6.0 *IN-VITRO* DRUG RELEASE STUDY

6.1 MATERIALS: Sodium carbonate was purchased from SD fine chemical (Mumbai, India). Folin Ciocalteu's phenol reagent and gallic acid from SRL Pvt. Ltd. (Mumbai, India).

6.2 PROCEDURE: *In-vitro* total polyphenol release study of CFE loaded hydrogel sheet was carried out using a vertical type of modified Franz diffusion (1-3). Dialysis membrane (cut-off 12,000 Da; HIMEDIA, Mumbai, India) was mounted on donor compartment with a diffusion surface area of 2.5 cm^2. Hydrogel sheet was placed in donor compartment. The receptor compartment (beaker) having 50 ml of SWF was covered to prevent the evaporation of release medium. The receptor compartment solution (50 mL of SWF) was stirred at 50 rpm speed using magnetic stirrer for 24 hr Samples of 3 ml were withdrawn from the beaker at specified time intervals (1, 2, 3, 4, 5, 6, 7, 8, 24 & 48 hr) and were replaced by same volume of fresh SWF. Colorimetric method based on Folin-Ciocalteu's phenol reagent was used to determine total polyphenol content of the aliquots (13). Aliquots were analyzed at 725 nm using UV-Visible spectrophotometer (Shimadzu, Japan). The total polyphenol content was expressed as mg of gallic acid equivalent/g of sample (mg GAE/g).The study was performed in three replicate.

6.3 KINETICS OF DRUG RELEASE

In order to examine the release mechanism of the prepared CFE-Hydrogel sheet, the result of the in vitro release study was fitted to following equations (4):

6.3.1 ZERO ORDER RELEASE EQUATION

$Q = k_0 t$ ……Equation (6.1)

Where, Q = amount of drug release at time t

K_0 = zero order release constant

t = time

Regression value of plot of amount of drug release versus time t gives the idea of release mechanism. R2 value nearer to indicating zero order release (4).

6.3.2 FIRST ORDER RELEASE EQUATION

$\ln (100-Q) = \ln Q_0 - K_1 t$ …….. Equation (6.2)

Where, Q = amount of drug release at time t

K_1 = first order release constant

The regression coefficient (R^2) value obtained from the log % ARR (Amount remaining to release) versus time, nearer to 1 indicates first order release. The dosage forms containing water soluble drug in porous matrices follow this model (5).

6.3.3 HIGHUCHI SQUARE ROOT OF TIME EQUATION

$Q = K_h t^{1/2}$ …….. Equation (6.3)

Where, Q = amount of drug release at time t

K_h = Higuchi square root of time release constant

The regression co-efficient of percentage drug release versus square root of time nearer to one Indicates anomalous release (6, 7). This relation can be used to describe the dissolution from several types of modified release pharmaceutical dosage form, as in the case of some Trans dermal systems and matrix with water soluble drug (8, 9).

6.3.4 KORSMEYER-PEPPAS EQUATION

$$\text{Log } (M_t/M_\alpha) = \text{Log } K + n \text{ Log } t \quad \text{.....Equation (6.4)}$$

Where, M_α = total drug release after infinite time

M_t/M_α = fractional drug release at time t

K = kinetic constant incorporation structural and geometrical characteristic of the drug/polymer system (devices).

n = diffusion exponents that characterizes the mechanism of drug release

t = time

Graph of log % drug release versus log time was plotted. n value was obtained and release kinetic was determined using following specifications. This type of drug release is controlled by combination of polymer swelling erosion and diffusion through the hydrated matrix (Diffusion and chain relaxation).

- The value of $n < 0.5$ or $n = 0.5$ indicating fickian diffusion
- The value of n between 0.5 to 1 indication non-fickian release
- The value of n=1, indication the zero order release or case 2 transport
- The value of $n > 1$, indication the super case 2 transport

This model is generally used to analyze the release of pharmaceutical polymeric dosage forms, when the release mechanism is not well known or when more than one type or release phenomena could be involved (10, 11).

6.3.5 HIXON-CROWELL CUBE ROOT MODEL EQUATION

Kinetic equation: $Q_0^{1/3} - Q_t^{1/3} = K_H C.t$ …… Equation (6.5)

Plot: $(Q_0^{1/3} - Q_t^{1/3})$ vs. t

Where, Q_o = initial concentration of drug present

Q_t = amount of drug release at time t

K_H is the kinetic constant for distribution from constantly changing surface area observed in slow dissolving tablet (12).

6.4 RESULTS AND DISCUSSION

The *in-vitro* release profiles of polyphenolic compound from CFE-hydrogel sheet are shown in figure 6.1. The release curve suggests initial fast release during first hour which may be due to the surface absorbed CFE. Later on release phase depicts a diffusive pattern of release typical of a hydrating, hydrogelling matrix system.

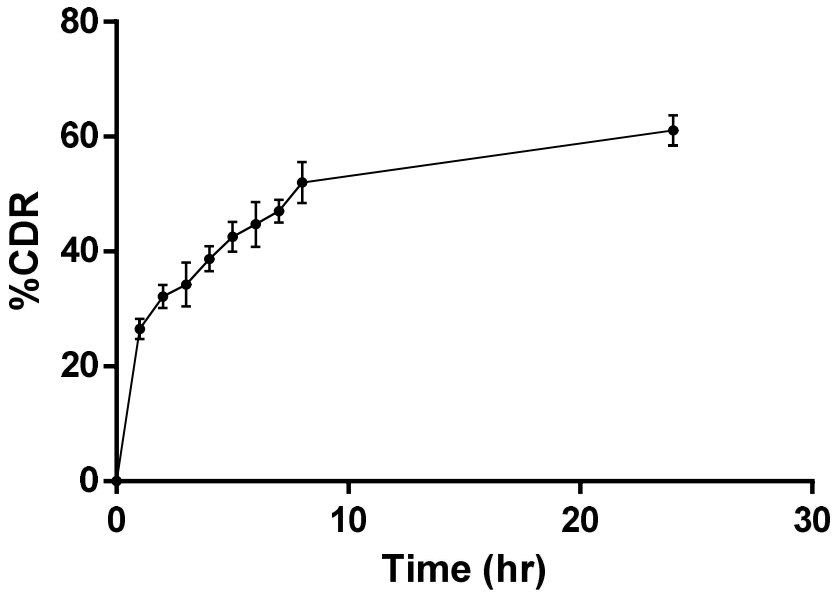

FIGURE 6.1: *In-vitro* cumulative total polyphenol compound release profile from CFE loaded hydrogel sheet

Kinetics of drug release: The release data were fitted to various kinetic models in order to calculate the release constant and regression coefficient (R^2). Results are shown in table 6.1 & Figure 6.2

TABLE 6.1: Release rate constants and correlation coefficients (R^2) calculated after fitting the release profiles obtained using different mathematical models

Mathematical models	**CFE loaded Hydrogel sheet**	
	R^2	**Release rate constants**
Zero order	0.767	1.351
First order	0.841	-0.011
Highuchi	0.915	9.077
Korsmeyer peppas	0.970	0.280
Hixon-crowell cube root	0.767	-1.351

CFE-loaded hydrogel sheet followed Korsmeyer Peppas release kinetics as Korsmeyer equation showed better fit than Highuchi, first order and zero order equations. This type of drug release is controlled by combination of polymer swelling erosion and diffusion through the hydrated matrix (Diffusion and chain relaxation).

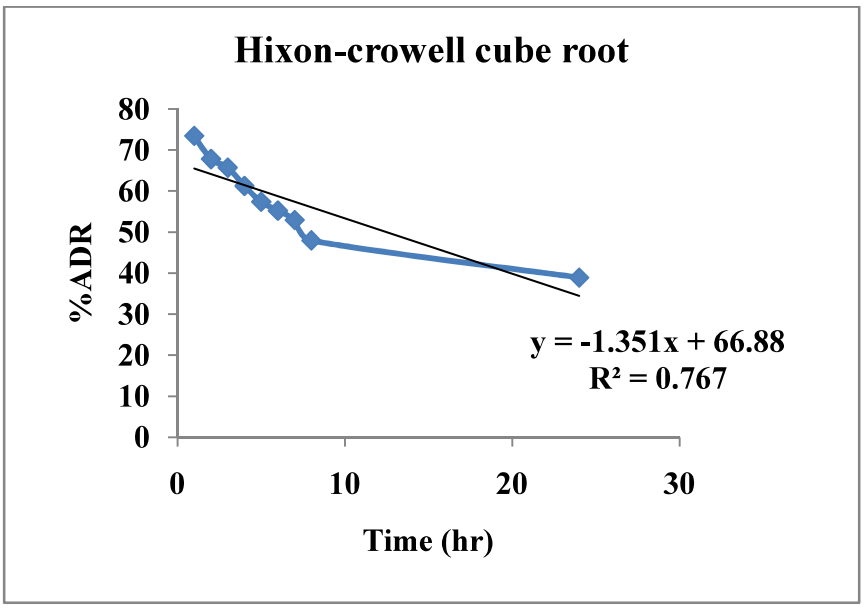

FIGURE 6.2: Hixon-crowell cube root release kinetics profile of total polyphenol release from CFE loaded hydrogel sheet

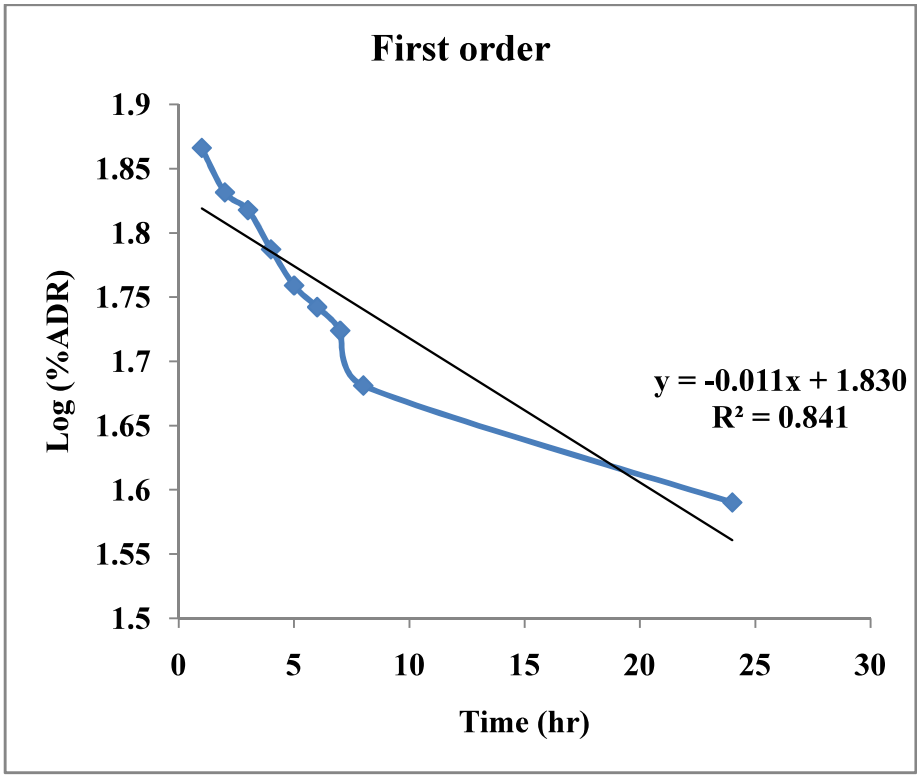

FIGURE 6.3: First order release kinetics profile of total polyphenol release from CFE loaded hydrogel sheet

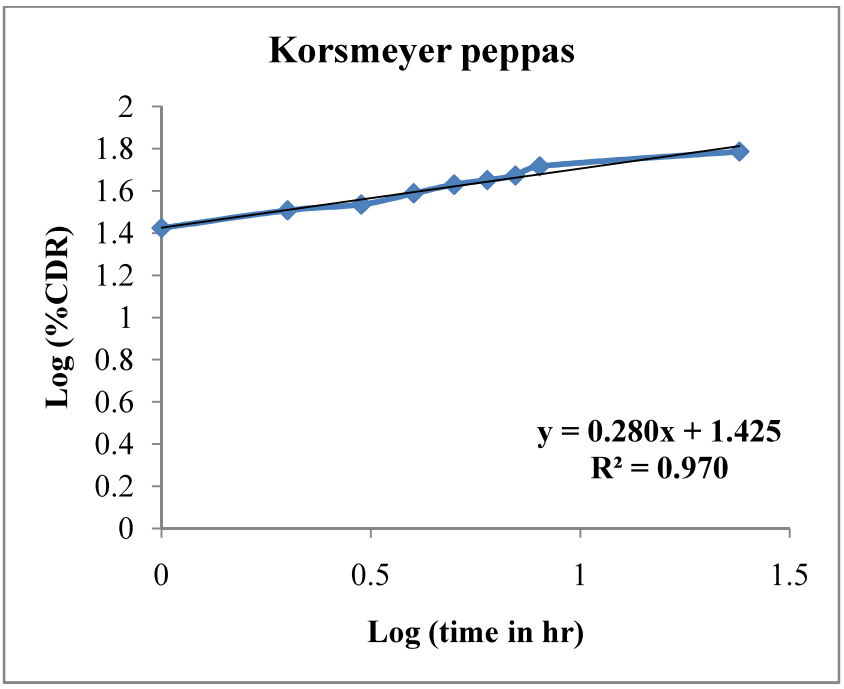

FIGURE 6.4: Korsmeyer peppas release kinetics profile of total polyphenol release from CFE loaded hydrogel sheet

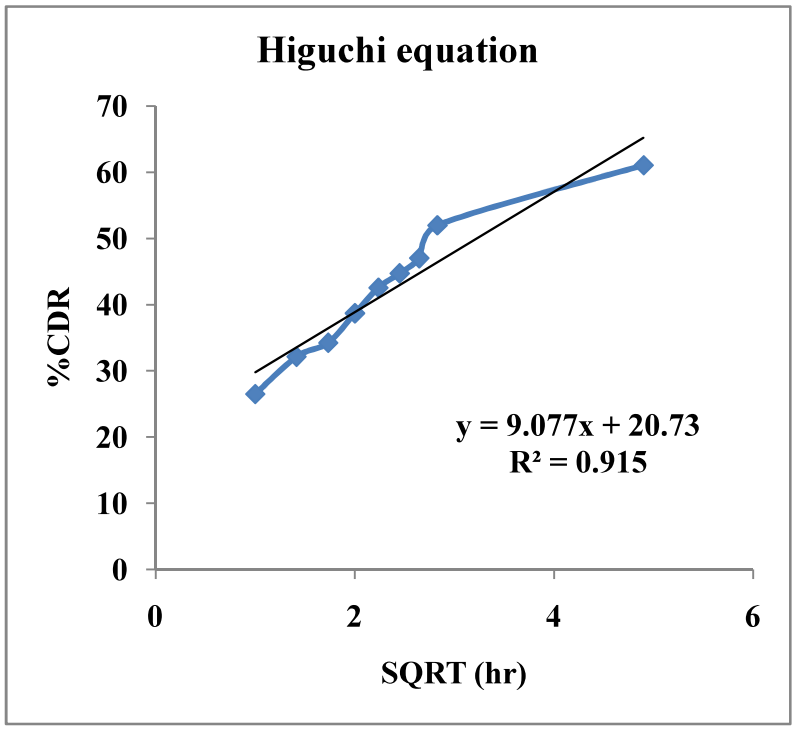

FIGURE 6.5: Higuchi release kinetics profile of total polyphenol release from CFE loaded hydrogel sheet

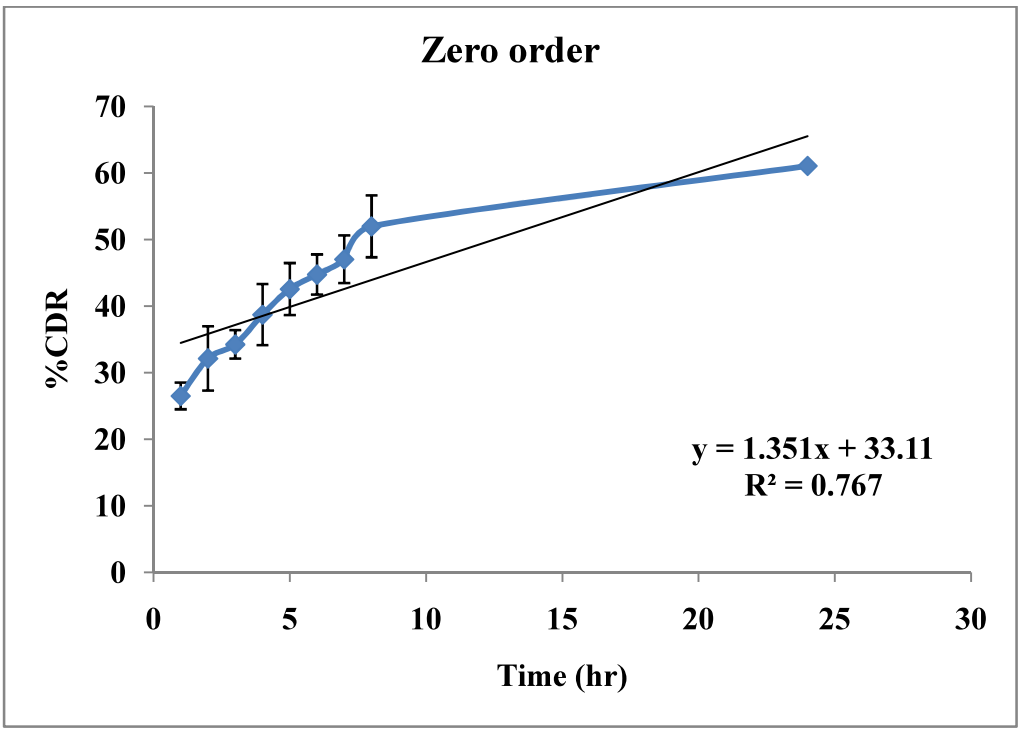

FIGURE 6.6: Zero order release kinetics profile of total polyphenol release from CFE loaded hydrogel sheet

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