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Multiple Emulsion Mediated Delivery of Azilsartan Medoxomil for Improved Solubility and Bioavailability

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Abstract- This study focuses on the formulation and characterization of water-in-oil-in-water (W/O/W) multiple emulsions of Azilsartan Medoxomil to enhance its solubility, stability, and oral drug delivery performance. A total of nine formulations (AZL1–AZL9) were developed using Span 20, Span 40, and Span 80 at varying concentrations as primary emulsifiers, while Tween 80 was employed as the secondary emulsifier. The prepared multiple emulsions were evaluated for various physicochemical and morphological parameters including visual appearance, organoleptic properties, microscopic examination, globule size, polydispersity index (PDI), zeta potential, viscosity, pH, conductivity, drug content, entrapment efficiency, in vitro drug release, and stability studies. Among all formulations, AZL6 exhibited superior characteristics with a mean globule size of 2.6 µm, uniform droplet distribution, high entrapment efficiency of 98.2 ± 1.1%, optimum viscosity, and cumulative drug release of 94.3% over 12 hours. The formulation also demonstrated good colloidal stability with a zeta potential value of −29.5 mV. Drug release kinetic studies revealed that the optimized formulation followed the Higuchi diffusion model and Super Case II transport mechanism, indicating both diffusion- and erosion-controlled release behavior. Furthermore, post-formulation stability studies, including centrifugation stress testing, confirmed the physical stability of the emulsion system. The findings of this investigation suggest that multiple emulsions represent a promising and effective delivery approach for poorly water-soluble drugs such as Azilsartan Medoxomil, with potential to improve oral bioavailability and therapeutic efficacy.

Keywords- Multiple Emulsion, Azilsartan Medoxomil, Drug Release Kinetics, Entrapment Efficiency, Zeta Potential, Solubility Enhancement, Oral Drug Delivery.

I. INTRODUCTION

Hypertension is a major chronic cardiovascular disorder and one of the leading causes of global morbidity and mortality. Proper regulation of blood pressure is essential to reduce the risk of serious complications such as stroke, myocardial infarction, heart failure, and renal dysfunction. Among the various classes of antihypertensive agents, Azilsartan Medoxomil, an angiotensin II receptor blocker (ARB), has attracted significant attention due to its potent antihypertensive efficacy, longer duration of action, and favorable safety profile [1]. Despite these therapeutic advantages, the clinical effectiveness of Azilsartan Medoxomil is limited by its poor aqueous solubility and low oral bioavailability. Furthermore, as a prodrug, it undergoes enzymatic hydrolysis in the gastrointestinal tract to produce the active metabolite, which may lead to variable absorption and inconsistent systemic availability [2].

To overcome these limitations, the development of advanced drug delivery systems capable of enhancing solubility, stability, and sustained drug release has become an important area of pharmaceutical research. Among the novel drug delivery approaches, multiple emulsions have emerged as promising carrier systems because of their unique structural organization and versatile pharmaceutical applications. Multiple emulsions are complex dispersed systems generally categorized as water-in-oil-in-water (W/O/W) or oil-in-water-in-oil (O/W/O) emulsions, in which both hydrophilic and lipophilic drugs can be effectively encapsulated [3]. These systems offer several advantages such as improved drug stability, protection of the active pharmaceutical ingredient from degradation, controlled and prolonged drug release, enhanced bioavailability, and reduced dosing frequency.

In recent years, multiple emulsions have gained considerable importance in oral drug delivery due to their capability to improve the delivery of poorly water-soluble drugs. The

internal aqueous phase of W/O/W emulsions can act as a reservoir system, enabling sustained release of the drug while minimizing drug degradation in the gastrointestinal environment. Additionally, the use of suitable surfactants and stabilizers contributes to improved emulsion stability and enhanced therapeutic performance [4]. Multiple emulsions also provide opportunities for site-specific delivery and modulation of drug release kinetics, thereby improving patient compliance and therapeutic outcomes.

The present study focuses on the formulation and characterization of W/O/W multiple emulsions containing Azilsartan Medoxomil with the objective of enhancing its solubility, stability, and oral bioavailability. The emulsions were prepared using a two-step emulsification technique employing different concentrations of Span 20, Span 40, and Span 80 as primary emulsifiers and Tween 80 as a secondary emulsifier to achieve a stable multiple emulsion system. The prepared formulations were evaluated for various physicochemical and morphological parameters including globule size, polydispersity index, zeta potential, viscosity, pH, conductivity, entrapment efficiency, drug content, in vitro drug release, and stability studies [5].

The outcomes of this research are expected to provide valuable insights into the potential application of multiple emulsions as an efficient drug delivery platform for poorly water-soluble antihypertensive drugs such as Azilsartan Medoxomil. The successful development of such a system may contribute toward improved therapeutic efficacy, enhanced patient compliance, and better management of hypertension.

II. MATERIALS AND METHODS

Azilsartan Medoxomil was bought from Med Chem in Mumbai. Opifex research labs provided Span 80, Tween 80, liquid paraffin. The other compounds utilized were of analytical grade.

Formulation of Azilsartan Medoxomil multiple emulsion: [6]

Method of Preparation

Multiple emulsions were prepared by using two step emulsification method: a) Preparation of primary emulsification; b) Secondary emulsification.

Primary emulsification: A quantity of 10 mL of distilled water containing 25 mg of Azilsartan Medoxomil was added gradually to 14 mL of the oil phase containing 25 mg of drug along with the primary emulsifier (Span 20, Span 40, or Span 80). The mixture was continuously stirred at 5000 rpm for 5 minutes to obtain the primary emulsion.

Secondary emulsification: An external aqueous phase containing the secondary emulsifier (Tween 80) along with 50 mg of Azilsartan Medoxomil was gradually incorporated into 20 mL of the previously prepared viscous primary emulsion. The mixture was then subjected to continuous stirring at 1000 rpm for 10 minutes to form the final multiple emulsion system.

The same procedure was followed for all formulations. The influence of the primary emulsifier was evaluated by preparing and comparing multiple formulations with different emulsifier compositions.

Formulation of AZL

Table 1: Formulation Design of Azilsartan Medoxomil Multiple Emulsions

Formulation Code	Drug (mg)	Span Type	Span (mL)	Tween 80 (mL)	Liquid Paraffin (mL)	Internal Aqueous Phase (mL)	External Buffer pH 6.8 (mL)	Total Volume (mL)	W:O:W Ratio
AZL1	100	S-20	0.5	0.8	12	8	20	40.5	8:12:20
AZL2	100	S-20	1.0	1.0	13	9	18	42.0	9:13:18
AZL3	100	S-20	1.5	1.2	14	10	16	42.7	10:14:16
AZL4	100	S-40	0.5	0.8	13	8	19	41.3	8:13:19
AZL5	100	S-40	1.0	1.0	14	10	15	41.0	10:14:15
AZL6	100	S-40	1.5	1.2	15	11	14	42.7	11:15:14
AZL7	100	S-80	0.5	0.8	14	9	17	41.3	9:14:17
AZL8	100	S-80	1.0	1.0	15	10	15	42.0	10:15:15
AZL9	100	S-80	1.5	1.2	16	12	12	42.7	12:16:12

Evaluation Studies of Azilsartan Medoxomil Multiple Emulsions

1. Visual Inspection [7,8]

The prepared emulsions were stored in transparent containers and periodically examined under normal and polarized light to evaluate their physical appearance, phase behavior, and homogeneity. Any signs of instability such as creaming, sedimentation, or coalescence were carefully recorded throughout the study period.

2. Organoleptic Characteristics [9, 10]

Stability assessment was performed by observing changes in color, phase separation, and liquefaction of both primary and multiple emulsions at predetermined time intervals (0 h, 1 h, 1 day, 3 days, 7 days, 14 days, 21 days, and 28 days) under different storage conditions.

3. Microscopic Examination [11, 12]

Microscopic analysis was carried out at 100× magnification using oil immersion to evaluate globule size distribution and structural integrity. Observations were performed immediately after preparation and at regular intervals up to 28 days.

4. Globule Size Analysis [13,14]

Globule size determination was performed using a light microscope equipped with a digital imaging system. Measurements were taken for freshly prepared emulsions and subsequently at predefined intervals over a 28-day storage period under different conditions.

5. Viscosity Determination

Viscosity of the formulations was measured using a Brookfield Viscometer (Model DV-II) at 50 and 100 rpm. All measurements were conducted in triplicate for both freshly prepared and stored emulsions to ensure reproducibility.

6. Entrapment Efficiency (%EE) [15,16]

Entrapment efficiency was determined by centrifuging freshly prepared W/O/W emulsions at 4000 rpm for 10 minutes. The separated aqueous phase was collected, filtered through a 0.22 µm membrane, suitably diluted with phosphate buffer (pH 6.8), and analyzed spectrophotometrically at 267.6 nm.

7. pH Measurement [17,18]

The pH of the emulsions was measured using a calibrated digital pH meter at regular time intervals (1, 3, 7, 14, 21, and 28 days) to assess chemical stability under various storage conditions.

8. Conductivity Test

Electrical conductivity of the emulsions was measured to determine the nature of the continuous phase. Higher conductivity values indicated a water-continuous phase, whereas lower values suggested an oil-continuous phase.

9. Zeta Potential Measurement

The zeta potential was measured to determine colloidal stability. A value over ±30 mV indicated an uninterrupted formulation due to electrostatic repulsion among particles.

10. In Vitro Drug Release Study

Drug release was investigated using a static diffusion cell apparatus with a 200 µm cellophane membrane. The donor compartment contained 15 mL of emulsion, while the receptor compartment was filled with 100 mL of phosphate buffer (pH 6.8) maintained at 37 ± 0.5 °C. Samples were withdrawn at predetermined time intervals and analyzed spectrophotometrically at 267.6 nm.

11. Drug Release Kinetics

To better understand the process of drug release, the release data were fitted to several kinetic models, including zero order, first order, higuchi, and peppas.

12. Ex Vivo Drug Release Study [11]

The ex vivo drug permeation study was conducted using the perfusion technique on isolated goat ileum. A specially designed U-shaped glass apparatus with an internal diameter of 1 cm was used, and a cannulated incision was made at the upper end of one arm to facilitate the experimental procedure.

13. Centrifugation Test

Physical stability was determined by centrifuging 10 g samples at 5000 rpm and 25°C. Tests were performed immediately upon preparation and repeated at 24 hours, 3, 7, 14, and 28 days.

III. RESULTS AND DISCUSSION

Compatibility studies by Fourier Transform Infrared (FTIR) spectroscopy:

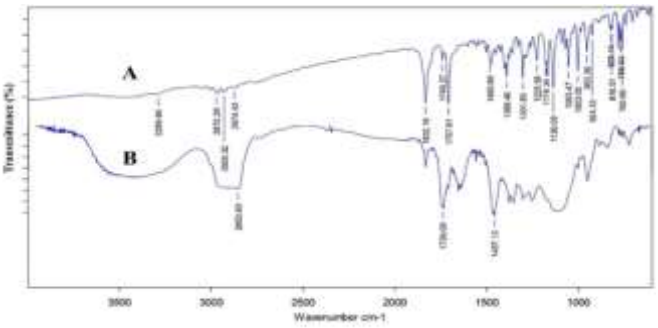


Figure No. 1: FTIR Spectrum of A) Azilsartan Medoxomil drug B) Azilsartan Medoxomil Multiple emulsion

FTIR analysis confirmed the chemical integrity of Azilsartan Medoxomil within the multiple emulsion formulation. No significant drug–excipient interactions were observed, as the

characteristic absorption peaks corresponding to the ester carbonyl group (C=O ~1730 cm^{-1}), aromatic functional groups (C=N/C=C ~1650 cm^{-1}), and O–H/N–H stretching (~3320 cm^{-1}) were retained in the formulation spectra.

A slight broadening of the O–H/N–H stretching region was noted, which may be attributed to mild hydrogen bonding interactions with the surfactant components. However, the overall spectral profile remained unchanged, indicating that the drug maintained its structural integrity. These findings confirm the compatibility and stability of Azilsartan Medoxomil with the excipients used in the multiple emulsion system.

Characterization of prepared Azilsartan Medoxomil multiple emulsions:

Visual Inspection:

Table No. 2: Visual Inspection of Azilsartan Medoxomil Multiple Emulsions Over 28 Days

Day	**Appearance (Normal Light)**	**Appearance (Polarized Light)**	**Creaming**	**Sedimentation**	**Coalescence**
0	Uniform, milky, homogenous	No birefringence	None	None	None
3	Uniform, stable	No birefringence	None	None	None
7	Slight creaming at top	No birefringence	Slight	None	None
14	Minor creaming persists	No birefringence	Slight	Minimal	None
21	Creaming stable	No birefringence	Slight	Slight	None
28	Uniform after shaking	No birefringence	Slight	Slight	None

Azilsartan Medoxomil multiple emulsions maintained good physical stability during 28 days, with a uniform appearance and limited, reversible creaming. No coalescence or drug crystallization was detected, indicating the formulation's integrity and appropriateness for drug administration.

Organoleptic Characteristics (Colour, Liquefaction, Phase Separation):

Table No. 3: Organoleptic properties of multiple emulsions

Time	Liquefaction			Colour			Phase separation			Centrifugation		
	8°C	25°C	40°C	8°C	25°C	40°C	8°C	25°C	40°C	8°C	25°C	40°C
0hr	-	-	-	mw	mw	mw	-	-	-	-	-	-
1hr	-	-	-	mw	mw	mw	-	-	-	-	-	-
24Hrs	-	-	-	mw	mw	mw	-	-	-	-	-	-
72Hrs	-	-	-	mw	mw	mw	-	-	-	-	-	-
7days	-	-	-	mw	mw	mw	-	-	-	-	-	-
14days	-	-	-	mw	mw	mw	-	+	-	-	+	+
21days	-	-	-	mw	mw	mw	-	+	+	+	+	+
28days	-	+	+	mw	mw	mw	-	+	+	+	+	+

No change (-); slight change (+); no more change (++) cw =creamy white; An oven was used to maintain the specified temperature conditions (n=3).

During the 28-day storage period, the primary emulsions retained a stable milky white appearance with no signs of liquefaction or phase separation under normal storage conditions. However, by Day 28, samples stored at 40°C exhibited liquefaction along with moderate phase separation, suggesting a degree of instability under elevated temperature conditions.

IV. MICROSCOPIC EXAMINATION

Table No. 4: Microscopic Observations of AZL1–AZL9

Formulation	Droplet Shape	Surface Appearance	Remarks
AZL1	Spherical, uniform	Smooth	Stable; well-formed emulsion

AZL2	Mostly spherical	Slightly coarse	Acceptable; minor irregularities
AZL3	Irregular	Uneven	Early signs of instability
AZL4	Spherical, uniform	Smooth	Good stability; consistent structure
AZL5	Slightly deformed	Slightly uneven	Needs optimization; minor instability
AZL6	Spherical, well-distributed	Smooth	Stable; homogenous emulsion
AZL7	Large, irregular	Coarse	Poor stability; likely coalescence
AZL8	Mostly spherical	Smooth	Acceptable; moderate size variation
AZL9	Mixed (spherical & irregular)	Uneven	Heterogeneous; signs of mild instability

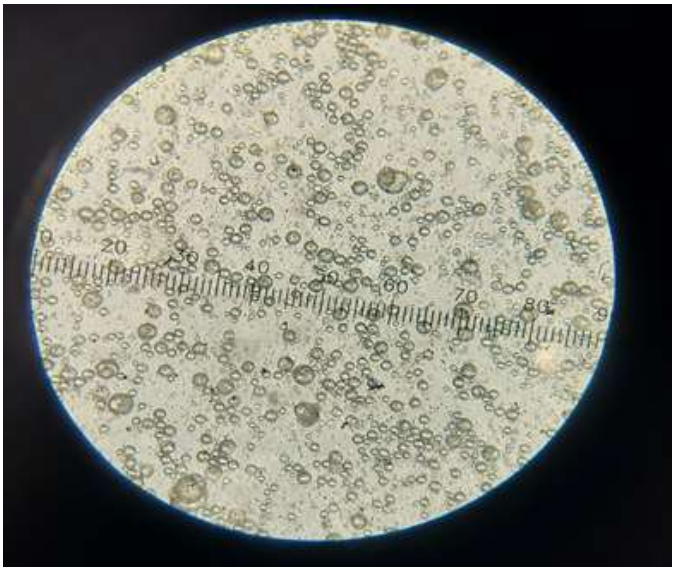


Figure No. 2: Microscopic view of Azilsartan Medoxomil multiple emulsions

Microscopic examination indicated that AZL1, AZL4, and AZL6 are stable formulations containing homogeneous, sphere droplets. AZL2, AZL5, and AZL8 exhibited considerable stability, whereas AZL3, AZL7, and AZL9 showed evidence of instability. AZL1, AZL4, and AZL6 were selected for further investigation.

Table No. 5: Evaluation studies of Azilsartan Medoxomil multiple emulsions

Formulation	**Avg. Globule size (µm)**	**Viscosity at 50 rpm (cP)**	**Viscosity at 100 rpm (cP)**	**EE (%)**	**pH (mean±SD)**	**Drug Content (% w/w)**	**Conductivity (µS/cm)**
AZL1	8.4	312 ± 5.0	262 ± 4.5	79.1 ± 1.7	6.84 ± 0.04	92.6 ± 1.1	42.4 ± 1.1
AZL2	2.7	282 ± 4.6	237 ± 4.1	85.8 ± 1.4	6.79 ± 0.05	93.8 ± 1.0	46.8 ± 1.3
AZL3	3.5	332 ± 5.9	272 ± 5.3	82.5 ± 1.5	6.73 ± 0.05	91.0 ± 1.4	39.2 ± 1.4
AZL4	7.3	297 ± 4.3	247 ± 3.8	76.9 ± 2.0	6.88 ± 0.04	91.9 ± 1.2	43.9 ± 1.2
AZL5	14.2	398 ± 7.4	348 ± 6.0	69.8 ± 2.3	6.61 ± 0.05	90.1 ± 1.5	33.1 ± 1.6
AZL6	2.6	262 ± 3.8	212 ± 3.4	98.2 ± 1.1	6.93 ± 0.03	95.1 ± 0.9	49.0 ± 1.0
AZL7	5.0	352 ± 6.3	297 ± 5.5	81.0 ± 1.8	6.68 ± 0.05	90.4 ± 1.3	37.0 ± 1.3
AZL8	6.5	302 ± 5.0	257 ± 4.4	74.8 ± 2.1	6.83 ± 0.04	92.3 ± 1.2	40.5 ± 1.2
AZL9	1.3	242 ± 3.5	202 ± 3.1	87.4 ± 1.3	6.91 ± 0.04	94.5 ± 1.0	47.6 ± 1.1

The physicochemical analysis of Azilsartan Medoxomil multiple emulsions revealed AZL 6 to be the most promising formulation, as proven by its superior structural and functional features. Microscopically, AZL 6, AZL 2, and AZL 9 exhibited homogenous, spherical droplets with low surface defects, resulting in decreased globule sizes (2.6, 2.7, and 1.3 µm) and increased physical stability. The use of Span 40 and Tween 80 in AZL 6 resulted in an ideal hydrophilic-lipophilic balance (HLB), forming a stable interfacial film that reduced coalescence and improved emulsion stability, which is consistent with previous research on tailored emulsifier mixtures in W/O/W systems.

Viscosity profiles showed formulation consistency, with AZL 6, AZL 9, and AZL 2 having reduced viscosities, indicating smooth shape and efficient flow behaviour. The increased viscosity seen in AZL- 5 is owing to the excess Span content, which causes coarse droplets and increased internal friction, supporting previous findings that emphasize the role of emulsifier concentration on rheological behaviour.

Entrapment efficiency (EE) was highest in AZL 6 (98.2%), demonstrating the importance of robust interfacial coatings in minimizing drug leakage. The positive relationship between smaller droplet size and improved EE is consistent with previous research showing droplet integrity in medication retention. Similarly, AZL 6 maintained the highest drug content (95.1%), indicating better stability and less leakage.
The pH values across all formulations remained within the approved oral administration range (6.6-7.0), with AZL 6 and AZL 9 having the most favourable profiles. Variations in pH for other formulations might be caused by surfactant hydrolysis or instability, as seen in earlier pH-sensitive emulsion systems.

The conductivity investigation confirmed aqueous phase continuity and droplet dispersion efficiency. AZL-6 showed the highest conductivity (49.0 µS/cm), indicating a well-distributed internal structure and successful emulsifier coupling. These results are congruent with those of Solans et al. (2003), who found higher ionic mobility in well-structured multiple emulsions.

Overall, our findings show that AZL-6 is the most stable and effective formulation for future development, with structural, rheological, and functional advantages based on established emulsion science ideas.

Zeta potential:

Table No. 6: Zeta Potential values of best formulation

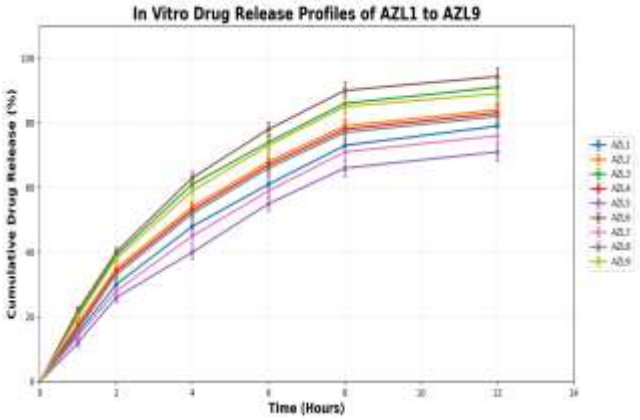
Formulation	Zeta Potential (mV)	Remarks
AZL 6	-29.5	High stability; sufficient repulsive forces between droplets

Figure No. 3: Zeta potential of formulation 6 (AZL 6)

AZL 6's zeta potential of -29.5 mV indicates strong electrostatic repulsion and great colloidal stability. Values over ±25 mV are often considered stable (Honary & Zahir, 2013). This verifies the formulation's resistance to coalescence and corresponds with its high entrapment efficiency and droplet shape, which have previously been seen in other stable systems (Shakeel et al., 2021).

In Vitro drug release studies of Azilsartan Medoxomil:

The graph below displays the cumulative percentage of drug release (mean ± SD) over time for different emulsion formulations (AZL 1-9). Every value is obtained from 3 measurements. AZL 6 showed the highest cumulative drug release (94.3% at 12 hours), owing to its excellent emulsifier composition, small globule size, and stable interfacial coating. These qualities increased surface area and facilitated drug diffusion. In contrast, AZL 5 and AZL 7 emitted fewer droplets, most likely due to their larger size and instability. Similar studies by Shakeel et al. (2020) and Pavoni et al. (2020) show that small droplet sizes and strong emulsifier systems significantly enhance release kinetics in a variety of emulsions.



In Vitro drug release Kinetics:

AZL- 6 released drugs using the Higuchi model (R^2 = 0.9736), demonstrating diffusion-controlled kinetics. However, the Korsmeyer-Peppas model (n = 2.2489) proposed a Super Case-II transport mechanism, which included polymer relaxation and erosion. Poor fits with zero- and first-order models revealed a non-constant, concentration-independent release. These findings lend credence to the complicated, regulated release behavior observed in different emulsion systems.

Table No. 7: Release kinetics and mechanisms of AZL 6

Formulation code	Zero order		First order		Higuchi		Korsmeyer -Peppas		Possible drug release mechanism
	Slope	R^2	Slope	R^2	Slope	R^2	R^2	N	
AZL 6	10.069	0.7187	0.1399	-6.688	29.565	0.9736	-9.385	2.2489	Super Case-II transport

Ex vivo drug release study

Ex vivo release investigations were conducted utilizing the U-shaped equipment. The goat ileum was hooked into this contraption, which was filled with several emulsions. Multiple emulsions were filled in the ileum and put in a dissolving tank containing pH 6.8 phosphate buffer. The substance discharged into the media was measured. Table 7 shows the cumulative medication release across various time intervals.

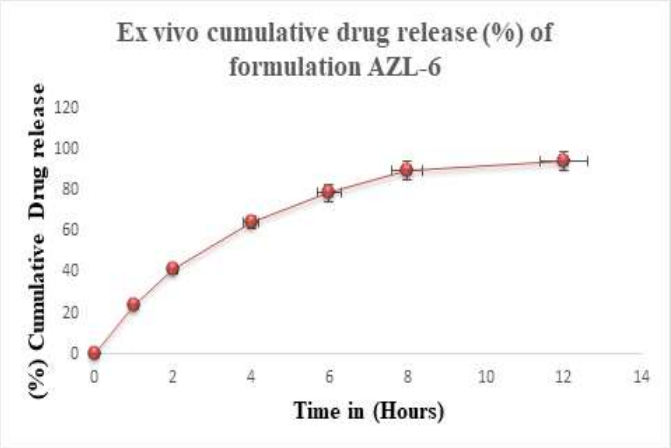


Figure No.4. Ex vivo cumulative, drug release (%) of formulation AZL 6 through goat ileum over 12 hours

Stability studies - Centrifugation tests:

Table No. 8: Centrifugation Stability Study Results (5000 rpm, 25°C, 10g sample)

Formulation	Time Point	Observation After Centrifugation	Stability Status
Primary Emulsion (PE)	0 h (immediate)	Slight creaming observed	Moderate
	24 hrs	Creaming intensified	Poor
	3 days	Phase separation started	Unstable
	7 days	Complete phase separation	Failed
	14 days	Complete separation	Failed
	28 days	Not tested (already destabilized)	--
Multiple Emulsion (AZL 6)	0 h (immediate)	No separation or creaming	Excellent

	24 hrs	No visible change	Stable
	3 days	Slight creaming (negligible)	Acceptable
	7 days	No coalescence or phase separation	Stable
	14 days	Consistent stability	Stable
	28 days	No separation, very fine droplets	Highly stable

Centrifugation stability studies revealed low kinetic stability of the primary emulsion (PE), with phase separation occurring by day 7. In contrast, AZL 6 maintained remarkable physical stability for 28 days, demonstrating just mild creaming by day 3 and no further advancement. This improved stability under stress circumstances demonstrates the W/O/W emulsion system's durability and is in line with its favorable zeta potential, droplet shape, and viscosity profile.

V. CONCLUSION

This study successfully created and assessed nine different emulsion formulations (AZL1-AZL-9) for oral administration of Azilsartan Medoxomil with the goal of improving solubility, bioavailability, and therapeutic effectiveness. AZL- 6 showed the most promising findings. It had the greatest entrapment effectiveness (98.2%), appropriate globule size (2.6 µm), and sustained in vitro drug release (94.3% after 12 hours). The pH (6.93 ± 0.03) showed acceptable gastric compatibility, whereas the zeta potential (-29.5 mV) demonstrated outstanding colloidal stability. The low viscosity and suitable conductivity (49.0 ± 1.0 µS/cm) made it easy to administer and maintain structural integrity.

Microscopic examination revealed spherical droplets without phase separation for up to 28 days, indicating strong physical stability. Drug release kinetics matched the Higuchi model, and Korsmeyer-Peppas analysis (n = 2.2489) revealed a Super Case-II transport mechanism that included both diffusion and polymer relaxation. Centrifugation stability proved the formulation's mechanical resilience.

Ex vivo experiments on goat ileum revealed a cumulative drug release of 94.3 ± 1.5% at 12 hours, consistent with in vitro results and demonstrating the formulation's sustained release in biological settings.

Overall, AZL- 6 provides a stable and efficient oral multiple emulsion solution for Azilsartan Medoxomil, with a high potential for improving therapeutic results and patient compliance.

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