



Formulation and Evaluation of Combination of Artemether and Lumefantrine as Tablets

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Article info

Received: 09/07/2021

Revised: 19/08/2021

Accepted: 26/09/2021

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Abstract

The objective of the present study was to fabricate and evaluate a combination of Artemether and Lumefantrine as tablets and to make Artemether in sustained form as to prolong its elimination time. Artemether was formulated in form of microspheres and was then formed into the tablet along with the Lumefantrine. Artemether microspheres were prepared and compressed into compressible tablet by direct compression process using the compressible excipients along with Lumefantrine, which entails the convenience of a sustained release product in one. The rationale behind this combination is that Artemether initially provides rapid symptomatic relief by reducing the number of parasites present before Lumefantrine eliminates any residual parasites. This is thought to minimize development of resistance because the malaria parasites are never exposed to Artemether alone due to its rapid elimination.

Although they may be exposed to Lumefantrine alone, the probability of resistance developing simultaneously to both drugs used in combination is thought to be low. The Artemether microspheres were formed by solvent evaporation technique using ethyl cellulose as a polymer, in presence of polyvinyl alcohol as surfactant. Due to the sustained property of polymer and surfactant property of polyvinyl alcohol, formulated microspheres can result in controlled release of drug. Ethyl cellulose coated microparticles have also demonstrated their capability to absorb pressure and therefore save the coating from fracture during tablet manufacturing process.

Keywords: Artemether, Lumefantrine, Formulation, Evaluation

Introduction

Malaria is one of the most important and scourge infectious diseases in developing areas of the world. Worldwide, over 40% of the population lives in areas where malaria transmission occurs i.e., parts of Asia, Africa, Middle East, South and Central America. It is estimated that 300-500 million cases of malaria occur each year resulting in 750,000- 2 million deaths (World malaria situation,1994). Microspheres are described as small particles ranging from 1-1000 micrometer in size for use as carriers of drugs and other therapeutic agents which consists of proteins or synthetic polymers that are biodegradable in

nature. The term microsphere describes a monolithic spherical structure in which the drug or therapeutic agent is distributed throughout the matrix either as a molecular dispersion or as a dispersion of particles (Nikam *et.al* 2012) They can also be defined as a structure consisting of continuous phase of one or more miscible polymers in which the particulate drug is dispersed at the macroscopic or molecular level. (Prasanth *et.al* 2011).

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Material and Methods

Artemether microspheres production procedure

Artemether microspheres were obtained by solvent evaporation technique. Firstly, polymer (ethylcellulose) was dissolved in dichloromethane and then the drug (Artemether) was added to the above solution. The polymer drug solution so obtained was injected into the PVA solution maintained at variable speed using mechanical stirrer. Stirring was continued for required period until all the dichloromethane evaporated. The formed microspheres were collected by filtration and washed with n-Hexane and dried to obtain free flowing microspheres.



Figure: Artemether microspheres

Table: Formulation codes of Artemether-EC Microspheres

Formulation Codes	Drug (Artemether) (mg)	Polymer (Ethyl Cellulose) (mg)	Solvent (DCM) (ml)	Medium (PVA) (%)	Stirring rate (rpm)
M1	500	1000	20	0.5	200
M2	500	1000	20	0.5	400
M3	500	1000	20	0.5	600 ¹²

M4	500	1250	20	0.5	200
M5	500	1250	20	0.5	400
M6	500	1250	20	0.5	600
M7	500	1000	20	0.3	200
M8	500	1000	20	0.3	400
M9	500	1000	20	0.3	600
M10	500	1250	20	0.3	200
M11	500	1250	20	0.3	400
M12	500	1250	20	0.3	600
M13	500	1000	20	0.1	200
M14	500	1000	20	0.1	400
M15	500	1000	20	0.1	600
M16	500	1250	20	0.1	200
M17	500	1250	20	0.1	400
M18	500	1250	20	0.1	600

Characterization Of Microspheres

Particle size analysis

All formulation showed a small mean size. The mean particle size for the drug loaded microspheres varies from 14.80 μm to 26.42 μm . The particle size distribution of microspheres is represented as follows.

Table: Particle size of microspheres

Formulation Codes	Particle Size (μm)	Formulation Codes	Particle Size (μm)
M1	18.45	M10	22.77
M2	18.30	M11	21.87
M3	15.00	M12	19.35
M4	23.12	M13	26.42
M5	19.10	M14	23.80
M6	16.92	M15	20.00
M7	21.00	M16	26.10
M8	21.00	M17	22.45

M9	14.80	M18	18.95
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Table: Size distribution of microspheres for different formulations

FC	0-5	6-10	11-15	16-20	21-25	26-30	31-40	41-50	51-60
M1	0	16	26	22	13	16	7	0	0
M2	0	12	33	14	22	16	3	0	0
M3	2	22	38	18	12	8	0	0	0
M4	0	8	26	19	10	14	12	9	2
M5	0	7	30	21	22	16	4	0	0
M6	0	18	36	15	13	17	1	0	0
M7	0	9	21	21	18	22	5	3	1
M8	0	11	20	15	25	16	12	1	0
M9	6	22	31	24	8	7	1	0	0
M10	0	6	29	15	17	16	6	8	3
M11	0	5	22	24	18	18	8	5	0
M12	0	19	26	17	7	15	16	0	0
M13	0	3	15	11	21	27	10	8	5
M14	0	5	28	7	24	10	18	6	2
M15	0	11	30	23	13	11	4	8	0
M16	0	1	18	24	14	15	14	9	5
M17	0	11	20	23	13	13	10	9	1
M18	0	22	32	14	9	11	3	7	2

Determination of Percentage yield of microspheres

The percentage yields for all formulations were determined. The values varied from 66.63% to 94%.

Table: Percentage yield (%) of microspheres

Formulation Codes	% Yield	Formulation Codes	% Yield
M1	84.87	M10	86.68
M2	79.40	M11	86.91
M3	94.00	M12	66.63
M4	90.28	M13	86.00
M5	86.00	M14	84.06
M6	70.34	M15	76.67
M7	88.33	M16	71.88
M8	92.27	M17	88.91
M9	91.73	M18	90.11

Determination of flow properties of microspheres

The prepared microspheres were evaluated for flow properties including bulk density, tapped density, Carr's index, Hausner ratio and angle of repose. Bulk density of all the batches was in the range of 0.63 – 0.68 gm/cm³. Tapped density in the range of 0.71 – 0.78 gm/cm³. Carr's index in range of 10.95 – 14.86 and Hausner ratio varies from 1.07 – 1.17 indicating excellent flow properties. Angle of repose was also found in the prescribed range showing excellent flow characteristics.

Table: Flow properties of Artemether microspheres

Formulation Codes	Bulk Density (gm/cm ³)	Tapped Density (gm/cm ³)	Carr's Index	Hausner Ratio	Angle of Repose (θ)
M1	0.66	0.76	13.15	1.15	20.21
M2	0.67	0.78	14.10	1.16	22.47
M3	0.66	0.77	14.28	1.16	19.63
M4	0.64	0.73	12.32	1.14	21.06
M5	0.65	0.73	10.95	1.07	20.13
M6	0.67	0.76	11.84	1.13	16.69
M7	0.68	0.79	13.92	1.13	20.51
M8	0.66	0.75	12.00	1.15	21.13
M9	0.66	0.76	13.15	1.15	18.30
M10	0.66	0.76	13.15	1.15	17.26

M11	0.65	0.75	13.33	1.15	18.14
M12	0.67	0.77	12.98	1.14	16.49
M13	0.64	0.75	14.66	1.17	18.33
M14	0.64	0.74	13.51	1.16	20.54
M15	0.67	0.76	11.84	1.13	17.05
M16	0.63	0.72	12.50	1.14	21.89
M17	0.61	0.71	14.08	1.16	19.94
M18	0.63	0.74	14.86	1.17	21.40

Drug Entrapment efficiency

To calculate the entrapment efficiency, accurately weighed quantity of microspheres (50 mg) were taken along with 50 ml of phosphate buffer pH 7.4 in a volumetric flask and kept for 24 hours. It was then filtered, suitably diluted and then analyzed by UV spectrophotometry at 216 nm. The EE was calculated and % EE varied from 52.81% to 74.42%. M9 formulation shows least percentage entrapment and M16 shows the highest.

Table: Entrapment efficiency (%) of microspheres

Formulation Codes	% Entrapment	Formulation Codes	% Entrapment
M1	66.20	M10	73.31
M2	60.49	M11	69.98
M3	54.06	M12	66.98
M4	70.57	M13	70.15
M5	67.42	M14	68.39
M6	63.23	M15	63.86
M7	67.81	M16	74.42
M8	61.15	M17	71.26
M9	52.81	M18	67.07

In vitro release studies of microspheres

In-vitro release of Artemether microspheres was carried out using the USP dissolution test apparatus at 37±0.5°C in 900 ml of phosphate buffer pH 7.4. Microspheres equivalent to 20 mg Artemether was placed in the muslin cloth and rotated at 100 rpm. A sample of 5 ml was withdrawn ¹⁶

at various time intervals and replaced with equal amount of medium to maintain the sink condition. The withdrawn samples were analyzed by UV spectrophotometer at 216 nm using phosphate buffer 7.4 as blank solution.

Table (a): Cumulative drug release (%) of M1 - M5

S.No.	Time (hrs)	% CDR				
		M1	M2	M3	M4	M5
1.	0	0	0	0	0	0
2.	0.5	3.12	3.76	3.12	3.64	7.45
3.	1	5.76	5.10	7.08	5.18	8.26
4.	2	10.44	13.05	10.44	10.85	13.02
5.	3	16.47	13.86	17.82	17.64	20.86
6.	4	22.65	21.93	24.63	21.63	27.26
7.	5	28.83	31.41	30.87	27.13	36.02
8.	6	34.89	40.35	34.47	34.37	44.10
9.	7	44.49	45.18	45.42	40.88	48.97
10.	8	51.78	56.85	56.28	48.23	54.11
11.	9	57.81	62.47	65.98	55.72	58.34
12.	10	63.03	68.38	71.65	57.85	61.95

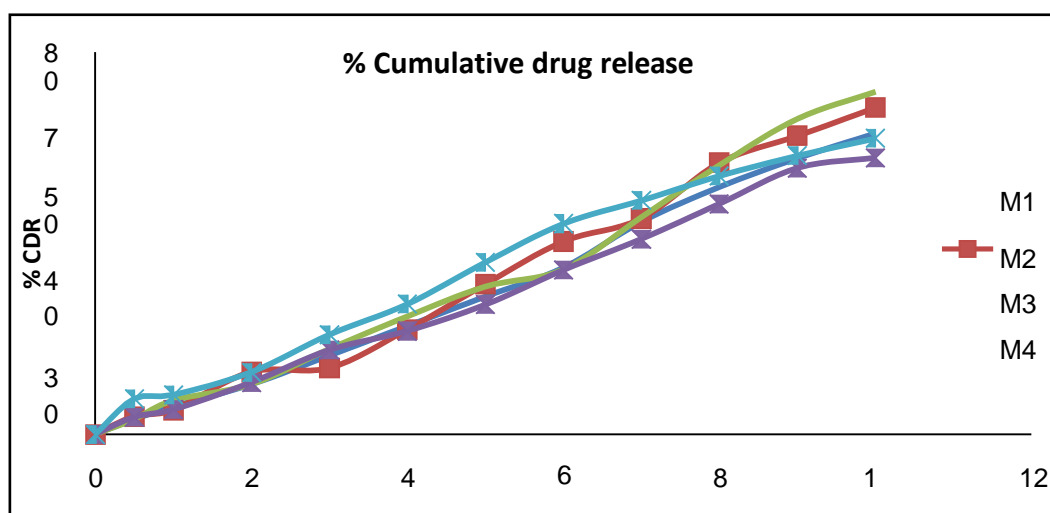


Figure (a): Cumulative drug release (%) of M1 - M5

% Cumulative drug release of formulation (M1-M5) varied from 57.85%-71.65%. M4 shows the minimum release after 10 hrs and M3 shows the maximum which may be due to the change in concentration of polymer or PVA or it could be due to the presence of other variables.

Table (b): Cumulative drug release (%) of M6 - M10

S.No.	Time (hrs)	% CDR				
		M6	M7	M8	M9	M10
1.	0	0	0	0	0	0
2.	0.5	9.03	4.41	5.07	2.44	5.91
3.	1	9.83	5.76	5.76	7.71	8.26
4.	2	17.67	9.12	9.78	15.06	14.56
5.	3	24.78	15.18	16.47	19.17	16.24
6.	4	31.99	21.27	21.93	27.99	23.28
7.	5	37.69	29.28	28.68	34.20	29.72
8.	6	43.99	36.87	36.96	40.32	36.96
9.	7	50.68	44.52	45.27	48.69	42.53
10.	8	56.32	50.97	54.99	58.17	48.41
11.	9	64.65	60.09	64.18	68.71	57.44
12.	10	69.19	69.01	71.79	76.72	63.38

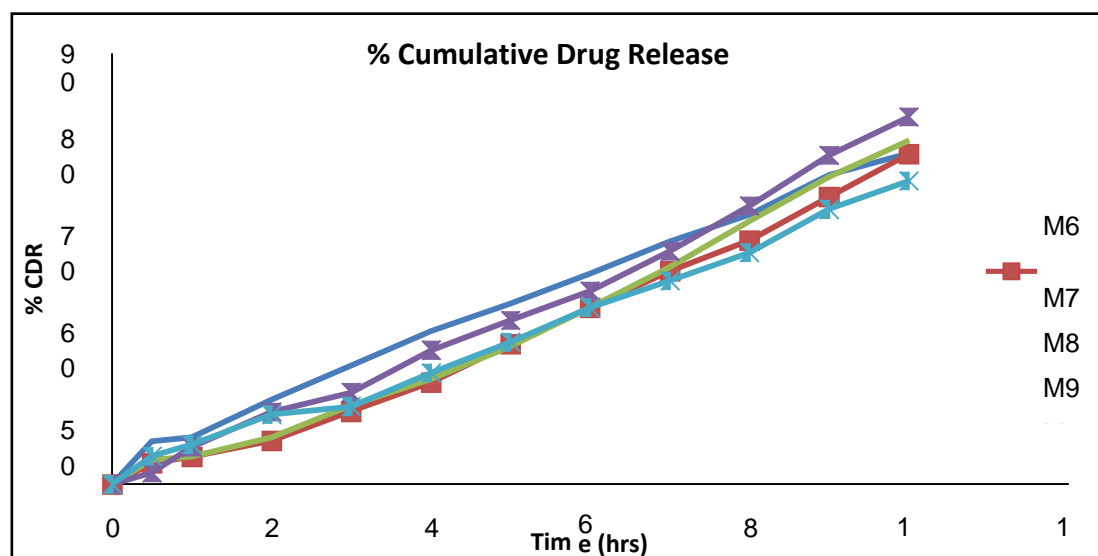


Figure (b): Cumulative drug release (%) of M6 - M10

% Cumulative drug release of formulation (M6-M10) varied from 63.38%-76.72%. M10 shows the minimum release after 10 hrs and M9 shows the maximum which may be due to the change in concentration of polymer or PVA or it could be due to the presence of other variables.

Table (c): Cumulative drug release (%) of M11 - M14

S.No.	Time (hrs)	% CDR			
		M11	M12	M13	M14
1.	0	0	0	0	0
2.	0.5	6.72	8.99	3.77	5.76
3.	1	15.96	17.60	7.08	7.11
4.	2	20.83	23.84	13.74	16.41
5.	3	24.92	29.47	18.54	24.51
6.	4	32.86	34.44	24.69	33.51
7.	5	40.92	40.04	34.86	42.99
8.	6	47.53	49.81	42.48	54.66
9.	7	53.27	60.41	49.56	61.24
10.	8	60.65	64.54	54.45	67.18
11.	9	65.87	68.66	66.92	71.47
12.	10	70.14	74.48	72.51	76.45

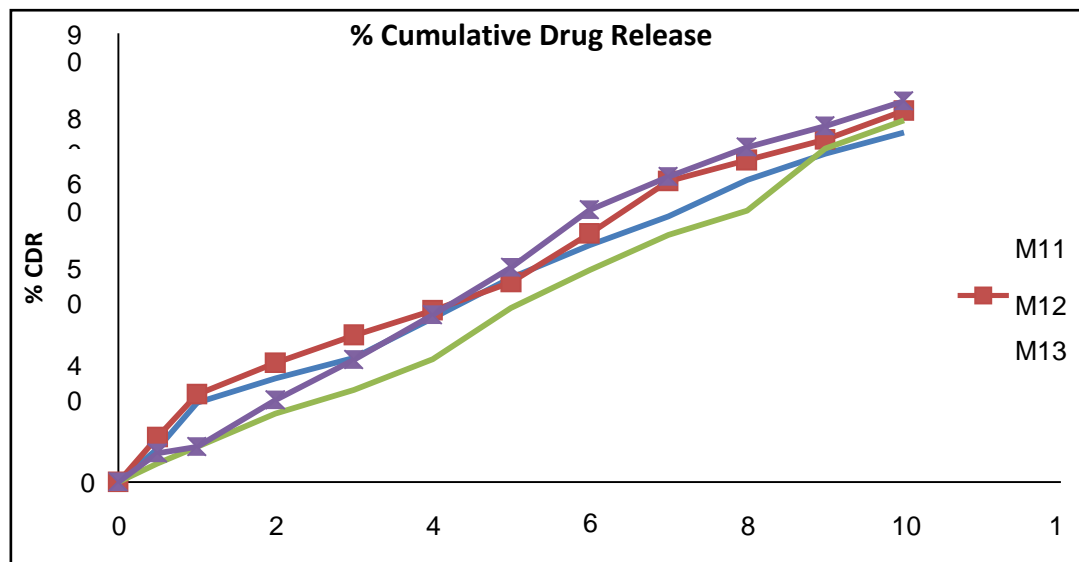


Figure (c): Cumulative drug release (%) of M11 - M14

% Cumulative drug release of formulation (M11-M14) varied from 70.14%-76.45%. M11 shows the minimum release after 10 hrs and M14 shows the maximum which may be due to the change in concentration of polymer or PVA or it could be due to the presence of other variables.

Table (d): Cumulative drug release (%) of M15 - M18

S.No.	Time (hrs)	% CDR			
		M15	M16	M17	M18
1.	0	0	0	0	0
2.	0.5	7.08	10.57	12.14	16.73
3.	1	9.75	12.91	12.95	19.14
4.	2	19.77	20.02	23.91	29.36
5.	3	29.22	26.39	32.62	35.87
6.	4	42.09	34.37	39.86	45.50
7.	5	48.51	43.23	46.48	49.63
8.	6	58.93	48.05	50.86	56.28
9.	7	64.84	55.51	56.49	61.74
10.	8	71.97	64.58	66.33	66.47
11.	9	79.15	69.89	72.35	70.28
12.	10	84.87	71.02	75.11	79.48

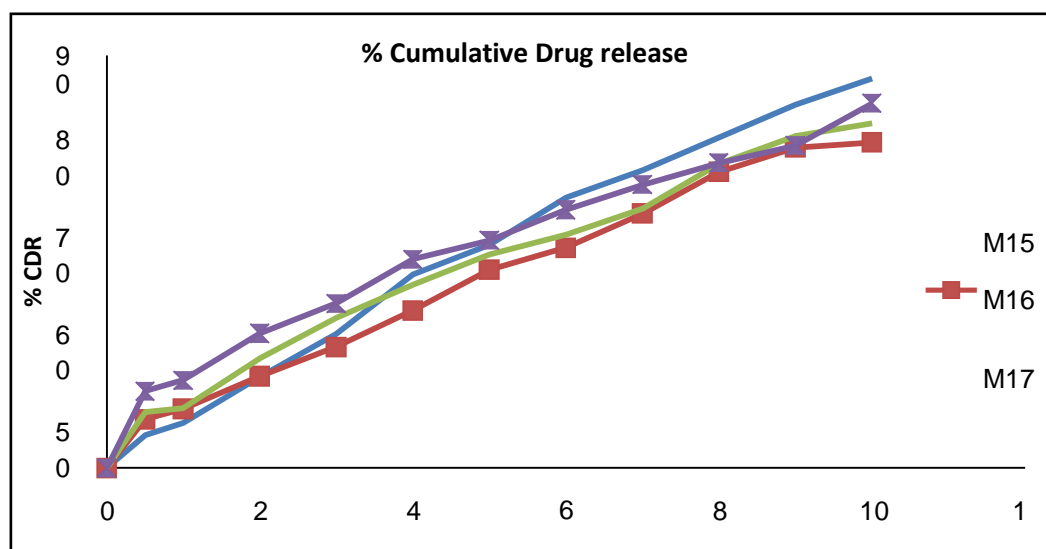


Figure (d): Cumulative drug release (%) of M15 - M18

% Cumulative drug release of formulation (M15-M18) varied from 57.85%-71.65%. M16 shows the minimum release after 10 hrs and M15 shows the maximum which may be due to the change in concentration of polymer or PVA or it could be due to the presence of other variables.

Effect of different formulation variables on various evaluation parameters

The influences of different formulation variables on various evaluation parameters were studied. The effects of polymer concentration (Ethylcellulose 1000-1250 mg), emulsifier concentrations (PVA concentration 0.1%-0.5%), and altered stirring speed of a mechanical stirrer (200, 400, 600 rpm) on microspheres characteristics (percentage yield, drug entrapment efficiency, particle size and cumulative drug release) were studied.

FC	Polymer (mg)	% Yield	% EE	Particle size (µm)	% CDR
M1	1000	84.87	66.20	18.45	63.03
M2	1000	79.40	60.49	18.30	68.38
M3	1000	94.00	54.06	15.00	71.65
M4	1250	90.28	70.57	23.12	57.85
M5	1250	86.00	67.42	19.10	61.95
M6	1250	70.34	63.23	16.92	69.19
M7	1000	88.33	67.81	21.00	69.01
M8	1000	92.27	61.15	21.00	71.79
M9	1000	91.73	52.81	14.80	76.72
M10	1250	86.68	73.31	22.77	63.38
M11	1250	86.91	69.98	21.87	70.14
M12	1250	66.63	66.98	19.35	74.48
M13	1000	86.00	70.15	26.42	72.51
M14	1000	84.06	68.39	23.80	76.45
M15	1000	76.67	63.86	20.00	84.87
M16	1250	71.88	74.42	26.10	71.02
M17	1250	88.91	71.26	22.45	75.11
M18	1250	90.11	67.07	18.95	79.48

The values for the % Yield for all formulations varied from 66.63% - 94% and is independent of the polymer concentration. The increase in the polymer concentration equals an approximately identical

increase in the entrapment efficiency. The particle size was dependent on the polymer concentration, as the increasing concentration increase the particle size. The particle size of microspheres prepared with 1000 mg of polymer ranged between 14.80 – 26.42 μm , with 1250 mg varies from 16.92 – 26.10 μm . It was observed that % cumulative drug release of formulations varied from 57.85%– 84.87%. The increase in the concentration of polymer results in the decrease in the % drug release. The increased polymer concentration might have led to increased density of the polymer matrix, resulting in an increased diffusional path length and consequent retardation of drug release. All the formulations prepared at 1000 mg concentration exhibited higher drug release than the formulation prepared at 1250mg.

Effect of stirring speed

Table : Effect of stirring speed on various parameters

FC	Stirring rate (rpm)	% Yield	% EE	Particle Size (μm)	% CR
M1	200	84.87	66.20	18.45	63.03
M2	400	79.40	60.49	18.30	68.38
M3	600	94.00	54.06	15.00	71.65
M4	200	90.28	70.57	23.12	57.85
M5	400	86.00	67.42	19.10	61.95
M6	600	70.34	63.23	16.92	69.19
M7	200	88.33	67.81	21.00	69.01
M8	400	92.27	61.15	21.00	71.79
M9	600	91.73	52.81	14.80	76.72
M10	200	86.68	73.31	22.77	63.38
M11	400	86.91	69.98	21.87	70.14
M12	600	66.63	66.98	19.35	74.48
M13	200	86.00	70.15	26.42	72.51
M14	400	84.06	68.39	23.80	76.45
M15	600	76.67	63.86	20.00	84.87
M16	200	71.88	74.42	26.10	71.02
M17	400	88.91	71.26	22.45	75.11
	600	90.11	67.07	18.95	79.48

M18					
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The prepared microspheres with the mean size ranging between 14.80 μm to 26.42 μm showed particle size dependence on the stirring speed. The results in the table confirmed that the microsphere mean size decreased with an increase in the stirring speed. The force of higher stirring distributes the internal phase into smaller droplets, resulting in the formation of smaller sized microspheres.

Formulation of Tablets containing Artemether microspheres and Lumefantrine

Tablets of Artemether (ART) microspheres and Lumefantrine (LUM) were prepared by direct compression technique. Microcrystalline cellulose (MCC) was used as a directly compressible diluent. Sodium starch glycolate (SSG) and Croscarmellose sodium (CCS) were used as superdisintegrants in a concentration of 1-5% of tablet weight. The corresponding amount of ART microspheres equivalent to 20 mg drug, lumefantrine, MCC and superdisintegrants were accurately weighed and blended. Thereafter the corresponding amount of magnesium stearate and colloidal silicon dioxide were added to the mixture. The mixture was allowed for direct compression into tablets weighing 300mg using a tablet punching machine with 8 mm flat faced punches.



Figure : Tablets containing artemether microspheres and lumefantrine

Table: Formulation codes of tablets containing ART Microspheres and LUM

FC	ART microsp here eq.to	LUM	MCC	SSG	CCS	Mag. Stearate	Colloidal Silicon Dioxide
MT1	20	120	107	3	-	7	3
MT2	20	120	105.5	4.5	-	7	3
MT3	20	120	104	6	-	7	3
MT4	20	120	102.5	7.5	-	7	3
MT5	20	120	101	9	-	7	3
MT6	20	120	99.5	10.5	-	7	3
MT7	20	120	98	12	-	7	3
MT8	20	120	96.5	13.5	-	7	3
MT9	20	120	95	15	-	7	3
MT10	20	120	97	-	3	7	3
MT11	20	120	95.5	-	4.5	7	3
MT12	20	120	94	-	6	7	3
MT13	20	120	92.5	-	7.5	7	3
MT14	20	120	91	-	9	7	3
MT15	20	120	89.5	-	10.5	7	3
MT16	20	120	88	-	12	7	3
MT17	20	120	86.5	-	13.5	7	3
MT18	20	120	85	-	15	7	3

*All quantities are in mg

Characterization of Tablets

Determination of thickness

3 tablets from each batch were taken randomly and their thickness was measured using Vernier calliper and the average value was calculated. It is expressed in millimetre. The thickness of each formulation was determined and was found to be relatively near about same. It was 4.83 mm to 4.88 mm.

Table : Thickness ofT ablets

Formulation Codes	Thickness (mm)	Formulation Codes	Thickness (mm)
MT1	4.88 ±0.01	MT10	4.87±0.01
MT2	4.85±0.01	MT11	4.87±0.02
MT3	4.83±0.02	MT12	4.88±0.03
MT4	4.88±0.03	MT13	4.86±0.01
MT5	4.86±0.02	MT14	4.85±0.03
MT6	4.86±0.01	MT15	4.88±0.01
MT7	4.88±0.04	MT16	4.87±0.02
MT8	4.87±0.01	MT17	4.84±0.01
MT9	4.83±0.02	MT18	4.88±0.01

Determination of diameter

3 tablets from each batch were taken randomly and their diameter was measured using Vernier calliper and the average value was calculated. It is expressed in millimetre. The diameter of each formulation was determined and was found to be relatively near about same. It was 9.05 mm to 9.08 mm.

Table 7.3.2: Diameter of Tablets

Formulation Codes	Diameter (mm)	Formulation Codes	Diameter(mm)
MT1	9.08±0.006	MT10	9.06±0.006
MT2	9.05±0.005	MT11	9.06±0.020
MT3	9.06±0.006	MT12	9.08±0.010
MT4	9.07±0.020	MT13	9.06±0.003
MT5	9.06±0.006	MT14	9.05±0.01
MT6	9.06±0.010	MT15	9.06±0.006
MT7	9.06±0.006	MT16	9.08±0.010
MT8	9.05±0.020	MT17	9.07±0.010
MT9	9.07±0.006	MT18	9.06±0.006

Determination of hardness

3 tablets from each batch were taken randomly and their hardness was measured using Monsanto hardness tester. It is expressed in Kg/cm². The hardness of each formulation was determined and was found to be relatively near about same. It was 4.33 to 6.00 Kg/cm².

Table 7.3.3: Hardness of Tablets

Formulation Codes	Hardness (mm)	Formulation Codes	Hardness (mm)
MT1	4.33±0.44	MT10	4.33±0.44
MT2	4.67±0.89	MT11	6.00±0.00
MT3	5.00±0.67	MT12	5.00±0.67
MT4	4.33±0.44	MT13	5.33±0.44
MT5	4.67±0.43	MT14	5.00±0.00
MT6	6.00±0.00	MT15	6.00±0.00
MT7	5.33±0.44	MT16	5.33±0.44
MT8	5.33±0.44	MT17	5.33±0.44
MT9	6.00±0.00	MT18	4.67±0.89

Determination of friability

The friability of the tablet was measured using Roche Friabilator. The friability of all the formulations was found to be between 0.75% to 0.92%, which was found to be within the pharmacopoeial requirement i.e. not more than 1% indicating good mechanical resistance of tablets sufficient.

Table: Friability of Tablets

Formulation Codes	Friability (mm)	Formulation Codes	Friability (mm)
MT1	0.75	MT10	0.85
MT2	0.85	MT11	0.78
MT3	0.87	MT12	0.88
MT4	0.79	MT13	0.89
MT5	0.80	MT14	0.85
MT6	0.88	MT15	0.86
MT7	0.85	MT16	0.86
MT8	0.86	MT17	0.81
MT9	0.92	MT18	0.85

Determination of Weight Variation

20 tablets were selected randomly from each formulation. These tablets were weighed and the average weight was calculated. The weight variation of each formulation was determined and was found to be relatively near about same. The average weights of tablets were found to be 297.45-303.24 mg. The acceptable weight range is $\pm 5\%$ as per IP for uniformity of weight thus indicating consistency in the preparation of the tablets and minimal batch to batch variation.

Table: Weight variation of tablets

Formulation Codes	Weight (mm)	Variation	Formulation Codes	Weight Variation (mm)
MT1	303.15	± 0.95	MT10	301.46
MT2	302.70	± 0.73	MT11	303.11
MT3	301.35	± 0.97	MT12	299.64
MT4	297.45	± 1.04	MT13	300.16
MT5	302.00	± 1.11	MT14	301.00
MT6	302.18	± 0.94	MT15	302.38
MT7	300.90	± 0.53	MT16	302.89
MT8	298.14	± 2.06	MT17	300.40
MT9	303.24	± 0.77	MT18	301.75

Determination of Drug Content

Five tablets were crushed and the powder equivalent to 20 mg of drug were accurately weighed and transferred to 10 ml volumetric flask. To this flask, small amount of 0.1 N HCl was added to dissolve it completely. Then, the volume of flask was made up to the mark with the same solvent. From this, 1ml of the aliquot was pipette out and transferred to 10 ml volumetric flask and the volume was made up to the mark with 0.1 N HCl. Solution was filtered, suitably diluted and drug content of lumefantrine was analyzed spectrophotometrically at 234 nm .

The drug content for all the formulations was determined. It was 96.32% - 99.43%

Table 7.3.6: Drug content of Tablets

Formulation Codes	Drug Content (%)	Formulation Codes	Drug Content (%)
MT1	98.91	MT10	99.24
MT2	99.16	MT11	95.49
MT3	98.67	MT12	97.89
MT4	99.31	MT13	98.99
MT5	98.13	MT14	98.64

MT6	96.32±0.14	MT15	99.43±0.24
MT7	98.62±0.23	MT16	99.33±0.21
MT8	94.38±0.46	MT17	96.54±0.17
MT9	97.94±0.68	MT18	98.77±0.19

The drug content for all the formulations was determined. It was 96.32% - 99.43%.

Microscopic evaluation of tableted microspheres

The morphology of the surface of tablets and broken diametrical surface of tablets was observed using a scanning electron microscopy (FEI, Model NOVANO 450). The tablets and the broken tablet surfaces were sputter coated with gold at a pressure of 5.13E to 4 pascal and 5 KV at 0° was maintained to get the photographs so as to observe the surface morphology of compressed microspheres.

The morphology of the prepared batches of tablet was evaluated by Scanning Electron Microscopy (SEM). Scanning Electron micrographs of the tablet surface and broken tablet surface are shown in figure at different magnifications of 200x, 24000x and 100000x for tablet and 50000x and 200000x respectively revealing the spherical and smooth surface. The microspheres appeared deformed but intact. This would explain the similar in vitro dissolution profiles for both the tablets and microspheres.

In vitro dissolution studies of Lumefantrine

Table : Cumulative drug release (%) of MT1 – MT18

S.No.	% CDR	Time (Min)						
		0	10	20	30	60	90	120
1.	M1	0	22.31	29.06	30.14	36.43	48.02	53.14
2.	M2	0	23.40	30.15	35.94	44.61	51.95	57.36
3.	M3	0	25.11	31.61	38.87	44.92	53.60	60.63
4.	M4	0	26.13	34.47	40.14	48.33	55.72	63.08
5.	M5	0	29.46	33.52	41.33	50.19	58.32	64.21
6.	M6	0	30.19	36.80	41.74	53.44	62.71	68.82
7.	M7	0	32.15	37.48	46.29	52.22	64.26	70.38
8.	M8	0	31.33	37.92	49.52	53.15	66.19	71.44
9.	M9	0	32.71	37.36	42.1	59.45	68.78	74.53

10	M10	0	33.6 4	40.70	45.42	51.17	62.04	69.63
11	M11	0	34.0 6	41.32	46.93	53.87	64.10	70.15
12	M12	0	35.2 6	42.81	49.32	57.02	66.15	72.49
13	M13	0	33.1 2	39.24	46.82	55.38	68.09	74.86
14	M14	0	34.0 3	47.32	52.26	60.15	67.12	75.32
15	M15	0	36.1 5	46.32	54.87	62.51	71.70	78.04
16	M16	0	36.6 0	44.12	49.33	51.08	63.95	78.11
17	M17	0	38.4 3	47.52	54.36	58.27	69.84	80.60
18	M18	0	39.3 6	50.21	59.08	66.52	74.39	81.14

The release rate of Lumefantrine from tablets for all the formulations. At 2nd hours release rate of drug was between 53.14% - 81.14%. M18 formulation shows the maximum release and M1 shows the minimum release. It showed that the release of the drug was dependent on the superdisintegrants used, in that Croscarmellose sodium can release drug faster compared to Sodium starch glycolate.

***In vitro* release of Lumefantrine from tablets**

In-vitro release of tablets containing microspheres was studied by using the USP dissolution test apparatus at 100 rpm using 900 ml 0.1 N HCl as dissolution medium. Temperature of the dissolution medium was maintained at 37±0.5°C. Aliquot of 5 ml of dissolution medium was withdrawn at predetermined time intervals. The volume of the of the dissolution medium was adjusted to 900 ml at every sampling time by replacing 5 ml with the same dissolution medium so as to maintain the sink conditions. The withdrawn samples were analyzed by UV spectrophotometer at 234 nm using 0.1 N HCl as blank solution and concentration of the drug was determined. (Kasid *et.al* 2013)

Table (a): Cumulative drug release (%) of MT1 - MT5

S.No.	% CDR	Time (Min)											
		0	0.5	1	2	3	4	5	6	7	8	9	10
1.	M1	0	3.71	6.73	14.28	26.54	34.32	42.19	46.28	52.84	59.36	61.88	66.14
2.	M2	0	4.17	8.11	12.24	24.51	33.96	44.81	49.28	56.12	61.47	64.15	69.81
3.	M3	0	3.10	7.18	15.33	24.11	34.67	48.31	52.43	58.55	60.19	68.30	73.12
4.	M4	0	2.96	6.84	13.15	20.27	27.19	32.83	39.22	46.32	50.34	55.34	61.22

						26		4	8	7	19		4
5.	M5	0	5.32	6.17	14.74	26.48	30.13	38.37	44.33	53.89	58.35	61.82	63.55
6.	M6	0	7.14	9.27	16.33	25.16	34.82	38.14	47.93	50.12	56.23	60.89	68.17
7.	M7	0	3.14	6.23	11.98	20.44	28.37	34.26	48.19	54.32	60.86	67.13	72.82
8.	M8	0	4.14	5.87	9.36	17.14	24.32	30.17	39.86	46.42	58.19	65.37	74.16
9.	M9	0	3.64	7.13	16.32	20.46	26.91	34.38	41.82	48.76	57.14	69.92	79.73
10	M10	0	4.06	6.18	11.31	17.96	22.71	29.48	36.17	44.81	52.28	59.16	66.97
11	M11	0	4.41	7.32	11.76	15.32	20.96	29.14	37.24	46.87	53.51	60.18	68.23
12	M12	0	4.85	9.40	13.26	21.32	28.84	36.73	45.15	51.33	62.81	70.73	78.34
13	M13	0	3.40	7.25	15.59	20.56	25.90	34.99	46.11	53.42	61.89	70.25	77.12
14	M14	0	4.95	8.77	15.06	22.93	30.04	38.10	47.68	59.30	67.85	75.08	80.02
15	M15	0	6.64	10.16	19.28	30.63	44.86	50.92	61.77	69.03	75.40	81.33	86.92
16	M16	0	8.49	13.06	21.57	29.42	37.23	45.81	53.15	59.31	62.84	66.05	70.13
17	M17	0	9.87	15.24	24.68	33.73	42.27	49.96	56.34	62.01	69.32	74.48	78.19
18	M18	0	8.15	15.86	28.81	37.33	48.98	57.26	61.07	69.63	73.12	79.06	81.03

The release rate of Artemether from the tableted microspheres for all the formulations. At 10th hours release rate of drug was between 61.24% - 86.92%. M15 formulation shows the maximum release and M4 shows the minimum release. It depicts that the release of artemether either from microspheres or from the tableted microspheres was found to be relatively near about same. A slight increase is observed in the release from tabletedmicrospheres.

KineticModelling

The dissolution data were analyzed according to various model dependent approaches (Zero order, First order, Higuchi, Hixson Crowell, Korsmeyer-Peppas) and the mode of drug release from the microspheric tablets was calculated plotting the curves. The kinetic model with highest value of coefficient of determination (R^2) was considered to be a more suitable model for all dissolution profiles.

Table (a): Kinetics of Drug release

Formulation Code	Zero Order		First Order		Higuchi	
	R ²	Slope	R ²	Slope	R ²	Slope
MT1	0.979	6.920	0.994	0.064	0.957	23.67
MT2	0.980	7.322	0.993	0.053	0.946	24.9
MT3	0.976	7.658	0.988	0.057	0.946	26.08
MT4	0.996	6.189	0.993	0.040	0.946	20.86
MT5	0.982	6.747	0.991	0.046	0.951	22.97
MT6	0.987	6.636	0.990	0.047	0.960	22.65
MT7	0.994	7.629	0.973	0.056	0.917	25.35
MT8	0.989	7.400	0.930	0.054	0.878	24.13
MT9	0.990	7.620	0.903	0.059	0.895	25.08
MT10	0.993	6.644	0.954	0.045	0.893	21.80
MT11	0.988	6.770	0.948	0.046	0.885	22.16
MT12	0.996	7.768	0.939	0.061	0.909	25.68
MT13	0.995	7.804	0.948	0.060	0.908	25.79
MT14	0.996	8.248	0.951	0.068	0.914	27.35
MT15	0.987	8.967	0.976	0.085	0.955	30.52
MT16	0.974	6.993	0.997	0.052	0.976	24.22
MT17	0.979	7.635	0.994	0.064	0.981	26.43
MT18	0.958	8.097	0.997	0.073	0.984	28.39

Table 7.3.11 (b): Kinetics of Drug release

Formulation Code	Korsmeyer – Peppas		Hisxon Crowell	
	R ²	Slope	R ²	Slope
MT1	0.831	1.225	0.995	0.146
MT2	0.820	1.213	0.993	0.157
MT3	0.850	1.287	0.990	0.168
MT4	0.851	1.206	0.998	0.125

MT5	0.797	1.158	0.992	0.141
MT6	0.732	1.061	0.995	0.141
MT7	0.869	1.286	0.986	0.166
MT8	0.848	1.224	0.957	0.160
MT9	0.839	1.222	0.943	0.172
MT10	0.839	1.176	0.972	0.137
MT11	0.819	1.149	0.966	0.141
MT12	0.801	1.169	0.967	0.176
MT13	0.849	1.249	0.972	0.175
MT14	0.807	1.195	0.974	0.192
MT15	0.762	1.180	0.994	0.227
MT16	0.679	1.023	0.994	0.154
MT17	0.652	1.013	0.998	0.181
MT18	0.674	1.068	0.994	0.200

The results of kinetic analysis provided the evidence that zero order was the best fit model for the dissolution data of all formulations as the plots showed the highest values of R^2 that indicated that the mode of drug release was independent of concentration of drug. All other models exhibited curvilinear plots having low values of R^2 when compared with that of zero order.

Conclusion

The objective of the present study was to fabricate and evaluate a combination of Artemether and Lumefantrine as tablets and to make Artemether in sustained form as to prolong its elimination time.

Artemether was formulated in form of microspheres and was then formed into the tablet along with the Lumefantrine. Artemether microspheres were prepared and compressed into compressible tablet by direct compression process using the compressible excipients along with Lumefantrine, which entails the convenience of a sustained release product in one.

The rationale behind this combination is that Artemether initially provides rapid symptomatic relief by reducing the number of parasites present before Lumefantrine eliminates any residual parasites. This is thought to minimize development of resistance because the malaria

parasites are never exposed to Artemether alone due to its rapid elimination. Although they may be exposed to Lumefantrine alone, the probability of resistance developing simultaneously to both drugs used in combination is thought to be low.

The Artemether microspheres were formed by solvent evaporation technique using ethyl cellulose as a polymer, in presence of polyvinyl alcohol as surfactant. Due to the sustained property of polymer and surfactant property of polyvinyl alcohol, formulated microspheres can result in controlled release of drug. Ethyl cellulose coated microparticles have also demonstrated their capability to absorb pressure and therefore save the coating from fracture during tablet manufacturing process.

Change in rpm also results in different particle size, entrapment efficiency and in vitro release from the microspheres. The increase in the stirring speed equals an approximately identical decrease

in the entrapment efficiency. Increasing the stirring speed delivers greater energy to the system, resulting in an increased breakdown of the forming microspheres and lower entrapment efficiency. The prepared microspheres with the mean size ranging between 14.80 μm to 26.42 μm showed particle size dependence on the stirring speed. The results confirmed that the microsphere mean size decreased with an increase in the stirring speed. The force of higher stirring distributes the internal phase into smaller droplets, resulting in the formation of smaller sized microspheres. The increase in the stirring rate results in the identical increase in the % drug release. All the formulations prepared at 600 rpm exhibited maximum drug release than their equivalent counterparts prepared at 400 and 200 rpm.

After successfully incorporating Artemether into microspheres, this study aimed to obtain tablets as a final oral dosage form. Artemether microspheres along with Lumefantrine were formulated into tablets by direct compression technique using the excipients. The microspheres were tableted using different concentration of superdisintegrants and were evaluated for various parameters.

Hence the present work suggest that, Artemether which has the lower half life and eliminates quickly from the body, when loaded with ethyl cellulose in form of microspheres and tableted along with Lumefantrine results in sustained release of drug in malaria. Therefore, Artemether and Lumefantrine in combination minimizes development of resistance as the malaria parasites are never exposed to artemether alone, so are considered as the best combination for treatment of malaria.

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Cite this article as:

Gautam M., Choukse R. and Patel R. (2021). Formulation and Evaluation of Combination of Artemether and Lumefantrine as Tablets, *Int. J. of Pharm. & Life Sci.*, 12(9):11-34.

Source of Support: Nil

Conflict of Interest: Not declared

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