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TO STUDY THE PREVALENCE OF MALARIA AND TO DETERMINE THE EFFICACY OF PREPARED NIOSOMES FOR COMBINATORIAL DRUG DELIVERY OF ANTI MALARIAL DRUGS

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Abstract

Background: Malaria is a caused by parasites that are transmitted through bites of Anopheles mosquitoes. Aim: The aim of the study is to assess the prevalence of malaria in the tertiary care hospital during the study period of 6 months. Methods: This a retrospective observational study conducting in both inpatients and outpatients diagnosed with malaria considering different age groups and other disease conditions. Data including the demographic details were obtained by medical history interview and niosomal formulation was subjected with the help of various suitable materials. Results & Discussion: 30 patients were admitted in the department of general medicine. The results were obtained from 30 patients with malaria disease who are enrolled in the study fulfilled after selection criteria on obtaining consent from the same among 30 patients. Peak prevalence of malaria occurred between the age group of 20-30.The total no. Of patients with malaria associated with hypertension and diabetes in males of age group between 20-60 was found to be 60% and females of age group between 20-60 was found to be 40%. Prevalence of stage–1(53%)is highest among the population. Conclusion: In this study, total 30 cases (n=30)were collected who have been diagnosed with malaria from the tertiary care hospital and studied the prevalence of occurrence of malaria between age groups and gender bias and prevalence of risk factors associated with malaria. The prepared niosomal drug delivery system and characterization was done and particle size (Span20< Brij35< tween 20<Tween 80), stability parameter, TEM & in vitro studies were performed in dialysis bag taking tween 80, tween 20, span 20, brij 35 niosomes and drug (primaquine) was compared by using RP-HPLC technique. The drug loaded niosomal preparations are effective for tween-80 niosomes for treatment of malaria.

Keywords: Malaria, Niosomal preparations, RP-HPLC technique, Non-ionic surfactants, Pharmaceutical preparations.

INTRODUCTION

Malaria is a caused by bites of infected female Anopheles mosquitoes. Malaria is caused by a type of microscopic parasite transmitted most commonly by mosquito bites. Liposomes that are made up of phospholipids [1-3]. The chemical stability, the low cost of excipients and the wide range of surfactant available to prepare niosomes make these carriers more attractive for both pharmaceutical and cosmetic applications [4-6].

AIM

- To assess the prevalence of malaria in the tertiary care hospital during the study period of 6 months.
- To determine the efficacy of prepared niosomes for anti malarial effect.

OBJECTIVES

- To study the prevalence of different stages of malaria.
- To evaluate risk factors associated with malaria for selected subjects.
- To study effectiveness of niosomes in malarial patients.
- Prepare stable and biocompatible niosomes of non-ionic surfactants and characterize them using various physico-chemical techniques.

- Stability evaluation of the plain and drug-loaded formulations.

MATERIALS AND METHODS

Materials and methods of drug loaded Niosomes

Polyoxyethylene, lauryl ether, sorbitan monostearate, Tween 80 (polyoxyethylene sorbitan monostearate), cholesterol, and deuterated water were obtained from Sigma Chemical Co.

Preparation of Blank and Drug-loaded Niosomes

The niosomes prepared by the conventional thin film method. The non-ionic surfactants, cholesterol were dissolved in chloroform in a round- bottomed flask and it was evaporated at 60 °C under reduced pressure at 100–150 rpm.

The obtained dry film was hydrated using Milli-Qwater. The dispersion was further sonicated for 60 min and then centrifuged to remove free surfactants and larger vesicles. It was hydrated using phosphate buffered saline (pH 7.4). The niosomal suspension was left to mature overnight at 4°C. It was followed by sonication and high speed centrifugation to obtain uniform sized Tween 80 drug co-loaded niosomes. The formation of niosomes was monitored by phase contrast microscopy.

Characterization of Niosomes

Optical microscopy

The dispersions were characterized under alight microscope after hydrating the thin film. Vesicles prior to sonication are known to be huge micro-sized structures and readily visible under a light microscope. Formation of vesicle scan be easily noted by the presence of multi lamellar spherical structures.

Particle size analyses

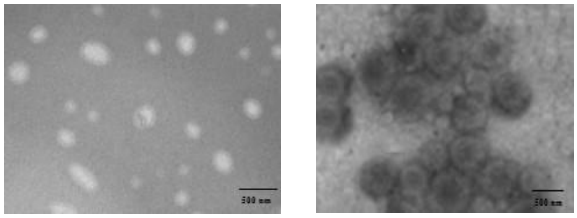
- The size of the niosomes is more dependent on the method of preparation and the subsequent downsizing and purification processes. Among the formulations, Span 20 niosomes exhibited the smallest particle size. The order of increasing particle size was: Span 20 < Brij 35 < Tween 20 < Tween 80. Span 20 has a molecular weight close to that of cholesterol, which may allow it to bind more tightly with cholesterol. This stronger interaction could result in smaller vesicular sizes compared to the other surfactants.

Phase transitions between micelles and vesicles by Turbidity measurements

- The structural transitions of the surfactant, cholesterol, were monitored by the optical density determined at λ= 500 nm.
- The concentration of cholesterol is increased, the surfactant micelles start solubilizing the cholesterol in it, until a thermodynamic equilibrium is achieved wherein the two systems can positively interact with each other forming bilayered structures, a peculiar characteristic attributed to vesicles.

Transmission electron microscopy

The morphology of hydrated niosomes was examined by Transmission electron microscopy using carbon coated gold grid. After absorption of the sample on the grid, the morphology was observed at 80 kV.



Tween80 Size:154.4nm | Brij35 Size:152.2 nm

Fig.01.TEM images of blank niosomes using various surfactants

Pyrene was used as a probe. Out of the five characteristic peaks, the difference of intensity ratio of first, third vibronic peaks of pyrene indicates the change of micro environment of the probe.

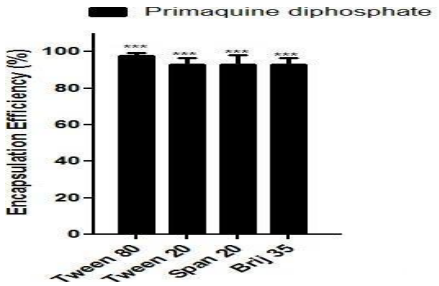
ΔGm values of PRI

Drug	KD	ΔGm (kJ/mole)
PRI	0.85 ± 0.20	0.34

- Kd=dissociation constant is the concentration of ligand

ENCAPSULATION EFFICIENCY DETERMINATION

The drug were centrifuged at 200 g for 5 min at 4°C. When co-loaded within niosomes, the encapsulation efficiency for the combined drugs was the highest in Tween 80

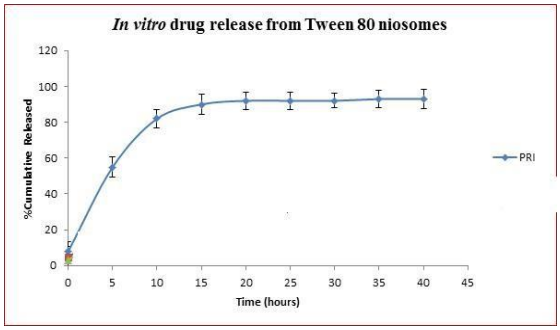


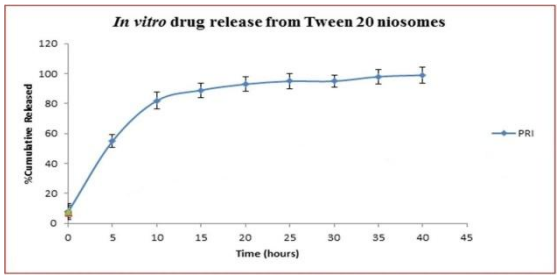
niosomes (PRI:97.5 ±1.3%).

FIG 02: Encapsulation Efficiency Determination

In-Vitro Drug Release

It was was determined using dialysis bag diffusion technique. The dialysis bags were hydrated in overnight. The drug-loaded niosome formulation was placed in a dialysis bag and it was placed in vessel containing 500 ml of dissolution medium consisting of 0.1 % Tween 80. The paddle speed was kept 50 rpm at 37 ± 2°C. At regular intervals 5 ml were removed from the vessel and replaced by fresh medium every time. Aliquots were centrifuged at 5000 rpm for 10 min and subsequently filtered through 0.22 µm syringe filter. The filtrates were assayed for drug concentration using the RP-HPLC. 80 niosomes [12-16].





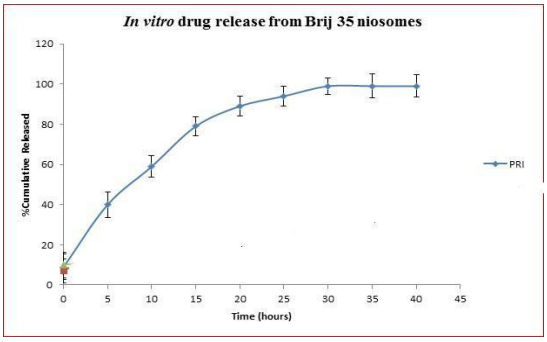


FIG 03: In Vitro Studies

Stability studies

The formulations were placed in vials, sealed for long term studies at 30°C±2°C and 65% RH±5%RH and 40°C ± 2°C and 75% RH ± 5% RH. As per modified International Conference on Harmonization guide lines. Samples were analyzed every 30 days for appearance, size, zeta potential and drug content.

Table 1: Ambient and accelerated studies of Tween 80 blank noisome formulations

Condition	Period	Size (nm)	pH	Zetapotential(mV)
30 ± 2°C 65 ± 5% RH	Day1	150 ± 17	7.42 ± 0.65	22 ± 5
	6 months	148 ± 21	7.35 ± 0.52	25 ± 4
40 ± 2°C 75 ± 5% RH	Day1	154±16	7.63 ± 0.65	23 ± 6
	6 months	152±13	7.49 ± 0.38	27 ± 3

Table 2: Ambient and accelerated studies of Tween 80 drug-loaded niosomes

Condition	Period	Size (nm)	pH	Zeta Potential (mv)	Drug Content (%)
30 ± 2°C 65 ± 5% RH	Day1	220 ± 15	7.39 ± 0.57	28 ± 4	99.07 ± 1.52
	6 months	217 ± 20	7.40 ± 0.43	27 ± 5	99.24 ± 1.38
40 ± 2°C 75 ± 5% RH	Day1	219 ± 18	7.57 ± 0.41	26 ± 6	99.16 ± 0.65
	6 months	223 ± 16	7.53 ± 0.44	27 ± 5	98.35 ± 0.83

METHODOLOGY

Study Site

This Retrospective study was conducted in the general medicine department of tertiary care hospital for a period of 6 months. Subjects who fulfilled the inclusion criterion were documented from the case sheets.

Duration of Study

The study conducted for a period of 6 months.

Study Participants:

The study was conducted of a total of 30 patients with different age groups who are diagnosed with malaria.

Inclusion Criteria

The study was chosen according to inclusion criteria who were diagnosed with malaria admitted in the hospital as well as out patients of both sex and different age groups.

Exclusion Criteria

- Patients with cancer.
- Pregnant
- Lactating women

Study Design

This a retrospective observational study conducting in both inpatients and outpatients diagnosed with malaria considering different age groups and other disease conditions.

Data Collection

Data including the demographic details were obtained by medical history interview.

RESULTS

A total no. of 30 patients who were admitted in the department of general medicine. The results were obtained from30 patients with malaria disease who are enrolled in the study fulfilled after selection criteria on obtaining consent from the same among 30 patients.

Prevalence between the Ages

Of the patients studied, males 2 and females 1(total 10%) were in the age group of 0-10, males 2 and females 2(total 13%) were in the age group of 10-20, males 8 and females 2(total 33%) were in the age group of 20-30, males 4 and females 3(total 23%) were in the age group of 30-40, males1 and females 2 (total10%) were in the age group of 40-50,males 3and females were in the age group of 50-60 as shown in the table and figure.

Table 3: Prevalence Between The Ages

Age	Males	Females	Percentage
0-10	2	1	10%
10-20	2	2	13%
20-30	8	2	33%
30-40	4	3	23%
40-50	1	2	10%
50-60	3	0	10%

Risk Factors Associated With Malaria

One of the total study populations, the most common risk factor associated with malaria are hypertension, diabetes. The total no. of patients with malaria associated with hypertension and diabetes in males of age group between 20-60 was found to be 60% and females of age group between 20-60 was found to be 40% as shown in table and figure.

Prevalance of Stages of Malaria in the Patients

Of the study, there are different stages of malaria. In stage 1males affected are 10 and females 6 total 16 patients (55%) and in stage 2 males affected are 4 and females 5 total 10 patients (33%) and in stage 3 males affected are 2 and females 3 total 5 patients(16%) as shown in table and figure.

Table 5: Prevalence of stages of malaria

STAGES	MALE	FEMALE	PERCENTAGE
1	10	6	53%
2	4	5	33%
3	2	3	16%

DISCUSSION

Anti malarials are predominantly used in the tropical and sub-tropical regions of the world. It is necessary to identify the effect of varying conditions in order to be able to use the excipients. Stable, niosomes have been prepared from various surfactant systems using thin film hydration method7-14. They have been further characterized using TEM, Particle size, micro polarity measurement, encapsulation efficiency and release studies. Tween 80 system was found to be best suited for the encapsulation of drug combined12-14. High penetration property of niosome encapsulated through biological membrane will help in the delivery of the anti- malarial drug15-17.

CONCLUSION

In this study, a total of 30 cases (n = 30) diagnosed with malaria were collected from a tertiary care hospital. The prevalence of malaria was analyzed across different age groups and genders, along with associated risk factors. Among the 30 patients, 10 (33%) were in the age group of 20–30 years, comprising 8 males and 2 females. Based on comorbid risk factors such as hypertension (HTN) and diabetes mellitus (DM), 60% of the affected males and 40% of the affected females were diagnosed with these conditions. Malaria staging among the patients was distributed as follows: Stage 1 – 53%, Stage 2 – 33%, and Stage 3 – 16%. In this study, a niosomal drug delivery system was prepared and characterized. Particle size analysis indicated the following order: Span 20 < Brij 35 < Tween 20 < Tween 80. Additional characterization parameters included stability studies, transmission electron microscopy (TEM), and in-vitro release studies using a dialysis bag. The niosomes were formulated with various surfactants (Tween 80, Tween 20, Span 20, Brij 35), and the antimalarial drug primaquine was incorporated. Drug release was evaluated and compared using the RP-HPLC technique. Among the formulations, Tween 80-based niosomes demonstrated the most effective results for malaria treatment. Overall, the niosomal preparations showed improved efficacy compared to conventional pharmaceutical formulations. A multidisciplinary approach incorporating such advanced drug delivery systems is recommended for the effective prevention and treatment of malaria.

REFERENCES

1. Korenromp EL, Williams BG, Gouws E, Dye C, Snow RW. Measurement of trends in childhood malaria mortality in Africa: an assessment of progress toward targets based on verbal autopsy. Lancet Infectious Diseases. 2003; 3: 349–358.
2. Bhadra D. Glycodendrimeric nanoparticulate carriers of primaquine phosphate for liver targeting. International Journal of Pharmaceutics. 2005; 295: 221–233.
3. Kumar GP, Rajeshwarrao P. Nonionic surfactant vesicular systems for effective drug delivery – an overview. Acta pharmaceutica sinica B. 2011; 1: 208–219.
4. LooareesuwanS,BuchachartK,WilairatanaP,ChalermrutK,RattanapongY,mradeeS, Siripiphat S, Chullawichit S, Thimasan K, Ittiverakul M, Triampon A, WalshDS. Primaquine-tolerant vivaxmalaria in Thailand.Annals of Tropical Medicine & Parasitology. 1997; 91: 939–943.
5. Murphy GS, Basri H, Purnomo, Andersen EM, Bangs MJ, Mount DL, Gorden J, Lal AA, PurwokusumoAR,HarjosuwarnoS,K.Sorensen,HoffmanSL.Vivaxmalariaresistant to treatment and prophylaxis with chloroquine.Lancet.1993;341: 96–

100.
6. Vauthier C, Couvreur P. Nanomedicines: a new approach for the treatment of serious diseases. Journal of Biomedical Nanotechnology. 2007; 3: 223–234.
7. Mihaly GW, Ward SA, Edwards G, Nicholl DD, Orme ML, Breckenridge AM: Pharmacokinetics of primaquineinman. I. Studies of the absolute bioavailability yandeffects of dose size. British Journal of Clinical Pharmacology.1985;19(6):745–750.
8. Vale, Nuno; Moreira, Rui; Gomes, Paula G. Primaquine revisited six decades after its discovery. European Journal of Medicinal Chemistry. 2009; 44(3): 937–953.
9. ManconiM, Valenti D,Sinico C, LaiF,Loy G,FaddaAM. Niosomesas cariers fortretinoin
10. II. Influence of vesicular incorporation on tretinoin photostability. International Journal of Pharmaceutics. 2003; 260: 261–272.
11. Mao M, Huang J, Zhu B, YeJ. The transition from vesicles to micelles induced by octane in aqueoussurfactanttwo-phasesystems.TheJournalofPhysicalChemistryB.2002; 106:219– 225.
12. Mukherjee B, Patra B, Layak B, Mukherjee A. Sustained release of acyclovir from nano- liposomes and nano-niosomes: An *in vitro* study. International Journal of Nanomedicine. 2007; 2: 213–225.
13. White NJ, Nosten F, Looareesuwan S, Watkins WM, Marsh K, Snow RW, Kokwaro G, Ouma J, Hien TT, Molyneux MW. Averting a malaria disaster. Lancet. 1999;353: 1965– 1967.
14. Marques J, Valle-Delgado JJ, Urbán P, et al. Adaptation of targeted nanocarriers to changing requirements in antimalarial drug delivery. Nanomedicine. 2017;13(2):515–525. doi: 10.1016/j.nano.2016.09.010. [DOI] [PMC free article] [PubMed] [Google Scholar]
15. Akbarzadeh A, Rezaei-Sadabady R, Davaran S, et al. Liposome: classification, preparation, and applications. Nanoscale Res Lett. 2013;8(1):102. doi: 10.1186/1556-276x-8-102. [DOI] [PMC free article] [PubMed] [Google Scholar]
16. Aderibigbe BA, Mhlwatika Z. Dual release kinetics of antimalarials from soy protein isolate-carbopol-polyacrylamide based hydrogels. J Appl Polym Sci. 2016;133(37):43918. doi: 10.1002/app.43918. [DOI] [Google Scholar]
17. Thakkar M, Brijesh S. Physicochemical investigation and in vivo activity of anti-malarial drugs co-loaded in Tween 80 niosomes. J Liposome Res. 2018;28(4):315–321. doi: 10.1080/08982104.2017.1376684. [DOI] [PubMed] [Google Scholar]
18. Ouji M, Augereau JM, Paloque L, Benoit-Vical F. Plasmodium falciparum resistance to artemisinin-based combination therapies: a sword of Damocles in the path toward malaria elimination. Parasite. 2018;25:24. doi: 10.1051/parasite/2018021. [DOI] [PMC free article] [PubMed] [Google Scholar]
19. Moyo, P., Mugumbate, G., Eloff, J. N., Louw, A. I., Maharaj, V. J., and Birkholtz, L. M. (2020). Natural products: A potential source of malaria transmission blocking drugs? Pharmaceuticals (Basel) 13(9), 251. doi:10.3390/ph13090251.
20. Moyo, P., Mugumbate, G., Eloff, J. N., Louw, A. I., Maharaj, V. J., and Birkholtz, L. M. (2020). Natural products: A potential source of malaria transmission blocking drugs? Pharmaceuticals (Basel) 13(9), 251. doi:10.3390/ph13090251
21. Thomas S, Ravishankaran S, Asokan A, Justin NJA, Kalsingh TMJ, Mathai MT, et al. Socio-demographic and household attributes may not necessarily infuence malaria: evidence from a cross sectional study of households in an urban slum setting of Chennai. India Malar J. 2018;17:4.