# Research Article

# Formulation, bio-equivalence and *in vitro - in vivo* correlation studies of a cashew gum-based clarithromycin sustained-release tablet

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**Abstract**

**Objective:** This study was conducted to develop a sustained release tablets of clarithromycin by wet granulation based on di erent combination ratios of cashew gum (CG) and hydroxy propyl methyl cellulose (HPMC) as retarding hydrophilic colloidal polymers. **Materials and methods**: The formulation was optimized on the basis of acceptable tablet properties and *in vitro* drug release. **Results:** Clarithromycin, CG and the other tablet excipients showed no physicochemical incompatibilities as observed in the FTIR and DTA analyses. There was no significant difference in drug release between the innovator product and formulation F6 when the HPMC concentration was modified in low percentage while increasing CG concentration besides satisfying FDA and compendia requirements for the f1 and f2 limits and amounts of drug released for each of the prescribed sampling time. Applying kinetic equation models, the mechanism of release of the drug from formulation F6 was found to follow Higuchi model, as the plots showed high linearity, with correlation coefficient (r) value of 0.95. In the bioequivalence studies, all the pharmacokinetic parameters obtained with formulation F and the innovator product were not significantly different and were found to be bioequivalent. **Conclusion:** The resulting formulation tablet produced complied within the specified technical limits as to content uniformity, weight variation, thickness, friability, disintegration and hardness. A positive correlation existed between the *in vivo* bioavailability and *in vitro* dissolution tests. Results of the present study indicated the suitability of CG in the preparation of sustained release formulation of clarithromycin.

*Keywords***:** Cashew Gum, Clarithromycin, Sustained-Release Tablet, Bioequivalence, *In vitro – in vivo* Correlational

## Introduction

The continuous search for hydrocolloids from plants paved the way for the discovery of cashew gum (CG) in the last decade

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due to the pantropical distribution of the cashew plant *Anacardium occidentale* L. Ofori-Kwakye et al.(2010) have demonstrated the properties of CG as a binder and coater in the development of Metronidazole tablets. De Paula et al. (1998) and Mothe et al. (2008) characterized the monosaccharide components of CG after acid hydrolysis, yielding galactose, rhamnose, arabinose and glucose. Hydrolytic products were later confirmed by Mothe and De Freitas (2013) by gas chromatography, including confirmation of functional groups by FTIR and $C^{13}$ NMR and decomposition monitoring by thermal analysis.

Okoye et al. (2012) differentiated CG derived from the Nigerian, Ghanaian and Brazilian varieties of *A. occidentale* by solid-state analyses using different spectroscopic and thermal methods. There is a trend encouraging drug manufacturers to substitute acacia gum with CG to reduce dependence on acacia, tragacanth and xanthan gums as excipients during formulation development and allow for the massive propagation of *A. occidentale* in countries that produce it in abundance.

Clarithromycin is a third-generation macrolide antibiotic that has, for the past decades, revolutionized oral antibacterial therapy for the eradication of peptic ulcer caused by Helicobacter pylori as well as bacterial infections of soft tissues, genito-urinary, gastro-intestinal, lower and upper respiratory tracts. However, patient compliance and antibiotic resistance are underlying problems that necessitate the development of oral sustained-release tablet formulations offering once daily oral dosing. Several attempts have been made to formulate clarithromycin into sustained-release tablets, including the fluid bed granulation process by Packiaraj et al. (2013), the direct compression technique of Mahalingan et al. (2009) and the floating matrix of Ravi et al. (2012). Furthermore, the high cost of the current innovator formulation for clarithromycin sustained-release tablets does not provide options for patients who seek less expensive but innovative products.

This study explores the possibility of utilizing CG as a hydrophilic colloidal polymer that will provide a sustained-release mechanism from a tablet formulation of clarithromycin which was optimized from the methods of the previously mentioned studies to determine if it satisfies similar *in vitro* release data as the innovator product. *In vitro* multi-point dissolution test and bio-equivalence studies were conducted to allow for a point-by-point (Level A) *in vitro – in vivo* correlational (IVIVC) analysis for both test formulation and innovator product. Good IVIV correlations can be utilized to predict bio-availability based on in vitro dissolution data, allows dosage form optimization with the fewest possible pharmacokinetic trials in humans, fix dissolution acceptance criteria and can be used as a surrogate for further bio-equivalence studies (Emami, 2006).

## Materials and methods

### Harvesting of exudates and preparation of cashew gum

Circular incisions by rubber tapping were made to injure the bark of 3 mature trees of *Anacardium occidentale* ("cashew") located at Mangaldan, Pangasinan. Herbarium specimens of the plant were submitted to the Philippine National Herbarium for authentication and were given a group accession number of 2135872. After 3 to 5 days, about 1 kg of the yellowish-brown exudates appearing in the incisions were removed and pulverized using a Wiley mill. The powdered exudates were made to swell in 1 liter of distilled water for 4 hours and then filtered. The resulting mucilage was added to 3 liters of acetone and then allowed to stand for 24 hours. The supernatant liquid was decanted and the white precipitate was spray dried. The resulting powdered was resuspended in 400 mL of distilled water, further precipitated with 750 mL of acetone and then spray dried after decantation to yield a total of 567 grams of (56.7 w/v %) of cashew gum (CG)

## Materials

Clarithromycin was provided by Deackchem, Inc. as a sample product. Talc and magnesium stearate were gifts from Medirare Trading Co. Microcrystalline cellulose (MCC), sodium dihydrogen phosphate dihydrate (SDPD) and polyethylene glycol 6000 (PEG 6000) were obtained from Dakila Trading, Inc. Hydroxypropylmethylcellulose 6 centipoise (HPMC 6 cps) and hypromellose 15 cps were requisitioned from Sigma-Aldrich, Inc. Klaricid 500 mg OD tablets (i.e., the innovator product from Abbott Lab., Inc.), bearing a lot number of 6055437 and expiring on February of 2018, was purchased from Mercury Drug, Inc.

## Tablet formulation

The 6 formulations (A to F) in Table 1 was patterned after the clarithromycin extended-release tablets designed by Mahalingan et al. (2009), taking into consideration the same ratio by which raw materials were compressed into the finished products and substituting HPMC K4M and HPMC K5M with CG, in the presence of HPMC 6 cps and hypromellose 15 cps, as the viscosity-building hydrophilic colloidal polymers. For the granulation, clarithromycin was mixed with an aqueous mixture of MCC, PEG 6000, HPMC 6 cps and hypromellose 15 cps. SDPD was added and the

**Table 1**. Test formulations of clarithromycin 500 mg sustained-release tablet

| Ingredients | Formulation (Amount in mg) | | | | | |
|---|---|---|---|---|---|---|
| | **A** | **B** | **C** | **D** | **E** | **F** |
| Clarithromycin | 500 | 500 | 500 | 500 | 500 | 500 |
| Cashew gum | 25 | 50 | 75 | 100 | 125 | 150 |
| HPMC 6 cps | 150 | 125 | 100 | 75 | 50 | 25 |
| Hypromellose 15 cps | 25 | 25 | 25 | 25 | 25 | 25 |
| Microcrystalline cellulose | 75 | 75 | 75 | 75 | 75 | 75 |
| Na dihydrogen phosphate dihydrate | 65 | 65 | 65 | 65 | 65 | 65 |
| PEG 6000 | 50 | 50 | 50 | 50 | 50 | 50 |
| Talc | 5 | 5 | . 5 | 5 | 5 | 5 |
| Magnesium stearate | 5 | 5 | 5 | 5 | 5 | 5 |
| Total | 900 | 900 | 900 | 900 | 900 | 900 |

wet mass was spray-dried at 60°C for 20 minutes and allowed to passed through sieve # 10. The granules were further refined by passing through sieve # 20 followed by lubrication with talc and magnesium stearate for 2 hours. The resulting tablet blend was tested for derived properties of powders and exhibited the following characteristics: a 1.15% w/v moisture content, bulk and tapped densities of 0.54 and 0.43, respectively, a 25% compressibility index, a Haussner ratio of 1.31 and an angle of repose of $33^0$ according to the method modified by Adeleke et al. (2012). The granules were pressed into tablets by direct compression using a Manesty 201C tableting machine.

## Excipient compatibility testing

Three samples, accurately weighed, were triturated in a mortar, namely: (1) 500 mg of clarithromycin; (2) 500 mg of clarithromycin + 500 mg of CG; and (3) 500 mg of the resulting tablet blend or granules obtained prior to compression. One gram portions of each sample was heated in a hermetic aluminum pan at 5°C per minute, between -70 and 100°C, using a Shimadzu DTA-50 thermal analyzer. Each sample was prepared as a KBr disc and scanned in a Perkin-Elmer 665 FTIR spectrometer.

## Formulation optimization by dissolution profiling

Clarithromycin release rates for formulations A to F and the innovator product (n = 6 each) were distributed by random into each of the 6 vessels of a USP Type 2 dissolution tester, with the paddles rotated at 50 rpm using 900 mL of 0.1 N HCl maintained at 37 ± 0.5°C as medium. Samples of 2 mL were withdrawn after 2, 4, 7.5 and 12 hours. Each sample withdrawn was immediately replaced with 2-mL portions of fresh medium. Samples were filtered (Millipore No. 1) and assayed for clarithromycin content using a Genesys 342 spectrophotometer from the calibration curve of clarithromycin USP reference standard.

## *In vitro* multipoint dissolution test

Similar dissolution experiment was performed using the optimized formulation and the innovator product (n = 6 each). This time, samples were withdrawn after 5, 10, 15, 20, 25, 30, 40, 50 and 60 minutes and 1.25, 1.67, 2, 2.5, 3, 4, 5, 8, 10, 12, 15, 18, 21 and 24 hours.

## Finish product analyses

Dosage units of the optimized formulation, and the innovator product were subjected to the following tests: weight variation, thickness, friability, disintegration time and hardness.

## Content uniformity

One tablet of the optimized formulation, powdered and accurately weighed, were transferred to a 500 mL volumetric flask containing 350 mL of methanol. The contents were mixed for 30 minutes, diluted to volume with methanol, sonicated for another 30 minutes and allowed to stand and settle for 16 hours. Exactly 3 mL of the supernatant was diluted to 100 mL with 0.067 M $KH_2PO_4$ (pH 4.0). Exactly 50 μL of this solution and a standard solution containing 125 mcg/mL of clarithromycin USP reference standard in 0.067 M $KH_2PO_4$ (pH 4.0) were injected separately into an HPLC Shimadzu 260-C system equipped with a Luna Phenomenex 4.6 mm x 15 mm C-18 column heated at $50^0$C and a 210 nm UV detector at a flow rate of 1 mL/minute using chloroform-acetonitrile-acetic acid (2:2:1) as mobile phase. The amount in mg of clarithromycin per tablet is taken from the formula: (50/3) (C/N) ($R_u$ /$R_s$), where C = 125 mcg/mL, N = 1 tablet taken for the assay, while $R_u$ and $R_s$ are the clarithromycin peak response obtained for the assay and standard preparations, respectively (Ghari et al., 2013). This test was performed individually for 10 dosage units of the optimized formulation.

## Accelerated stability study

Dosage units of the optimized formulation were individually blister-packed in aluminum foils and stored at 37 ± $2^0$C, 45 ± $2^0$C and 55 ± $2^0$C at 75% RH for 6 to 12 months. Samples were withdrawn at 2-month intervals and assayed for clarithromycin content by HPLC (Ghari et al., 2013).

## Bio-equivalence study

### *Study design*

The study was conducted in a private ward at a 300-bed capacity tertiary-level private hospital at San Carlos City, Pangasinan, Philippines. It is a prospective, randomized open-label 2-way cross-over study involving 24 healthy male Filipino volunteers. The volunteers received 500 mg single dose of either the optimized formulation (A) or the innovator product (B) in 2 separate sessions. During the first session (days 1 and 2), volunteers were randomly divided into 2 groups of 12 volunteers each, such that groups 1 and 2 received A and B, respectively. On the second session (days 10 and 11), after a washout period of 7 days, groups 1 and 2 received formulations B and A, respectively. Each volunteer received the treatment with 250 mL of distilled water in the morning after overnight fasting. Volunteers were fed a standard meal 4 hour after dosing. In each of the 2 sessions, volunteers were monitored closely for adverse reactions (Kumar and Sanjita, 2013).

### *Subjects*

The volunteers, ranging from 18 to 32 years old, have a mean age of 29.5 ± 2.3 years, a mean body weight of 70.2 ± 5.6 kg,

a mean height of $1.72 \pm 0.05$ m and a mean BMI of $23.14 \pm 1.4$. Healthy subjects were selected after undergoing to medical history, physical examination, chest X-ray, ECG, hematological/serological screenings, biochemical profiling, urinalysis and fecalysis. Exclusion criteria included any history of gastro-intestinal and hepato-biliary diseases that may affect the pharmacokinetics of clarithromycin, chain smokers, any history of drug allergies, severe alcoholism, blood donation within 30 days and intake of any medications or food supplements 7 days prior to the first session of the study. Volunteers were strictly abstained from smoking, coffee, alcohol, soda, junk foods, medications and commercial beverages. Volunteers with any incidences of vomiting and other serious adverse drug reactions throughout the duration of the study were withdrawn. An informed consent was signed by each volunteer, giving information as to possible risks, side-effects, benefits, procedures and objectives of the study in addition to their rights as volunteers according to the Declaration of Helsinki and the ICH Guidelines with the study protocol reviewed and approved by the Ethics Committee of the Institutional Review Board.

**Blood sample collection**

Venous blood samples were collected from a common catheter inserted into the right forearm of each volunteer, such that this catheter remained attached for the entire duration of the blood sampling time per study session. Five mL of blood samples were collected into heparinized tubes at 5, 10, 15, 30, 40 and 60 minutes and 2, 3, 5, 7.5, 10, 12, 15, 18, 24, 36 and 48 hours after dosing. Blood samples were centrifuged at 3000 rpm for 20 minutes to isolate the plasma. Plasma samples were stored at -20°C until ready for analysis.

**Validation of the bioanalytical method**

A fast and simple reversed phase binary HPLC method (Shimadzu 260-C system) was adopted to quantify clarithromycin in plasma samples using chloroform-acetonitrile-acetic acid (2:2:1) as mobile phase. A Luna Phenomenex 4.6 mm x 15 mm C-18 column heated at $50^0$C was used and clarithromycin was quantified using a 210 nm UV detector at a flow rate of 1 mL/minute to induce separation at a retention time at 8.2 minutes. Exactly 50 µL of plasma samples diluted 1:5 with the mobile phase were injected. This method was validated in terms of accuracy, precision, limits of detection and quantification, specificity and linearity. The interday and intraday precision of quality control samples showed less than 3.5% coefficients of variation. The calibration curve registered a linearity of 0.9978 (Niopas and Daftsios, 2001).

**Statistical data analyses**

Means ± CV are compared by the 2-way analysis of variance and 90% confidence interval where $p < 0.05$ is considered significant. The shelf-life of the test formulation during the accelerated stability was calculated using the method of least square for the first-order Arrhenius equation. The area under the curve from zero to 48 hours ($AUC_{0-48}$) was computed by the trapezoidal rule. The AUC to infinity ($AUC_{0-\infty}$ ) was computed, thus: $AUC_{0-\infty} = AUC_{0-48} + C_{48}/K_{el}$, where $C_{48}$ is the last quantified plasma concentration after 48 hours and $k_{el}$ is the elimination rate constant which was computed from the terminal slope of the $AUC_{0-48}$ where the semilogarithmic values of the plasma concentration were plotted against time. The time to maximum concentration ($T_{max}$) was computed, thus: $T_{max} = 2.3 \log (k_a/k) / k_a – k$, where $k_a$ is the absorption rate constant determined by the method of residuals from the terminal slope of the semilogarithmic plot of elimination slope of $AUC_{0-48}$. The fraction of drug absorbed per time $Ab/Ab\infty$ was computed by the Wagner-Nelson method, thus: $Ab/Ab\infty = C_p + k_{el} [AUC_{0-t}] / k [AUC_{0-\infty}]$, where $C_p$ is plasma concentration at time t (Gohel et al., 2005).

Dissolution profiles between the innovator and the sample formulations are compared by the similarity factor $f_2$ which is computed, thus: $f_2 = 50 \log [\{1 + 1/n \sum (R_t - T)2\}_t^{-0.5} \times 100]$, where Rt and Tt are the percentage of drug dissolved Q at each time point for the innovator and sample formulations, respectively, n is the number of dissolution sampling time and t is the time point for collecting dissolution samples. Furthermore, the dissimilarity factor $f_1$ is computed, thus: $f_1 = [\{\sum | R_t - T_t | / \sum R_t\} \times 100$. The in vitro mean dissolution time MDT is computed, thus: $MDT = \sum T_{mid} \Delta M / \sum \Delta M$, where $T_{mid}$ is the midpoint between i and i-1 and $\Delta M$ is the additional amount of drug dissolved between i and i-1.

To analyze the mechanism of dissolution of clarithromycin, the following kinetic models were applied: zero order, where cumulative concentration Q was plotted against time t; (2) first order, where log Q was plotted against time; (3) Higuchi release, where Q was plotted against square root of time; (4) Hixson-Crowell's cube root law, where $Mo^{1/3} – Mt^{1/3}$ was plotted against t such that Mo is the initial amount of the drug in the sample (~ 500 mg) and Mt is the cumulative amount of drug (in mg) released at t; and (5) Korsmeyer-Peppa's model, where log Q was plotted against log t.

**Results and discussion**

**Compatibility testing**

Based on the differential thermal analyses in Figure 1 clarithromycin and its tablet excipients showed characteristic melting trends without any significant changes in the endotherms and curvatures of individual thermogram to imply that the excipients selected can be combined with

clarithromycin as no physico-chemical incompatibility problems are evident. The drug-excipient compatibility represents an important phase in the pre-formulation stages for the development of the CG-based clarithromycin sustained release tablets as it can influence the dissolution profile of the drug.
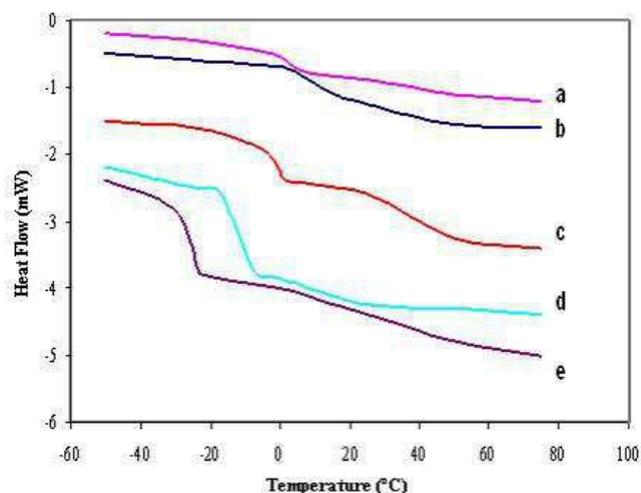


**Figure 1.** Comparative thermograms of (a) clarithromycin, (b) cashew gum, (c) clarithromycin + cashew gum, and (d/e) tablet blend of clarithromycin

In figure 2, FTIR analysis indicates the spectra showing characteristic peaks of clarithromycin and the tablet blend. Clarithromycin appears to be compatible with its excipients due to the absence of new absorption bands, including alteration of their intensities that may correspond to additional organic functional groups associated with the formation of degradation products, indicating the absence physicochemical incompatibilities. The result of FTIR corresponds to the DTA analysis, thus making it feasible for the development of a stable CG-based clarithromycin sustained release tablets.
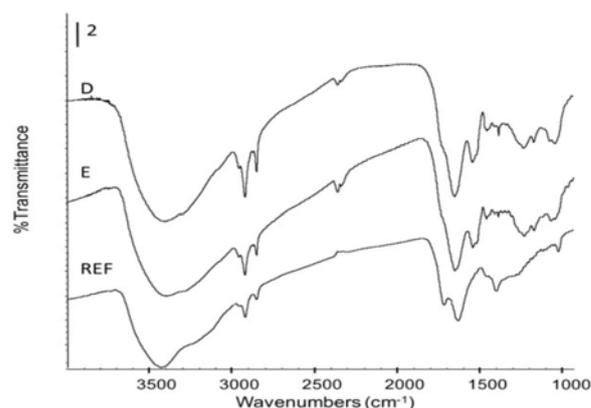


**Figure 2.** FTIR spectra of (D) clarithromycin, (E) clarithromycin + CG, and (REF) tablet blend of clarithromycin

**Dissolution test profiles**

Table 2 compares the drug release profiles of the 6 test formulations after 2, 4, 7.5 and 12 hours of dissolution as specified in the USP-NF 34/29. Several studies dealing with the sustained-release of drugs from tablets combine different techniques in granulation during optimization (Packiaraj et al., 2013; Mahalingan et al., 2009). In this study, suitable combination ratios of CG/HPMC polymers were determined to control clarithromycin release from the test formulations. Target release profile was generated for a 12-hour dosage regimen and dissolution profiles of the different formulations were compared based on compendia requirements for the f1 and f2 limits and amounts of drug released for each of the prescribe sampling time. Results showed that the dissolution profile of formulation F has the in vitro release patterns similar to that of the innovator product. Similarly, formulation F satisfied all USP limits as manifested in the in vitro multipoint dissolution profile (Figure 3) for f1 and f2 values with no significant differences as to the innovator product. These can be attributed to the increase in the ratio of CG and decrease in

**Table 2.** Comparative dissolution profiles of various test formulations of clarithromycin 500mg sustained-release tablet

| Sampling Time in Hours | Mean % Q for Test Formulation | | | | | | Innovator Product |
|---|---|---|---|---|---|---|---|
| | **A** | **B** | **C** | **D** | **E** | **F** | |
| 2 (nmt 25%*) | 3.66 ± 0.85 | 4.55 ± 1.24 | 8.33 ± 2.77 | 12.47 ± 2.98 | 21.3 ± 2.5 | 19.42 ± 2.13 | 21.6 ± 1.87 |
| 4 (20 - 40%*) | 15.64 ± 1.22 | 13.44 ± 2.65 | 17.93 ± 2.11 | 25.18 ± 2.75 | 28.9 ± 3.2 | 33.27 ± 2.3 | 31.5 ± 4.1 |
| 7.5 (45 - 75%*) | 28.67 ± 4.12 | 33.82 ± 3.71 | 36.75 ± 4.19 | 48.21 ± 3.98 | 66.3 ± 5.1 | 67.11 ± 3.4 | 72.3 ± 4.8 |
| 12 (nlt 80%*) | 77.3 ± 5.5 | 66.77 ± 4.76 | 65.34 ± 5.79 | 72.11 ± 4.85 | 77.3 ± 6.6 | 86.39 ± 5.6 | 87.4 ± 4.3 |
| f1 (nmt 15%) | 18.23% | 17.91% | 19.78% | 21.15% | 14.65% | 9.33% | --- |
| f2 (nlt 50%) | 41.47% | 39.68% | 44.76% | 43.25% | 47.88% | 61.36% | --- |

*USP-NF 34/29; n = 6 per formulation

HPMC which provides the desired drug-release. According to the study of Ofori-Kwakye, K. et al (2010), while 4 – 8 % of CG delivers immediate drug release, higher concentrations exhibit greater viscosity for CG and, thus, may promote sustained drug release.
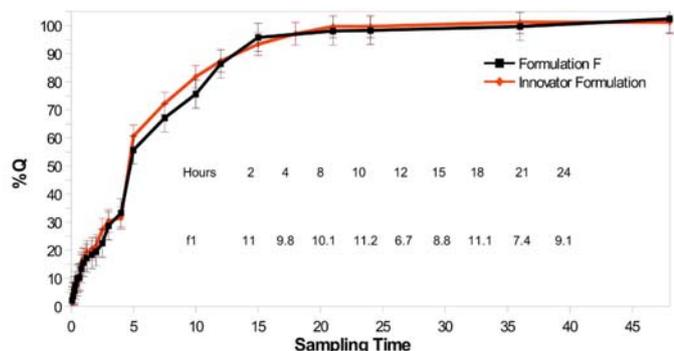


**Figure 3.** Comparative *in vitro* multipoint dissolution studies between the optimized and innovator formulations

### Release kinetics and mechanism between the optimized and innovator formulations

The results of the kinetic models used in the assessment of the dissolution data are summarized in Table 3. Regression values (r) were used to assess the best fit so that the higher the r value the better the fit of the dissolution profile to that kinetic model. Higher r values were obtained for the Higuchi model than the other kinetic models. This occurred in both the optimized and innovator formulations. Higuchi describes that drug release as a diffusion process based on the Fick's law is dependent on square root of time. This model has been used to describe drug dissolution from several types of modified release pharmaceutical

**Table 3.** The drug release kinetics of optimized and innovator formulations

| R values | Kinetic Models | | | | | |
|---|---|---|---|---|---|---|
| | Zero-Order | First-Order | Higuchi | Hixson-Crowell | Korsemeyer-Peppa | n value |
| **Formulation F** | 0.67 | 0.76 | 0.95 | 0.82 | 0.87 | 0.51 |
| **Innovator Formulation** | 0.63 | 0.69 | 0.93 | 0.74 | 0.91 | 0.76 |

**Table 4.** Correlation between release exponent (n) values for the Korsemeyer–Peppas equation and drug release mechanisms depending on the geometry shape

| Release exponent (n) | | | Release mechanism |
|---|---|---|---|
| Thin film | Cylindrical | Sphere | |
| 0.5 | 0.45 | 0.43 | Fickian diffusion |
| 0.5 < n < 1.0 | 0.45 < n < 0.89 | 0.43 < n < 0.85 | Anomalous transport |
| 1.0 | 0.89 | 0.85 | Case II transport |

dosage forms like transdermal systems and matrix tablets with water soluble drugs. The dissolution data was fitted into the Korsemeyer-Peppas equation to determine the exact mechanism of drug release. The drug release data gave 'n' values between 0.45 and 0.89 (Table 4). Drug release from the matrix tablets follows anomalous transport. This means drug release involves swelling of the matrix tablets and subsequent erosion which occurs simultaneously and, thus, contributing to the overall drug release rate.

### Bioequivalence and pharmacokinetic profile

Plasma concentration-time profile is highly affected by the performance of the dosage formulation. Despite the similarity provided by usually employed test dissolution methodology, bioequivalence between the innovator and test formulations may still vary. In this study, in vitro dissolution study of formulation F conforms to the in vivo bioequivalence test (Figure 4), suggesting that cashew gum facilitates the bioavailability of clarithromycin. The pharmacokinetic and bioequivalent values (Table 5) obtained with formulation F and the innovator product were not significantly different and were all within the FDA bioequivalence acceptance criteria of 80-125% in the two-sided 90% confidence interval. This reflects that formulation F containing CG exhibit pharmacokinetic parameters comparable to the innovator product and thus can be used as a generic substitute.

### Quality control tests

Table 6 presents the results of in-process quality control tests performed on both optimized formulation and innovator product. Formulation F complied within the specified technical specifications and manifested no considerable differences as compared to the innovator product in terms of
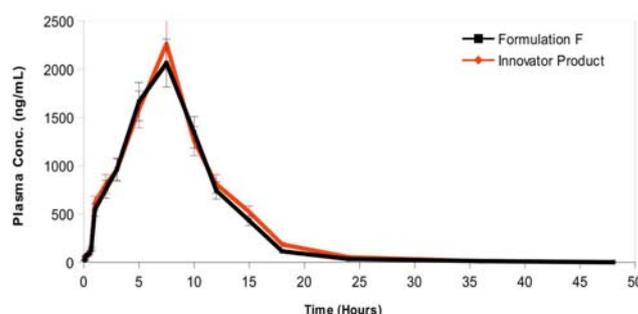


**Figure 4.** Comparative areas under the curve between the optimized formulation and the innovator product of clarithromycin 500mg sr tablet

**Table 5.** Comparative pharmacokinetics derived from bio-equivalence study between the optimized formulation and innovator product of clarithromycin 500mg SR tablet

| Parameter | Formulation F | Innovator Product | P Values | 90% Confidence Interval (80 - 125%) |
|---|---|---|---|---|
| $AUC_{0-t}$ (ng-hr/mL) | 28,751 ± 7,534 | 27,833 ± 6,345 | < 0.001 | 93.5 – 112.5 |
| $AUC_{0-\infty}$ (ng-hr/mL) | 31,453 ± 9,236 | 30,870 ± 8,453 | < 0.001 | 88.4 – 106.4 |
| $C_{max}$ (ng/mL) | 2,065 ± 413 | 2265 ± 313 | < 0.001 | 95.1 – 111.6 |
| $T_{max}$ (hr) | 7.45 ± 2.34 | 7.77 ± 2.14 | < 0.001 | 93.9 – 115.4 |
| $K_a$ ($hr^{-1}$) | 0.4253 ± 0.105 | 0.4587 ± 0.157 | < 0.01 | 92.4 – 117.5 |
| $K_{el}$ ($hr^{-1}$) | 0.1155 ± 0.034 | 0.134 ± 0.075 | < 0.01 | 84.6 – 110.4 |
| $T_{1/2}$ (hr) | 6.05 ± 1.67 | 6.18 ± 2.18 | < 0.01 | 85.7 – 112.5 |
| Cl (L/hr) | 0.05 ± 0.01 | 0.06 ± 0.02 | < 0.001 | 89.5 – 107.4 |

**Table 6.** Quality control tests for the optimized formulation and innovator product of Clarithromycin 500mg sustained-release tablet

| Parameter | Technical Specification | Method | Results as Mean ± SD | |
|---|---|---|---|---|
| | | | Formulation F | Innovator Product* |
| Content uniformity | Mean LC ± 3% CV n = 20 | HPLC (see 2.6) | 502.4 mg ± 2.12% | 501.2 mg ± 1.98% |
| Weight variation | Mean ± 3 SD n = 20 | Sartorius 1150 | 979.2 mg < 1SD | 945.6 mg < 1.2 SD |
| Thickness | Mean ± 3 SD n = 10 | Digital Vernier caliper | 8.45 mm < 1.3SD | 8.21 mm < 1.2 SD |
| Friability | Nmt 1% n = 10 | Roche 258 | 0.24% ± 0.08 | 0.31% ± 0.05 |
| Disintegration | Within 1 hour n = 6 | Basket/SGF $37^0$C | 45 ± 4.4 mins. | 39 ± 5.2 |
| Hardness | Nlt 4 kg n = 10 | Pfizer HT | 5.21 ± 0.25 kg | 5.33 ± 0.19 kg |

SGF = simulated gastric fluid; SD = standard deviation; LC = % label claim; Innovator Product = Klaricid 500mg OD Tab. (Abbott Laboratories, Inc.)

the different test parameters. This is similar to a study conducted by Ofori-Kwakye et al. (2013) where the physicochemical properties of the optimized formulation (diclofenac sodium + CG) were found to be within the prescribed limits. This can be attributed to efficient blending of the hydrophilic polymers with CG to show improvement in the physicochemical and release retarding properties of the optimized sustained-release product. Thus, test results are indicative that the CG can be used as matrix carriers for sustained drug release delivery without presenting bioavailability problems.

### *In vivo – in vitro* correlation between the test formulation and innovator drug

Clarithromycin exhibits poor aqueous solubility that limits its absorption and is, thus, classified under drugs belonging to Biopharmaceutics Classification System (BCS) Class II. Manani et al. (2017) reported that clarithromycin undergoes rapid degradation under conditions of low pH that exist in gastric fluid. Since its systemic bioavailability is dependent on gastric stability and intestinal absorption, protection from acid degradation may play a key role in enhancing its bioavailability.

Quality problems associated with generic products of clarithromycin have been reported in the past (Nightingale, 2005), with a significant percentage of generic products failing to meet pharmacopoeia specifications for assay and dissolution, as well as failing the test for equivalence to innovator products. However, in figure 5, not only in the *in-vitro* testing did the optimized formulation provide similar drug release characteristics as the innovator product but even under pathophysiological or in vivo conditions. The results show that a positive correlation exists between the in vivo bioavailability and in vitro dissolution tests and no significant difference between the optimized and innovator formulations were observed.

### Conclusion

Sustained release clarithromycin matrix tablets designed for oral administration were formulated using various blends of CG and HPMC. Formulation F showed the requisite *in vitro* sustained drug release, as well as difference and similarity properties, and were found to be comparable with
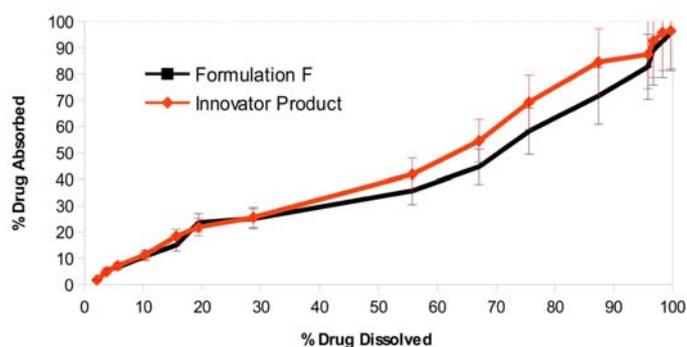
**Figure 5.** Level A *in vivo – in vitro* correlation between the optimized formulation and innovator product of clarithromycin 500mg SR tablet

the innovator product in terms of pharmacokinetic and bioavailability profiles. The study has demonstrated that the blending of a suitable ratio of CG and hydrophilic polymers improves the overall sustained release properties leading to the formation of an optimized formulation suitable for oral administration.

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