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FORMULATION AND DEVELOPMENT OF COLON TARGETED MATRIX TABLET OF TENOFOVIR DF

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ABSTRACT

Tenofovir disoproxil fumarate (a prodrug of tenofovir), marketed by Gilead Sciences under the trade name Viread®, belongs to a class of antiretroviral drugs known as nucleotide analogue reverse transcriptase inhibitors (nRTIs), which block reverse transcriptase, an enzyme crucial to viral production in HIV-infected people. In vivo tenofovir disoproxil fumarate is converted to tenofovir, an acyclic nucleoside phosphonate (nucleotide) analog of adenosine 5'-monophosphate. Tenofovir inhibits the activity of HIV reverse transcriptase by competing with the natural substrate deoxyadenosine 5'-triphosphate and, after incorporation into DNA, by DNA chain termination. The present investigation was concerned with the development of the colon targeted tablets, which after oral administration were designed to prevent the drug release in stomach and small intestine. It improves the bioavailability of the drug as well as its half life. Various formulations were developed by using release rate controlling polymers like *Xanthan Gum*, Guar Gum by direct compression method. Wet granulation method was followed to manufacture the matrix tablets of Tenofovir DF. The tenofovir df matrix tablets were further coated with Eudragit S-100 solution. All the tablets were evaluated for following different parameters which includes general appearance Thickness and diameter, Drug content, hardness, Friability, Uniformity of weight, Dissolution rate studies etc.

KEYWORDS: Tenofovir disoproxil fumarate, Matrix tablet, wet granulation, Guar gum, Eudragit.

INTRODUCTION

Tenofovir disoproxil fumarate (a prodrug of tenofovir), marketed by Gilead Sciences under the trade name Viread®, belongs to a class of antiretroviral drugs known as nucleotide analogue reverse transcriptase inhibitors (nRTIs), which block reverse transcriptase, an enzyme crucial to viral production in HIV-infected people.In vivo tenofovir disoproxil fumarate is converted to tenofovir, an acyclic nucleoside phosphonate (nucleotide) analog of adenosine 5'-monophosphate.

Tenofovir inhibits the activity of HIV reverse transcriptase by competing with the natural substrate deoxyadenosine 5'-triphosphate and, after incorporation into DNA, by DNA chain termination. Specifically, the drugs are analogues of the naturally occurring deoxynucleotides needed to synthesize the viral DNA and they compete with the natural deoxynucleotides for incorporation into the growing viral DNA chain. However, unlike the natural deoxynucleotides substrates, NRTIs and NTRTIs (nucleoside/tide reverse transcriptase inhibitors) lack a 3'-hydroxyl group on the deoxyribose moiety. As a result, following incorporation of an NRTI or an NtRTI, the next incoming deoxynucleotide cannot form the next 5'-3' phosphodiester bond needed to extend the DNA chain. Thus, when an NRTI or NtRTI is incorporated, viral DNA synthesis is halted, a process known as chain termination. All NRTIs and NtRTIs are classified as competitive substrate inhibitors.

Tenofovir is used with other medications to help control HIV infection, thereby improving quality of life. It helps to decrease the amount of HIV in the body so immune system can work better. It also lowers risk of getting HIV disease complications (such as new infections, cancer).

MATERIALS AND METHODS

Wet granulation method was followed to manufacture the matrix tablets of Tenofovir DF. Six different formulations (F1, F2, F3, F4, F5 & F6) were prepared by Wet granulation method. All the polymers selected, drug and excipients were passed through sieve no. 40 before using into formulation. The amount and ratio of drug and polymers were weighed as per given in table No. 1 and all the formulation were used for further evaluations parameters. Polymers selected for tablets are:

- Xanthan Gum
- Guar Gum

Steps involved in the manufacture of tablets:-

First the drug; polymer and other excipients selected were weighed. Required quantity of drug, polymer, Fillers (Excipients like *Xanthan Gum*, Guar Gum are natural polymer) were weighed properly and transferred into mortar pastle and mix & triturate then added binder solution (isopropyl alcohol + pvp) dropwise. Prepared wet mass and further passed through sieve, drying, screening, and punch the granules Tablet was prepared.

Enteric coating of the matrix tablets

The tenofovir df matrix tablets were further coated with Eudragit S-100 solution. A different concentration (1% w/v) of coating solution of Eudragit S-100 was prepared in a mixture of Isopropyl alcohol: acetone (1:1). The coating of the matrix tablets was performed by immersion in the coating solution followed by spray coating technique.

Evaluation of tablets

All the tablets were evaluated for following different parameters which includes.

General Appearance

Five tablets from different batches were randomly selected and organoleptic properties such as color, odor, taste, shape, were evaluated. Appearance was judged visually. Very good (+++), good (++), fair (+) poor (-), very poor (- -).

Thickness and diameter

Thickness and diameter of tablets were determined using Vernier caliper. Five tablets from each batch were used, and an average value was calculated.

Drug content

Twenty tablets were taken and amount of drug present in each tablet was determined. The tablets were crushed in a mortar and the powder equivalent to 100mg of drug was transferred to 100ml standard flask. The powder was dissolved in 50 ml of 0.1 N HCL and made up to volume with of 0.1 N HCl. The sample was mixed thoroughly and filtered through a 0.45µ membrane filter. The filtered solution was diluted suitably and for drug content by UV spectrophotometer at λ max of 258 nm using of 0.1 N HCL as blank.

Hardness

For each formulation, the hardness of five tablets was determined using the Monsanto hardness tester.

Friability

The friability of a sample of 10 tablets was measured using a Friability tester (Electro Lab). Ten tablets were weighed, rotated at 25 rpm for 10 minutes. Tablets were reweighed after removal of fines (dedusted) and the percentage of weight loss was calculated.

Uniformity of weight

Twenty tablets were randomly selected from each batch individually weighed, the average weight and standard deviation of 20 tablets was calculated.

Dissolution rate studies

The prepared tablets were evaluated for *in vitro* drug release. The drug release studies were carried out using USP XXII paddle type Dissolution test apparatus. The dissolution study was carried out in 900 ml dissolution medium which was stirred at 75 rpm maintained at 37±0.2°C. The scheme of using the simulated fluids at different timing was as follows:

- *1st hour:* Simulated gastric fluid (SGF) of pH 1.2.
- *2th to 12th hour:* Simulated intestinal fluid (SIF) of pH 6.8.

A tablet placed in dissolution media (900 ml) at 37±0.2°C. Samples were withdrawn at different time interval and compensated with same amount of fresh dissolution medium. Volume of sample withdrawn was made up to 10ml by PBS (pH 6.8). The samples withdrawn were assayed spectrophotometrically at 258.0 nm using UV visible spectrophotometer. The release of drug was calculated with the help of Standard curve of Tenofovir DF.

The observations of drug release for the drug in uncoated formulation and coated formulation is tabulated in Table.

Mathematical treatment of *in-vitro* release data: The quantitative analysis of the values obtained in dissolution/release tests is easier when mathematical formulas that express the dissolution results as a function of some of the dosage forms characteristics are used.

1. Zero-order kinetics: The pharmaceutical dosage forms following this profile release the same amount of drug by unit of time and it is the ideal method of drug release in order to achieve a pharmacological prolonged action. The following relation can, in a simple way, express this model:

$$Q_t = Q_o + K_o t$$

where Q_t is the amount of drug dissolved in time t, Q_o is the initial amount of drug in the solution (most times, Q_o=0) and K_o is the zero order release constant (Bourne, 2002).

2. First-order kinetics: The following relation expresses this model:

$$\log Q_t = \log Q_o + \frac{K_1 t}{2.303}$$

where Q_t is the amount of drug dissolved in time t, Q_o is the initial amount of drug in the solution and K_1 is the zero order release constant.[56]

3. Higuchi model: Higuchi developed several theoretical models to study the release of water-soluble and low soluble drugs in semi-solid and/or solid matrixes. Mathematical expressions were obtained for drug particles dispersed in a uniform matrix behaving as the diffusion media.[56-57]

4. Korsmeyer-Peppas model: Korsmeyer *et al.* used a simple empirical equation to describe general solute release behaviour from controlled release polymer matrices:

$$\frac{M_t}{M_\infty} = a\, t^n$$

OBSERVATION

Table 1: Various formulations of Tenofovir DF Gastro Retentive tablets.

Excipients(mg)	F1	F2	F3	F4	F5	F6
Tenofovir DF	100	100	100	100	100	100
MCC	35	35	35	35	35	35
DBP	150	150	150	150	150	150
Lactose	250	150	250	150	250	150
Starch	35	35	35	35	35	35
SSG	20	20	20	20	20	20
Xanthan Gum	100	200	-	-	50	100
Guar Gum	-	-	100	200	50	100
Mg *Stearate*	5	5	5	5	5	5
Talc	5	5	5	5	5	5
Total Weight	700	700	700	700	700	700

Table 7.2: Result of pre-compression properties of Tenofovir DF FGR tablets.

Material	Angle of repose (Degree)	Bulk density (gm/ml)	Tapped density (gm/ml)	Compressibility index	Hausner ratio
Tenofovir DF					
F1	32.25	0.714	0.800	10.714	1.120
F2	31.45	0.690	0.806	14.483	1.169
F3	30.25	0.699	0.826	15.385	1.182
F4	32.23	0.704	0.833	15.493	1.183
F5	31.12	0.690	0.826	16.552	1.198
F6	32.25	0.676	0.840	19.595	1.244

Table 3: Results of post compression properties of Tenofovir DF FGR tablets.

Formulation code	Thickness (mm)	Hardness (kg/cm2)	Weight variation (mg)	Friability (%)	Drug content (%)
F1	3.21±0.12	5.4	705±5	0.580 ± 0.10	98.12±0.45
F2	3.20± 0.14	5.3	710±4	0.510± 0.08	98.98±0.25
F3	3.22± 0.12	5.5	712±6	0.560 ± 0.12	98.45±0.23
F4	3.22± 0.13	5.8	705±7	0.554 ± 0.04	98.15±0.14
F5	3.23± 0.16	5.4	713±5	0.548 ± 0.07	98.78±0.45
F6	3.25± 0.14	5.3	715±3	0.545± 0.05	99.45±0.56

Table 6: Interpretation of diffusional release mechanisms.

Release exponent (*n*)	Drug transport mechanism	Rate as a function of time
0.5	Fickian diffusion	$t^{-0.5}$
0.5<*n*<1.0	Anomalous transport	t^{n-1}
1.0	Case-II transport	Zero-order release
Higher than 1.0	Super Case-II transport	t^{n-1}

Table 7: Cumulative % drug release of all formulation (F1-F6).

S. No.	Dissolution medium	Time (hrs)	% Cumulative Drug Release					
			F1	F2	F3	F4	F5	F6
1.	SIF (pH 6.8)	0.5	2.25	2.35	3.3	2.5	4.5	1.45±0.45
2.		1	5.65	6.65	4.45	5.12	6.45	3.65±1.25
3.	SIF (pH 6.8)	1.5	12.25	10.12	15.65	13.12	21.45	22.26±1.36
4.		2	22.65	19.98	23.12	20.14	25.65	33.56±2.45
5.		3	36.65	30.25	30.25	25.65	33.14	54.56±3.65
6.		4	45.56	40.56	40.56	38.98	40.65	64.58±4.58
7.		5	59.98	55.56	60.25	55.45	60.56	75.58±3.25
		6	60.25	54.47	65.45	60.15	69.98	83.36±2.56
8.		8	73.25	70.12	75.45	73.12	78.98	95.45±1.25
9.		12	78.98	68.78	65.45	68.98	89.98	99.89±3.36

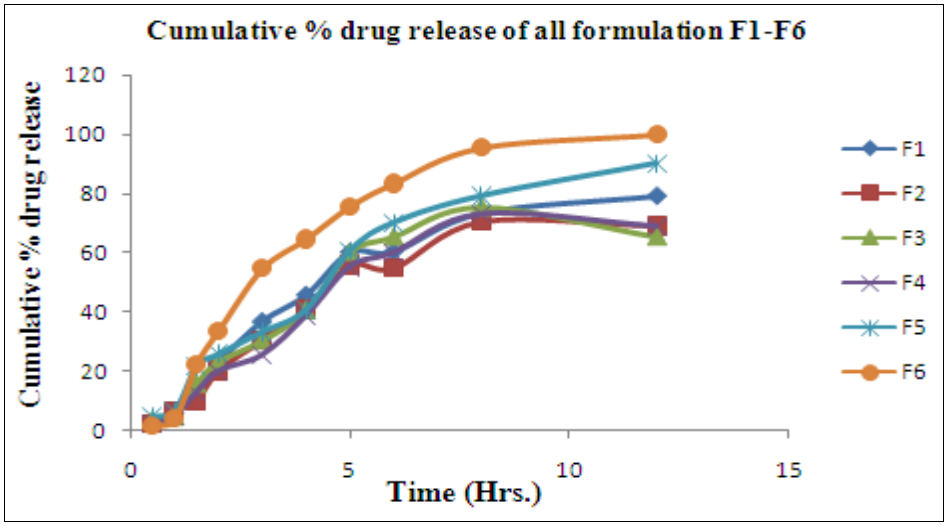

Figure 1: Graph of Cumulative % drug release of all formulation.

Table 8: Cumulative % drug release.

S. No.	Dissolution medium	Time (hrs)	% Cumulative Drug Release Tenofovir DF FGR tablets F6
1.	SIF (pH 6.8)	0.5	1.45±0.45
2.		1	3.65±1.25
3.	SIF (pH 6.8)	1.5	22.26±1.36
4.		2	33.56±2.45
5.		3	54.56±3.65
6.		4	64.58±4.58
7.		5	75.58±3.25
		6	83.36±2.56
8.		8	95.45±1.25
9.		12	99.89±3.36

Release kinetics of Tenofovir DF floating tablets

Table 9: *In-vitro* drug release data for optimized formulation F6.

Time (h)	Square Root of Time(h)$^{1/2}$	Log Time	Cumulative *% Drug Release	Log Cumulative % Drug Release	Cumulative % Drug Remaining	Log Cumulative % Drug Remaining
0.5	0.707	-0.301	1.45	0.161	1.45	0.161
1	1.000	0.000	3.65	0.562	3.65	0.562
1.5	1.225	0.176	22.26	1.348	22.26	1.348
2	1.414	0.301	33.56	1.526	33.56	1.526
3	1.732	0.477	54.56	1.737	54.56	1.737
4	2.000	0.602	64.58	1.810	64.58	1.810
5	2.236	0.699	75.58	1.878	75.58	1.878
6	2.449	0.778	83.36	1.921	83.36	1.921
8	2.828	0.903	95.45	1.980	95.45	1.980
12	3.464	1.079	99.89	2.000	99.89	2.000

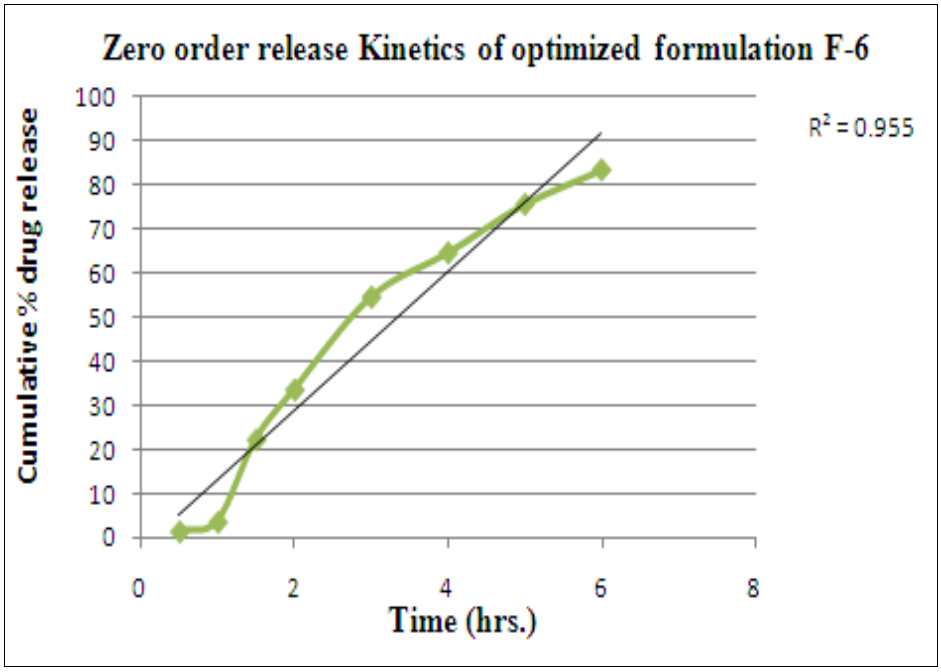

Figure 2: Cumulative % drug released Vs Time.

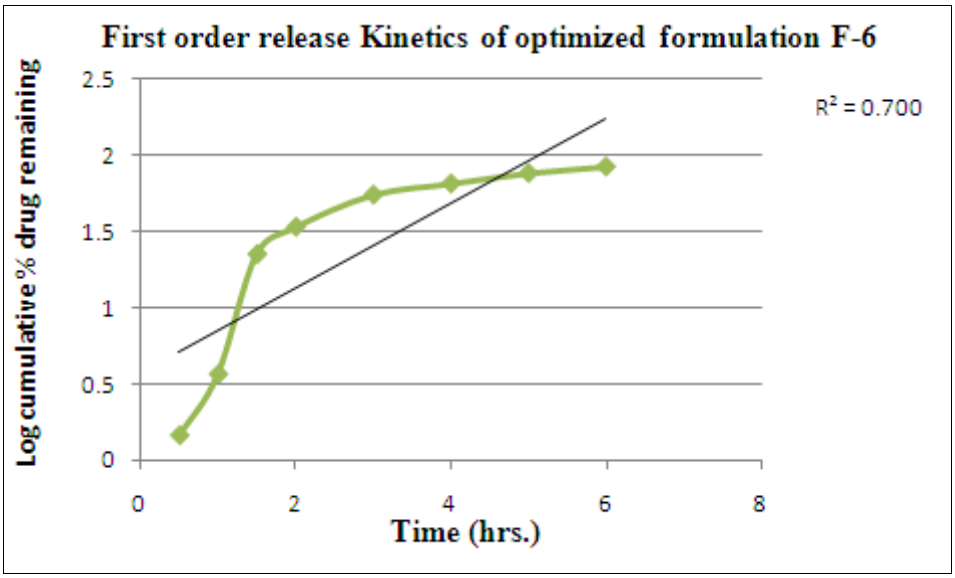

Figure 3: Log cumulative % drug remaining Vs Time.

Table 10: Regression analysis data of Tenofovir DF Tablets.

Batch	Zero Order	First Order
	R^2	R^2
F6	0.955	0.700

When the regression coefficient values of were compared, it was observed that 'r' values of zero order was maximum i.e. 0.955 hence indicating drug release from formulations was found to follow zero order kinetics.

CONCLUSION

Results of the present research work demonstrate that the combination of polymers successfully employed for formulating the colon release matrix tablets of Tenofovir DF. The present investigation was concerned with the development of the colon targeted tablets, which after oral administration were designed to prevent the drug release in stomach and small intestine. It improves the bioavailability of the drug as well as its half life. Various formulations were developed by using release rate controlling polymers like *Xanthan Gum,* Guar Gum by direct compression method. Developed film coated colon targeted tablets possessed the required physicochemical parameters such as hardness, friability, weight variation, drug content. Drug release studies shows that F6 shows good release behavior in colon and restricts release in stomach and intestine as compare to F1–F6. Therefore, it was concluded that the most satisfactory formulation is (F6).

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