastfeeding [7]. Conventional AZT tablets have systemic availability of 65 % (range 52 to 75 %), while its concentration is maintained within the therapeutic range of 106.9-1069 µg/L. It is 30-38 % protein bound and is metabolized in the liver with a half-life of 3-5 h and renally excreted. According to Vanhove *et al.* [8] using parametric model; the mean pharmacokinetic parameters, the Tmax, Cmax, AUC, $T_{1/2}$ of AZT were 1.29 h, 347 ng/ml, 1260 ng h/ml, 0.76 h for AZT respectively. Modern treatment regimens typically use lower dosages of zidovudine (e.g. 300 mg) two times a day. As of 1996, AZT, like other antiretroviral drugs, is almost always used as part of highly active antiretroviral therapy (HAART). That is, it is combined with other drugs in order to prevent mutation of HIV into an AZT-resistant form [9, 10]. Like most other drugs, this drug had its own share of side effects. Also, the necessity of taking the drug daily poses a problem of compliance, since the conventional 300-mg tablet dosage form is taken twice daily. Despite the *in vitro* efficacy of antiretroviral drugs, AZT inclusive, pharmacological treatment is compromised by its inherent physico-chemical properties and limitations in bio-distribution and cellular uptake leading to impediment in the effective drug bioavailability owing to restricted transport across the blood-brain barrier (BBB) as human immuno viral reservoir in the central nervous system (CNS) is high. Protease inhibitors have limited ability to reach the CNS, with majority of this class of drugs not detected in human cerebrospinal fluid (CSF). Body compartments with blood-tissue barriers prevent drug penetration, thus preventing the eradication of latent viral pools. Active transport mechanisms involving proteins

of the ATP-binding cassette (P-glycoprotein) that are present, for example, in central nervous system (CNS) prevent the penetration of anti-HIV drugs into the brain [11]. However, many promising anti-retroviral (ARV) agents are unfortunately compromised by disadvantageous physicochemical properties which lead to poor biodistribution and insufficient cellular uptake [12, 13]. Owing to the shortcomings in pharmacokinetic data (very short half due to first-pass metabolism and fast renal excretion) and variable bio-distribution (poor BBB transport and cerebrospinal fluid barrier) there is need to formulate these drugs in novel drug delivery system like solid lipid microparticles (SLM)s.

The very slow progress in the efficacy of the treatment of severe diseases has suggested a growing need for a multidisciplinary approach to the delivery of therapeutics to targets in tissues [14]. There is growing interest and investment in the use of lipid-based systems in drug discovery and product development to effectively overcome physical and biological barriers related to poor aqueous solubility and stability, membrane permeability, drug efflux and availability [15]. Solid lipids have the advantage of providing more flexibility in controlling the drug release and protecting the encapsulated ingredients from chemical degradation. Also, they allow for the incorporation of hydrophilic or hydrophobic drugs [16, 17]. SLMs can avoid use of organic solvent, non-toxicity of the carrier system, chemical and physical storage stability for both the carrier and the drug, low cost of ingredients, ease in the preparation and high scale-up potential [18-20]. Controlled drug delivery technology represents one of the frontier areas of science which involves a multidisciplinary scientific approach, contributing to human health care. The need for non-invasive drug delivery systems is a result of patient's non compliance and poor acceptability of the existing delivery regimens, limited market size for drug companies and drug users, coupled with a high cost of disease management. It has been proved *in vitro* that SLMs can reasonably be considered as a suitable carrier to provide a sustained release to the incorporated substance. This work was geared towards utilizing these unique properties of lipids in formulating a sustained release form of zidovudine to be delivered once daily.

Thus, the aim of this study was to evaluate the pharmacokinetics and biodistribution of zidovudine-loaded SLMs in immune-compromised rats in comparison with the commercially available immediate release zidovudine tablets. Results of the pharmacokinetics and bio-distribution of sustained-release SLMs containing AZT in immune-compromised rats were compared with the pharmacokinetics and bio-distribution properties of conventional oral tablets of AZT.

Material and Methods

Materials

The materials used were Phospholipon® 90H, a hydrogenated lecithin (Phospholipid GmbH, Köln, Germany), thiomersal, Poloxamer 188 (a non-ionic tri-block copolymers composed of a central hydrophobic chain of polyoxypropylene flanked by two hydrophilic chains of polyoxythylene (BASF, Germany), zidovudine (a gift from Fidson Healthcare, Nigeria), hydrochloric acid, sodium chloride, sodium hydroxide, monobasic sodium phosphate (Merck, Darmstadt, Germany), cyclophosphamide (Oncomide®, Khandelwal, India) and distilled water (Lion Water, University of Nigeria, Nsukka). All other reagents and solvents were of analytical grade and were used as supplied.

Methods

Extraction of goat fat

Goat fat was extracted from the adipose tissue of *Capra hircus*. The extraneous materials were manually separated from the adipose tissue, which was then rendered by the wet process as outlined by Attama and Nkemnele [21]. The adipose tissue was grated and subjected to moist heat by boiling with about half its weight of water in a water bath for 45 min. The molten fat was separated from the aqueous phase after filtering with a muslin cloth. The fat was stored in a refrigerator until used.

Formulation of the solidified reverse micellar solution (SRMS)

According to the procedure outlined by Friedrich and Müller-Goyman [22], formulation of the solidified reverse micellar solutions (SRMS) was prepared with different ratios of goat fat and Phospholipid as follows: 1:1, 2:1, 3:1 and 2:3 respectively. All the preparations were prepared by melt solidification.

Formulation of SRMS-based solid lipid microparticles (SLMs)

The reverse micellar microparticles (solid lipid microparticles) were prepared to contain: lipid matrix (7.5 % w/w), zidovudine (0, 1, 2, 3, 5 %w/w), Poloxamer 188 (1 %w/w), thiomersal (0.001 %w/w), sorbitol (4 %w/w) and water (to 100 %w/w). The lipid matrix consisted of goat fat and Phospholipon® 90H. For each batch, the lipid matrix was placed in a stainless steel bowl and heated at 70 ^{o}C until it had completely melted. The drug was poured into the melted matrix and mixed. The remaining excipients were weighed out appropriately and mixed with the corresponding quantity of water. The excipients mixture with water at 70 C was poured into the lipid matrix-drug mixture and homogenized at 5000 rpm for 10 min with Ultra-turrax homogenizer (IKA® 25, Bonn-Bad Godesberg, Germany), a creamy emulsion was formed. The hot emulsion was then poured into bottle and allowed to recrystallize at room temperature for 24 h. The same procedure was repeated for the various lipid matrices (1:1, 2:1, 3:1 and 2:3). The dispersions were left on the shelf at room temperature to determine their short-term stability by monitoring visually the formulations daily for one week. At the end of the week, it was observed that only the 1:1 ratio was stable while the other ratios were unstable, thus they were

discarded. The stable dispersion was then lyophilized to get the microparticles.

Preparation of simulated intestinal fluid (SIF) without pancreatin

This was done by dissolving 6.8 g of monobasic potassium sulphate in 250 ml of distilled water. The resulting solution was made up to 1000 ml with distilled water. The pH of the solution was checked and adjusted to a pH (Suntex TS-2, Taiwan) of 7.2 using 0.2 N NaOH.

Preparation of simulated gastric fluid (SGF) without pepsin

A 1.0 g of NaCl was dissolved in 500 ml of distilled water and 7 ml of conc. HCl was added. The resulting solution was made up to 1000 ml with distilled water. The pH of the medium was checked with pH (Suntex TS-2, Taiwan) and adjusted to 1.2 with a 2.0 N HCl.

Preparation of calibration plot

The wavelengths of maximum absorption were determined by scanning some samples in the various media used (SIF, SGF, ethanol and plasma). From this, the calibration plots of zidovudine in these media were obtained and used to calculate the corresponding concentrations of drug released from them. A calibration curve was obtained at five concentration levels of a zidovudine standard solution (0.01- 0.06 mg/ml) showing high linearity.

Release studies

A sequential drug release in different release media was carried out. In each case a 0.5 g of each of the solidified reverse micellar microparticles (SRMMs) was placed in a dialysis membrane (MWCO 6000-8000 Spectrum Labs, The Netherland) tied at both ends and suspended in 250 ml of SGF, placed in a dissolution apparatus set to rotate at 100 rpm at a temperature of 37 ^{o}C. At intervals of 5 min, 10 min, 20 min, 30 min, 1 h and 2 h respectively, 5 ml aliquots of the dissolution medium (SGF, pH 1.2) were collected and immediately replaced with 5 ml of fresh SGF. After 2 h, the dissolution medium was changed and replaced with freshly prepared SIF (pH 7.2). The drug release in this medium was then assessed at hourly intervals for 10 h. Sequel to this the withdrawn samples were collected and analyzed using a UV-VIS spectrophotometer (UNICO-3102, England) at the appropriate predetermined wavelengths.

Determination of kinetics and mechanism of release

The dissolution data for the SLMs were also analyzed to determine the *in vitro* kinetics of release. Four kinetic models including the zero-order release equation, first order equation, Higuchi square root equation and Korsmeyer equation.

$Qt = K_o t$ (Zero order equation) ..... Eqn 1

$\ln Qt = \ln Q_o - K_1 t$ (First order equation) ..... Eqn 2

$Qt = Kh.S\sqrt{t}$ (Higuchi equation based on fickian diffusion)... Eqn 3

Where Q is the amount of drug released in time t, Q_o is the initial amount of drug in the microparticles, K_o, K_1, and K_h are the rate constants of zero order, first order and Higuchi rate equations respectively.

Also, the Peppas and Korsenmeyer equation (Power law) was used.

$Mt/M\infty = Kt^n$ ..... Eqn 4

Where M_t is the amount of drug released at time t. $M\infty$ is the amount of drug released at time $=\infty$, n is the diffusional exponent indicative of the mechanism of drug release, K is the power law constant, $Mt/M\infty$ is the fraction of the drug released; If $n \leq 0.43$, a Fickian diffusion (case I+); $0.43 \leq n < 0.89$, a non-Fickian transport and $n \geq 0.89$, a case II transport (zero order, drug release mechanism dominates).

In vivo drug release

The animal experimental protocols were in accordance with the guidelines for conducing animal experiments stipulated by our Institution's Animal Ethics Committee and in compliance with the Federation of European Laboratory Animal Science Association and the European Community Council Directive of November 24, 1986 (86/609/EEC) [23]. Fifteen albino Wistar rats weighing 180-250 g were used for the *in vivo* test. The rats were divided into five groups of three rats per group. They were fed for one week for acclimatization. Afterwards, a single dose of 30 mg/kg of cyclophosphamide (Oncomide®) was administered to the rats IP (intraperitoneally) to induce immune suppression. The immune suppressed rats were allowed to starve for 24 h. At the end of this period, blood was withdrawn from each of the rats (t=0). Subsequently, 1 mg of zidovudine was administered to the rats in one group while the equivalent weight of reverse micellar preparations that would give 1 mg of zidovudine was administered to each of the remaining four groups. After administration, blood samples were withdrawn from the retro-orbital plexus of the rats at intervals of 1, 3, 5, 8, 12 and 15 h respectively with the aid of heparinised capillary tubes. After collection, these samples were placed in EDTA bottles and refrigerated. Thereafter, the collected blood samples were centrifuged (Abishkar centrifuge, India) at 500 rpm for 10 min. This plasma was then diluted 50-fold with a plasma solution and their absorbance checked using a UV-VIS spectrophotometer at a predetermined wavelength of 255 nm. The concentration maximum (Cmax), the time to reach the concentration maximum (Tmax) and area under curve (AUC) were also determined.

Biodistribution studies

The biodistribution of zidovudine from the SRMM to some organs of the body was studied using healthy albino rats. Four rats were used for the study. A 1 ml solution of pure zidovudine (1 %) was administered to two of the rats while the remaining two rats received the equivalent dose of zidovudine from B1 batch. Distilled water was used to disperse the two drugs. They were then fed orally to the rats that had previously been injected with a dose of 30 mg/kg of cyclophosphamide and allowed to survive on water alone for 24 h. After feeding the animals, one from each group was sacrificed at intervals of 1 h and 1.5 h respectively. The sacrificed animals had their kidneys, livers, spleens and brains harvested. The harvested organs were homogenized and the homogenate soaked in ethanol for 30 min and then filtered. The filtrates were then analysed with a UV-VIS spectrophotometer at the predetermined wavelength to determine their absorbance. The absorbance values were converted to concentration by reference to Beer's plot of zidovudine in ethanol.

Data and statistical analysis

All experiments were performed at least in triplicates for validity of statistical analysis. Results were expressed as mean $\pm$ SD. ANOVA was performed on the data sets generated using Microsoft excel and differences were considered significant for p values $<$ 0.05.

Results and Discussion

A stable polydispersed SLMs was obtained.

Calibration plots

Calibration plots indicated linear relationships between absorbance and concentration of zidovudine with all the solvents used. The following relationships were obtained; Ethanol: $A_{264} = 0.333C$ ($r^2 = 0.9460$); SGF pH 1.2: $A_{267} = 0.420C$ ($r^2 = 0.9340$); SIF pH 7.2: $A_{267} = 0.486C$ ($r^2 = 0.9520$); Plasma: $A_{255} = 0.095C$ ($r^2 = 0.9910$). High linearity ($r^2 > 0.9$) was obtained for the calibration plot. With $r^2 > 0.9$ for all the solvents, proper analysis of AZT could be performed at the wavelength of maximum absorption of AZT in these solvents.

In vitro release studies

In vitro drug dissolution studies showed that the B1 formulation containing 1 % of AZT gave a gradual release of zidovudine. Batches B3 and B4 containing 3 % and 5 % of AZT respectively had an initial high release which could be adduced to the presence of zidovudine on the surface of the microparticles as a result of saturation of the lipid matrix. Reddy and Murthy [24] suggested that this initial burst release (batch B3 with 16.07 % of drug release within 5 min and batch B4 with 36.44 % of drug release within 5 min) could be as a result of adsorption on the surface of the microparticle or precipitation from the superficial lipid matrix. This could be advantageous in a formulation since it provides a quantity of drug to the body within a short time for therapeutic activity to commence before subsequent quantities are delivered gradually to maintain this activity. In contrast, the commercial zidovudine tablet released almost all of its drug content within 5 min characteristic of immediate release. According to Galinsky and Svenson [25], prolonged release dosage forms like this solidified reverse micellar microparticles have additional advantage of giving a lesser fluctuation in blood levels of the drug than with rapidly absorbed dosage forms. Particulate drug delivery systems thus perform better than tablets in prolongation of drug release.

Release kinetic studies

The result of the *in vitro* release of the different batches was fitted into zero order, first order, Higuchi and Korsmeyer models to determine the kinetics of release as shown in Table 1.

Table 1: Kinetic models for the release studies

Formulation batches of microparticle	zero order		first order		Higuchi		Korsenmeyer	
	r	K_o	r	K_1	r	K_h	r	n
B1 in SGF	0.8970	0.054	0.8370	0.007	0.9910	1.166	0.9970	0.538
B1 in SIF	0.8930	0.024	0.9390	0.002	0.9860	1.173	0.9870	0.520
B2 in SGF	0.8340	0.087	0.7860	0.008	0.9660	1.311	0.9590	0.580
B2 in SIF	0.9420	0.025	0.9330	0.002	0.9800	0.599	0.9800	0.466
B3 in SGF	0.7990	0.159	0.7850	0.020	0.9700	0.361	0.9530	0.865
B3 in SIF	0.8740	0.012	0.8710	0.001	0.9310	0.381	0.9820	0.351
B4 in SGF	0.8870	0.144	0.8050	0.010	0.9800	5.971	0.9060	0.882
B4 in SIF	0.9200	0.035	0.8330	0.002	0.9220	1.374	9.9490	0.559
AZT in SGF	0.9000	0.186	0.7350	0.008	0.9920	0.485	0.9700	0.285
AZT in SIF	0.8730	0.034	0.8230	0.002	0.9580	0.283	0.9800	0.288

∗key: r =regression coefficient, K_0= Zero order rate constant, K_1= First order rate constant, K_h= Higuchi rate constant, n = Korsenmeyer constant, B1 (contained 1 % of AZT in the solidified reverse micellar microparticle), B2 (contained 2 % of AZT in the solidified reverse micellar microparticle), B3 (contained 3 % of AZT in the solidified reverse micellar microparticle), B4 (contained 5 % of AZT in the solidified reverse micellar microparticle).

Analysis of kinetics

Determining the regression coefficients assessed the fitness of the data into various kinetic models of the batches. The rate constants for the respective models were calculated from their slopes [26]. The statistical analysis showed a high significance difference ($p < 0.05$) between the AZT-loaded SLMs and the AZT tablet.

In vivo release studies

The B1 batch containing 1 % AZT maintained a gradual release of AZT *in vivo* up to the 15th h while batches B2 and B3 batches containing 2 % and 3 % of AZT respectively had optimum release at the 8th h before their gradual decline in release while the batch B4 containing 5 % of AZT and zidovudine tablet had optimum release at the 5th h.

Pharmacokinetic parameters

The result of the Cmax, Tmax and AUC for the batches is presented in Table 2.

From the data in Table 2, the Tmax and AUCs for B1 batch containing 1 % AZT and zidovuine tablet formulations were almost similar.

Table 2: Pharmacokinetic parameters

Batch Formulations	Cmax (µg/ml)	Tmax (hrs)	AUC (µghr/ml)
B1	116.05	5	1,378.68
B2	124.21	8	1,270.28
B3	128.95	8	1,197.97
B4	138.95	5	1,314.74
AZT tabs	149.47	5	1,402.86

*key: r =regression coefficient, K_0= Zero order rate constant, K_1= First order rate constant,

K_h= Higuchi rate constant, n = Korsenmeyer constant, Cmax = maximum concentration

Tmax = time to reach maximum concentration, AUC = area under curve

Bio-distribution studies

The bio-distribution profile of AZT tablet and batch B1 containing 1 % AZT in the various organs were ascertained to determine which part of the organs has the highest bioavailability of the drug are shown Figs. 1 and 2. The bio-distribution studies of batch B1 containing 1 % AZT showed a time dependent change in the concentrations of the drug in four organs of the body: liver, kidney, spleen, and brain, with the drug achieving its highest concentration in the liver and least in the brain.

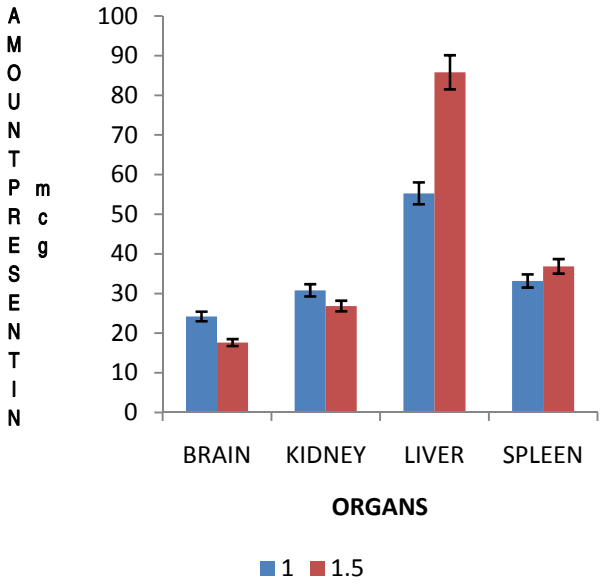

Figure 1: Biodistribution studies of zidovudine tablet
1 is the biodistribution at 1 h; 1.5 is biodistribution at 1.5 h

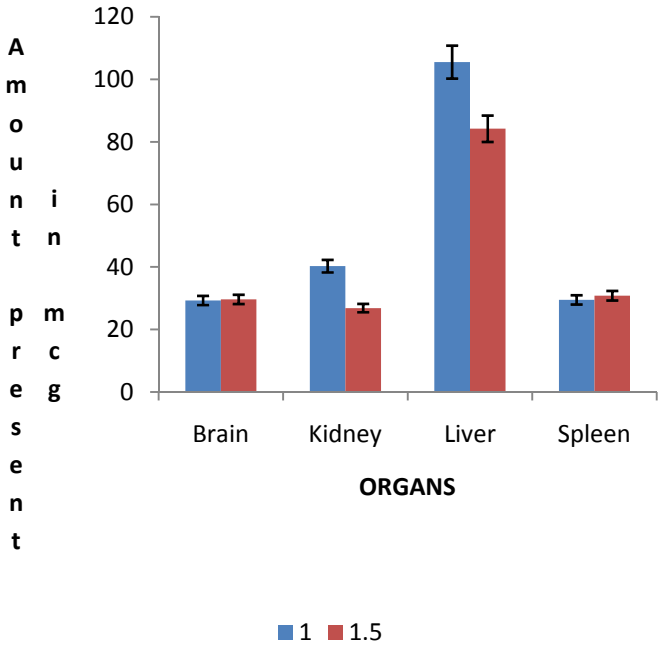

Figure 2: Biodistribution studies of batch B1 containing 1 % AZT
1 is the biodistribution at 1 h; 1.5 is biodistribution at 1.5 h

The results of the calibration plots in different media showed linearity between absorbance and concentrations of the zidovudine which implies first order kinetics. Analysis of the mechanism of drug release from the microparticles showed that the drug release followed both diffusion and dissolution mechanisms and Fickian. Release kinetic data for all the models are shown in Table 1. Drug

release data of conventional tablet was fitted into first order equation (r^2 = 0.7350 in SGF and 0.8230 in SIF). The result of the kinetic analysis of the *in vitro* release profiles (Table 1) revealed that most of the SLM batches predominantly followed the Higuchi model of release while B1 and B2 containing 1 % and 2 % of AZT respectively both showed the Higuchi and Korsmeyer models of release. This implies that zidovudine-loaded SLMs undergo a change in surface area as zidovudine dissolved and diffused out of the hydrated or solvated SLMs in these batches. Moreover, since most of the formulations followed the Higuchi square root model, indicating a diffusion controlled release as expected from a heterogeneous matrix system [27, 28], it implies that the smooth surface of the SRMS-based zidovudine-loaded SLMs have enough pores and channels to permit controlled drug release. Therefore, the study indicated combined effect of diffusion and erosion mechanisms for controlled drug release. The results obtained as shown revealed that the best fit release kinetics was obtained with the Korsmeyer's plot while the Higuchi plot also showed a high correlation for the data. This showed some potential for use of this preparation as a sustained release form for zidovudine. *In vitro* drug release studies revealed that batch B1 released its drug content gradually while batches B3 and B4 containing 3 % and 5 % respectively had an initial burst release before subsequently releasing gradually in comparison with the conventional zidovudine tablets that released most of its drug content within 5 min.

The pharmacokinetic parameters determined *in vivo* (Table 2) showed that the C_{max} of the drugs increased with increasing drug loading. Batch B4 containing 5 % AZT had the highest C_{max} of 138.95 µg/ml. However, the T_{max} were at variance with their C_{max}. Batches B2 and B3 containing 2 % and 3 % AZT respectively had similar T_{max} of 8 h while the B1 and B4 containing 1 % and 5 % respectively had a T_{max} of 5 h. The AUCs of the SLMs when compared with that of the conventional release tablet showed that the formulations had a concentration dependent decrease in their relative bioavailabilities of 0.983, 0.905 and 0.854 for batches B1, B2 and B3 respectively while that of batch B4 was 0.937. Thus drug release from the matrix of the microparticles could be described as non-Fickian for batches B1, B2 and B3 batches with values within the range: $0.43 < n < 0.85$, while drug release from the zidovudine tablet was Fickian with an *n*-value of < 0.43. This means that drug was released via a diffusion and dissolution mechanism. The difference in the release mechanisms of the formulated microparticles and the commercial tablets may be due to the tortuosity of the matrix of the microparticles, which thus restricted the path of diffusion whereas the simple tablet matrix of the commercial tablets made diffusion from its matrix easier and faster. *In vivo* studies also revealed that batch B1containing 1 % AZT showed the best properties as it sustained the drug release most (up to the 15th h) with drug plasma concentrations within the therapeutic range of zidovudine (106.7-1067.0 µg/ml). Other batches reached peak plasma concentrations at the 8th h and subsequent concentrations were below the therapeutic range of zidovudine. The biodistribution of a molecule in an organ depends on a series of pharmacokinetic parameters (physicochemical and physiological) such as administration route, dosage form, drug liberation rate, diffusion, penetration, permeation, and distribution within the body fluids and tissues; biotransformation type, amount, and rate; recycling processes; and elimination [29]. Distribution rate for each organ tissue is determined by the blood perfusion and the facility of passing through vessels and penetrating the cell [30]. Diffusion time for different tissues, identification of target tissue, and residence time are important information revealed by biodistribution studies [30]. Biodistribution studies revealed a time dependent concentration of zidovudine in the various organs tested (liver, spleen, kidney and brain) with the highest concentration occurring in the liver and the least in the brain. A comparison of the results obtained for the zidovudine tablet (Fig.1) and the batch B1 containing 1 % AZT (Fig. 2) revealed greater brain penetration of the microparticles possibly due to its higher lipophilicity thus enabling it to cross the highly lipophilic blood-brain barrier (BBB). Also other organs (liver and kidney) had a greater concentration of the drug suggesting greater cell permeation. In the brain, the amount of zidovudine tablet at 1 h and 1.50 h were 24.21 µg and 17.63 µg while that of batch B1 were 29.27 µg and 29.63 µg respectively. In the kidney, the amount of pure zidovudine at 1 h and 1.50 h were 30.79 µg and 26.84 µg while that of batch B1were 40.26 µg and 26.84 µg respectively. In the liver the amount of zidovudine tablet at 1 h and 1.50 h were 55.26 µg and 85.79 µg while that of batch B1 were 105.53 µg and 84.21 µg respectively. In the spleen the amount of pure zidovudine at 1 h and 1.50 h were 33.16 µg and 36.84 µg while that of batch B1were 29.47 µg and 30.79 µg respectively. The biodistribution of AZT from batch B1 in the kidney, brain, liver were more than the biodistribution of zidovudine tablet in those organs. At the spleen, batch B1 has a lower biodistribution than the zidovudine tablet. It is reported in the literature that more than 30% release of drug within 1 h of dissolution indicates the chance of dose dumping. The results showed probability of dose dumping from conventional AZT tablets. The incorporation of zidovudine into SLM reduced the chances of dose dumping. AZT has a very narrow (0.4-4.0 µmol/L) therapeutic index [31]. The commercially available conventional tablet produces initial high plasma concentration owing to absence of release modifiers, which may cause unwanted toxic effects like bone marrow depression that sometimes leads to withdrawal of drug therapy [32]. The smooth and extended absorption phase coupled with maintenance of plasma concentration for longer duration after administration in matrix tablets suggests reduced chance of dose-dependent side effects of AZT [33].

The bio-distribution in terms of elimination and retention of the drug in different organs vis-à-vis the normal elimination route of the drug showed; in the brain the SLM-loaded AZT has higher retention than pure AZT; in the kidney and liver the SLM –loaded AZT even though had higher bio-distribution in these organs, the elimination was also faster thereby preventing unwanted toxic effect as seen in pure AZT and in the spleen the pure AZT had higher bio-distribution and higher retention. Therefore batch B1 containing 1 % AZT with highest entrapment efficiency, loading capacity and smallest particle size when compared with other batches [34]

exhibited the best release profile up to the 15th h and better bio-distribution than the conventional AZT.

Conclusion

Results of the present study demonstrated that SLMs of zidovudine could successfully be employed for formulating sustained-release dosage forms of AZT. The investigated SLM showed that batch B1 formulated with 1 % AZT was capable of maintaining constant plasma AZT concentration through 15 h and the chance of dose-dumping associated with conventional AZT tablet will be eliminated. This formulation of SLM can be expected to reduce the frequency of administration and decrease the dose-dependent side effects associated with repeated administration of conventional AZT tablets as a result of very short half due to first-pass metabolism and fast renal excretion and variable bio-distribution; the gradual release of the SLMs with concentrations within the therapeutic range of AZT as opposed to the initial burst release of the conventional release tablets would be advantageous in reducing the dose-dependent side effects associated with repeated administration of conventional AZT tablets. The biodistribution studies of the SLMs showed ability to distribute to the major organs of the body achieving concentrations that were slightly higher than that of conventional tablets, this could enhance efficacy of the drug.

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Declaration of interest

The authors report no declarations of interest as no financial assistance or grant was received by authors in the course of this work.

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