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# Effect of some surfactants on the release of metronidazole from suppositories formulated with goat fat and palm kernel oil admixtures

**Anthony A. Attama***

*Department of Pharmaceutics, University of Nigeria, Nsukka 410001, Nigeria.*

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**Abstract**

Metronidazole (MDZ) suppositories were formulated by pour moulding using goat fat (GF)–palm kernel oil (PKO) admixture (3:1) as the base. The effect of three surfactants – cationic (cetrimide), anionic (sodium laurylsulphate) and non-ionic (Tween 65) was evaluated. The mechanism of release of the active ingredient from the suppositories was equally investigated. It was found that Tween 65 (T-65) and sodium laurylsulphate (SLS) improved the release of MDZ significantly ($p>0.05$) at all the concentrations used. Cetrimide (CTM) did not improve the release of MDZ at all the concentrations used. It was also found that release of MDZ from the suppositories was largely governed by diffusion. Statistical analysis ($p>0.05$) of the results showed T-65 performed better than SLS in enhancing the release of MDZ from the suppositories, and could be used to enhance the release of MDZ from suppositories formulated with this novel suppository base.

*Keywords*: Surfactants; Goat fat; Palm kernel oil; Metronidazole; Suppositories; Drug release

## Introduction

A wide range of materials has been used as suppository bases including fractionated palm kernel oil, theobroma oil, glycerol gelatin, the polyethylene glycols and surfactants. Suppository bases have been combined to further improve on the gains derivable from a single base. In this regard, theobroma oil–other fat combinations, polyethylene glycol–silica and polyethylene glycol-polysorbate are some of the combinations that have been employed in formulation (Kitagawa *et al.*, 1987). Drug release rate from suppositories is mainly conditioned by excipient characteristics, temperature, fusion rate, viscosity and hydro-lipophilic characteristic of the active (Bornschein *et al*., 1980). A lot of auxiliary agents such as glycerides, silica gel, insoluble powders, carbomers, cellulose derivatives and surfactants have been incorporated into lipophilic suppository bases to enhance the release of incorporated drug (Baichwal and Lohit, 1970; Dal Zotto *et al.*, 1991). Different effects were observed with these agents. It has been shown that absorption from suppositories is dependent on the nature of the suppository base or other additives and the solubility of the drug in the suppository base (Schmitt and Guentert, 1990), and that surfactants possess a high potential to promote absorption across rectal mucosa

* Corresponding author. *E-mail address*: aaattama@yahoo.com Tel: +234 (0) 8036618204

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(Nishihata *et al.*, 1980). Some workers have investigated the possibility of using some additives to prolong or modulate the release of drugs from suppositories (Hosny *et al.*, 1996; Realdon *et al.*, 1997; Choi *et al.*, 1998). It was the objective of this work to investigate the effect of some surfactants on the release of MDZ from suppositories formulated with GF-PKO admixture. GF is extracted from the adipose tissue of *Capra hircus*. Purified GF has a solid consistency and melts at 51 $^{o}$C making it imperative for the addition of an agent that will lower the melting point to the range acceptable for suppositories that melt before releasing the incorporated drug. MDZ is an anti-amoebic drug that has been formulated into suppositories and tablets with different lipid matrices (Lund, 1994; Attama *et al.*, 2004; Ofoefule *et al.*, 2004; Özyazici *et al.*, 2006). In all the bases used for suppository formulation, theobroma oil was ranked the least in terms of the release studies carried out. It is thus necessary to develop a cheap base or modify the existing bases for the formulation of MDZ suppositories that would release maximally, the incorporated drug. The base used in this study has been evaluated for use in the formulation of salicylic acid suppositories (Attama *et al.*, 2000), but the effect of additives on the release behaviour of the active was not studied.

## Experimental

*Materials.* Palm kernel oil, PKO (Golden Oil), cetrimide (CTM), sodium laurylsulphate (SLS), Tween 65 (T-65), theobroma oil, and glycerin (Merck), activated charcoal and bentonite (BDH) were used as procured from their suppliers. Goat fat (GF) was obtained from a batch processed in our laboratory. All other reagents and solvents were of analytical grade and were used without further purification.

*Extraction of GF*. GF was extracted from the adipose tissues of *Capra hircus*. The fatty mass was collected fresh from the abattoir. The extraneous materials were manually separated from the adipose tissue and thereafter wet-rendered (Young, 1986). Briefly, this involved boiling the adipose tissue in about half its weight of water for 45 min. The molten fat–water mixture was filtered with a muslin cloth and allowed to cool for 24 h at 28 $^{o}$C. The solidified fat was manually separated from the aqueous phase.

*Deodorization and bleaching of GF*. GF possesses a characteristic odour, which usually necessitates deodorization. Deodorization and bleaching were done together by heating the extracted GF at 90 $^{o}$C for 30 min with a combination of activated charcoal and bleaching earth (bentonite) in the ratio of 10:1 per 20 g of GF, accompanied with continuous agitation. At the end of the heating, the hot mixture was filtered through Whatman No. 3 filter paper and the filtrate allowed to solidify at room temperature (Richardson, 1978).

*Determination of melting points of the base combinations.* The melting points of different admixtures of PKO and GF were determined in a melting point apparatus (model MFB-600-010F, Gallenkamp England). The best combination (1:3 ratio of PKO and GF) was selected for further studies.

*Preparation of suppositories.* All the suppositories were prepared to contain 200 mg of MDZ each. The appropriate quantities of the bases (Table 1) were weighed. Pour moulding was used for the production of the suppositories after incorporating the appropriate quantities of the surfactants, as shown in Table 1. Lubrication was done using glycerin and a 1 g mould was used. Prior to removal of the suppositories from the moulds it was cooled at 0 $^{o}$C for 30 min. Theobroma oil was used as the standard suppository base. Eleven batches of suppositories were prepared.

*Evaluation of suppositories.* Appearance: Six suppositories from each batch were selected and cut longitudinally. The internal and external surfaces were examined using a hand lens for uniformity in appearance, presence or absence of air bubbles, brittle fracture, and for the presence of contraction holes.

Weight uniformity: Twenty suppositories were selected from each batch and used for the test. In each case, the suppositories were weighed together using a balance (Sauter, KGD-7470 Germany). The result was analyzed statistically.

Absolute drug content**:** Twenty suppositories were weighed together and the mean weight calculated. They were crushed together and a quantity of the mass equivalent to the mean weight for the particular batch was weighed and melted in 60 ml of phosphate buffer at 37 $^{o}$C, and the volume adjusted to 100 ml with the buffer, and later filtered. The filtrate was diluted appropriately and analysed for MDZ at 277 nm using a spectrophotometer (SP6-450, UV-Vis, Pye Unicam). The absolute drug content was calculated with reference to standard Beer's plot for MDZ. The absolute drug content reported for each batch was the mean of five determinations.

Liquefaction time: A modification of the method described by Setnikar and Fantelli (1962) was used to determine the liquefaction time. One suppository from each batch was wrapped with a dialysing membrane and tied with an inextensible thread. The set-up was suspended in phosphate buffer thermoregulated at 37.0 ± 1.0 $^{o}$C. The suppository was observed carefully and the time taken for the suppository to melt completely was recorded. Average of five determinations was taken as the liquefaction time for each batch.

Release studies**:** The method of Dal Zotto *et al.* (1991) was adopted for this study. A piece of dialysis tube (Visking Tubing, London UK) 10 cm long, 2.5 cm diameter closed at one end, was soaked in distilled water overnight at room temperature. After the addition of 5 ml of phosphate buffer (pH 6.8) the open side was tied with a thread to prevent leakage. The set up was then suspended in a dissolution tester (DTD, Erweka, Germany), containing 500 ml phosphate buffer maintained at a temperature of 37.0 ± 1.0 $^{o}$C and agitation rate of 100 rpm, as the dissolution medium. At predetermined time intervals, 5 ml of the medium was sampled and filtered, and the dissolution medium replaced with a fresh 5 ml portion of phosphate buffer. The filtrate was analysed in the spectrophotometer above at 277 nm.

## Results and Discussion

The melting point of 1:3 blend of PKO and GF was found to be 39.0 ± 0.2 $^{o}$C. This temperature is ideal for suppository formulation using admixtures of PKO and GF considering the melting point of pure GF (51 $^{o}$C) and PKO (31 – 36 $^{o}$C). A suppository formulated with GF alone will not release its active because of the hardness. Similarly a suppository formulated with PKO alone will not withstand tropical temperature. The new base combination with melting point of 39.0 ± 0.2 $^{o}$C may not completely melt in the rectum but will soften, crack or deform due to pressure in the rectal cavity thus releasing its actives. It can also be stored comfortably at room temperature in the tropics.

The internal and external surfaces of the longitudinally dissected suppositories were uniform in appearance. There was no colour change and no air bubbles, no holes or brittle fracture and could withstand the shock of transportation and other mechanical stresses. The result of the weight uniformity test showed that the weight variations in the different batches of the suppositories were not significantly different with each other ($p>0.05$). They had mean weights and coefficients of variation of 1.19 ± 1.76 %; 1.18 ± 2.3 %; 1.19 ± 3.61 %; 1.22 ± 2.94 %,

1.19 ± 2.60 %; 1.21 ± 1.97 %; 1.21 ± 1.32 %, 1.19 ± 4.87 %; 1.24 ± 5.39 %; 1.26 ± 6.48 %; and 1.21 ± 1.81 % for batches 1 – 11 respectively. Weight variation affects drug content and careful control of weight is needed to ensure drug content uniformity. The results of the absolute drug content and liquefaction time are presented in Table 2. The contents of the active ingredient of the suppositories are within the range specified in the British Pharmacopoeia (BP, 1988).

**Table 1.** Quantities* of the ingredients used in suppository formulation

| Batch | PKO:GF (g) | Surfactants (%w/w) | | | Drug (g) |
|---|---|---|---|---|---|
| | | SLS | CTM | T-65 | |
| 1 | 44.1 | - | - | - | 10 |
| 2 | 44.1 | 0.5 | - | - | 10 |
| 3 | 44.1 | 1 | - | - | 10 |
| 4 | 44.1 | 1.5 | - | - | 10 |
| 5 | 44.1 | - | 0.5 | - | 10 |
| 6 | 44.1 | - | 1 | - | 10 |
| 7 | 44.1 | - | 1.5 | - | 10 |
| 8 | 44.1 | - | - | 0.5 | 10 |
| 9 | 44.1 | - | - | 1 | 10 |
| 10 | 44.1(TEO)** | - | - | 1.5 | 10 |

*Quantities presented are for 50 suppositories, **TEO = Theobroma oil

**Table 2.** Result of some physical parameters evaluated

| Batch | ADC* (mg ± SD) | LT** (Min ± SD) |
|---|---|---|
| 1 | 198.2 ± 1.5 | 22.5 ± 1.4 |
| 2 | 195.7 ± 2.8 | 10.0 ±2.2 |
| 3 | 200.2 ± 1.4 | 12.7 ± 2.8 |
| 4 | 210.3 ± 4.1 | 90.5 ± 3.1 |
| 5 | 190.9 ± 2.8 | 20.4 ± 4.2 |
| 6 | 195.7 ± 1.4 | 17.5 ± 2.8 |
| 7 | 205.1 ± 2.5 | 15.3 ± 2.2 |
| 8 | 205.4 ± 1.4 | 15.1 ± 2.8 |
| 9 | 210.7 ± 4.7 | 10.3 ± 2.2 |
| 10 | 211.5 ± 3.1 | 5.2 ± 1.1 |
| 11 | 198.8 ± 2.5 | 11.7 ± 2.1 |

* ADC = Absolute Drug Content, ** LT = Liquefaction Time

**Table 3.** Release kinetic parameters.

| Batch | Release exponent (*n*) | Kinetic constant (*K*) | Regression coefficient ($R^2$) |
|---|---|---|---|
| 1 | 1.1583 | -0.3527 | 0.9846 |
| 2 | 0.9250 | 0.1292 | 0.9733 |
| 3 | 0.8892 | 0.1847 | 0.9746 |
| 4 | 0.7809 | 0.3977 | 0.9721 |
| 5 | 0.9880 | 0.0398 | 0.9913 |
| 6 | 0.9935 | -0.0735 | 0.9135 |
| 7 | 1.9739 | -0.6422 | 0.9765 |
| 8 | 0.9739 | 0.0543 | 0.9897 |
| 9 | 0.7192 | 0.5634 | 0.9905 |
| 10 | 0.7657 | 0.5186 | 0.9825 |
| 11 | 0.6499 | 0.7424 | 0.9846 |

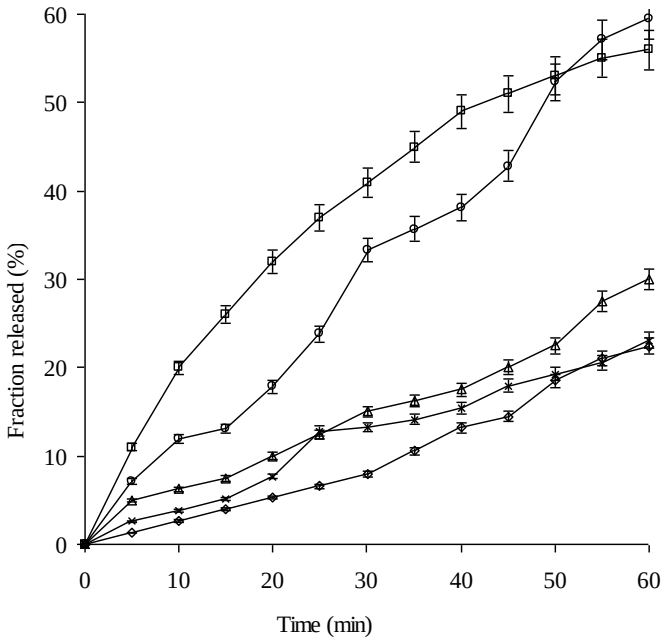


Fig. 1. Release of MDZ from the suppositories containing 10 mg of surfactant per suppository

PKO:GF (1:3) Theobroma oil SLS CTM T-65

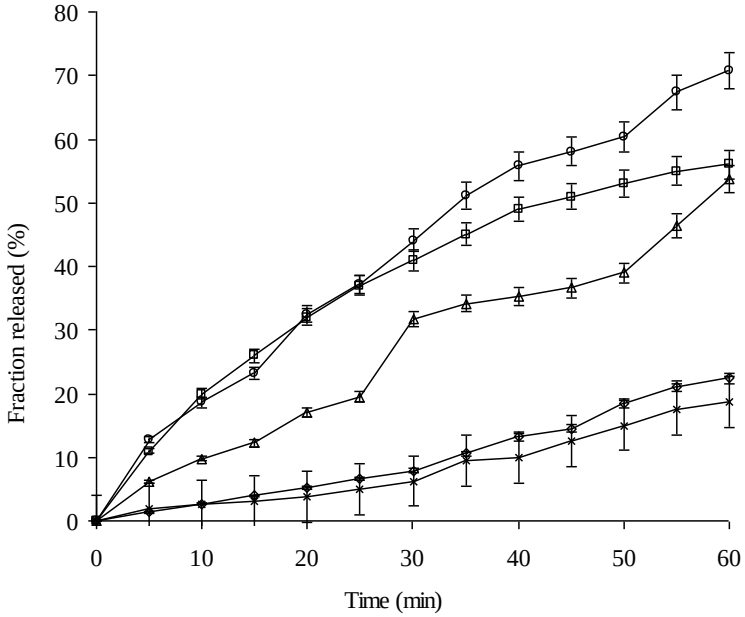


Fig. 2. Release of MDZ from the suppositories containing 20 mg of surfactant per suppository

PKO:GF (1:3) Theobroma oil SLS CTM T-65

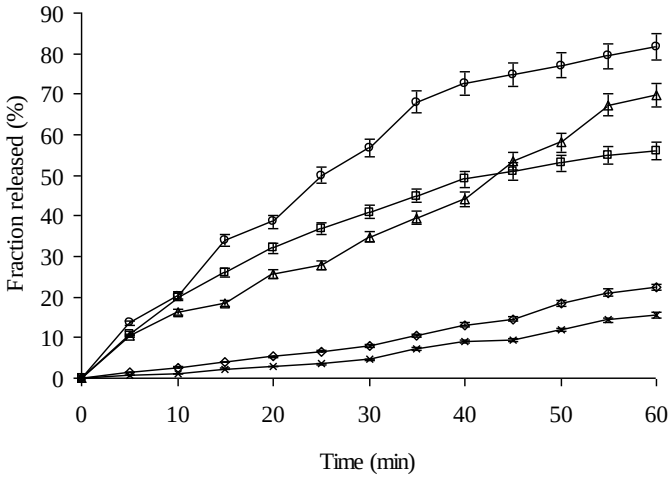


Fig. 3. Release of MDZ from the suppositories containing 30 mg of surfactant per suppository

PKO:GF (1:3) Theobroma oil SLS CTM T-65

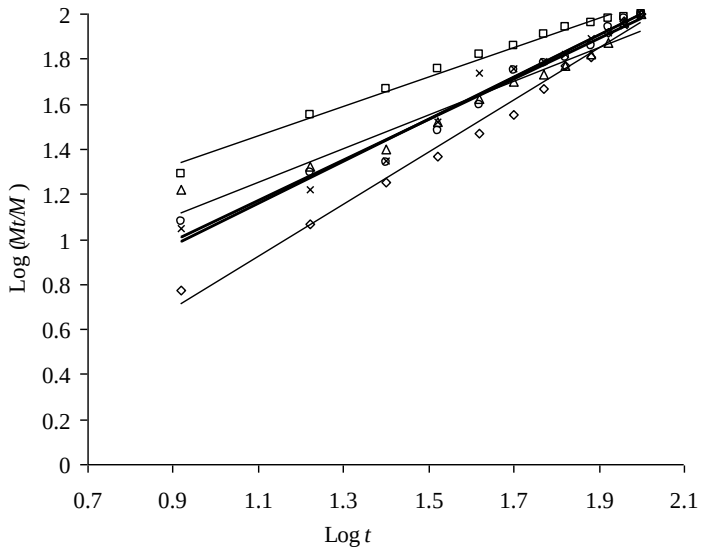


Fig. 4. Log (*Mt/M* ) versus log *t* at 10 mg of surfactant per suppository

◇ PKO:GF (1:3) □ Theobroma oil △ SLS × CTM ○ T-65

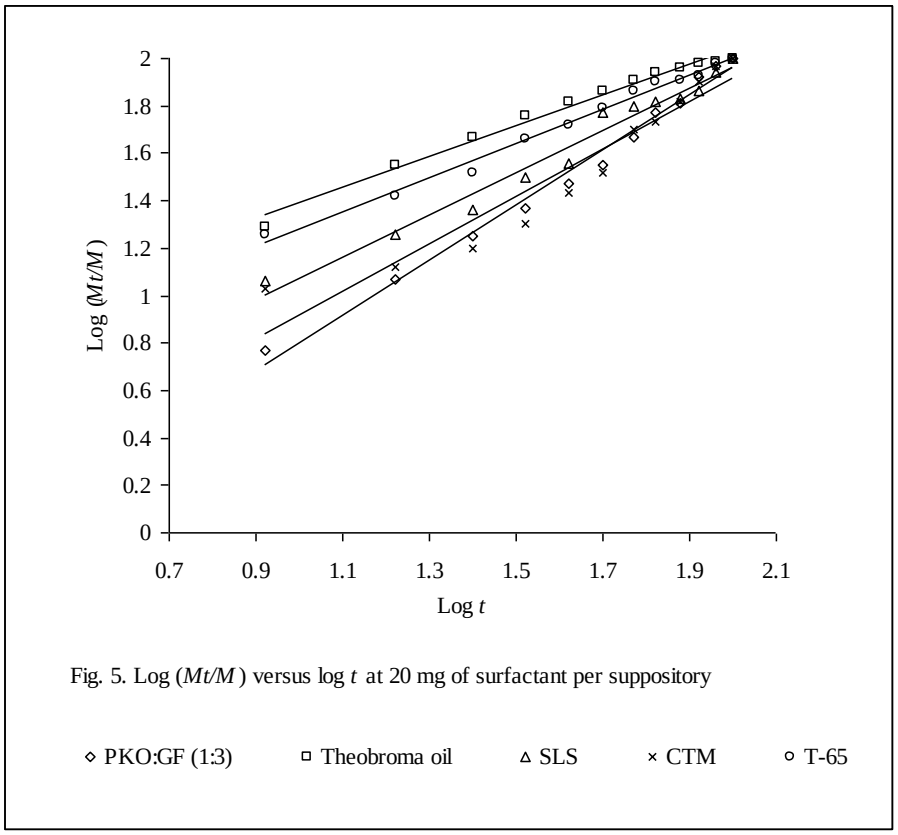


Fig. 5. Log (*Mt/M*) versus log *t* at 20 mg of surfactant per suppository

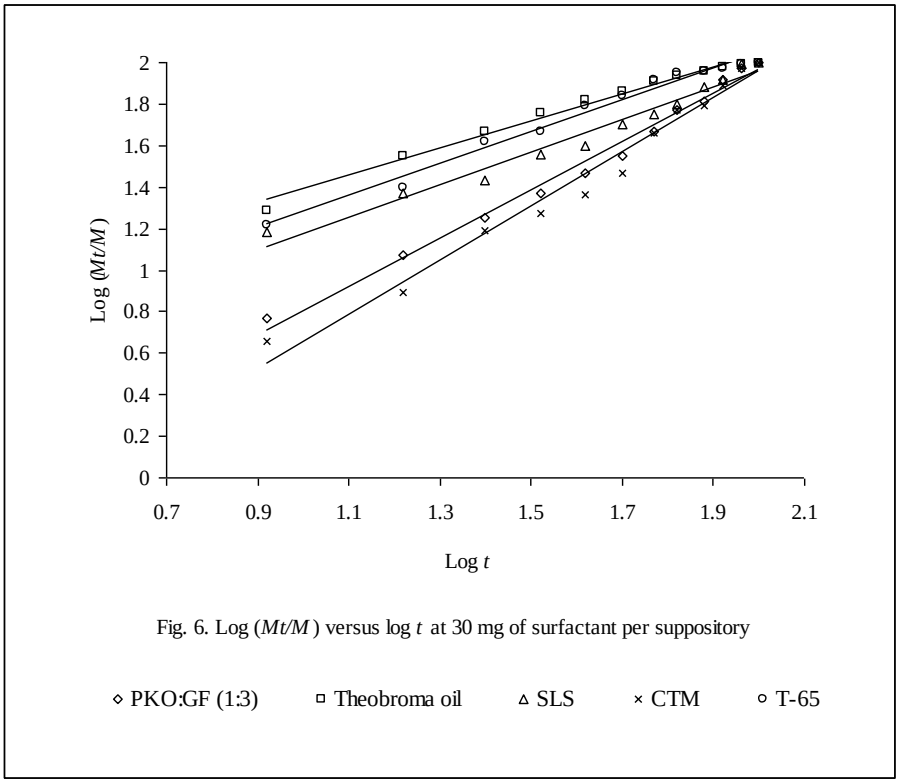


Fig. 6. Log (*Mt/M*) versus log *t* at 30 mg of surfactant per suppository

Variation of active ingredient content is usually due to weight variation and inappropriate mixing or due to sedimentation. However, this only applies to suppositories in which the active ingredient is dispersed in the base. It may also arise due to inadequate drainage of the lubricant in the mould before pouring the mix.

The knowledge of the liquefaction time of suppositories is pertinent for formulations that contain drugs for systemic action, which must be absorbed. A suppository, which takes too long to liquefy, may be expelled together with the drug it includes. From Table 2, it could be inferred that the liquefaction time is inversely related to surfactant concentration. Suppositories containing the three surfactants exhibited shorter liquefaction times than the reference, that is, those containing only PKO and GF without surfactant (Batch 1). The decreased liquefaction time was due to the combined action of heat and emulsification at lipid/fat–aqueous fluid interface. Absence of wetting and emulsification could be the cause of the seemingly high liquefaction time of the suppositories containing no surfactant. A maximum time of 10 min has been suggested for suppositories that are indicated for systemic or general action, which liquefy before complete release and absorption of the incorporated drug (Erbe, 1960). Suppositories from batches 2, 4, 9 and 10 had shorter liquefaction times than the suppositories containing only theobroma oil (Batch 11) implying that the included active would be released faster.

The results of the release studies are presented in Figs. 1-3. There was a general increase in percentage of MDZ released with increase in the amount or proportion of the surfactants. Many variables such as concentration and chemical nature of the surfactant and potential interactions of surfactant and drug determine the effect of surfactants on drug release and absorption from fatty suppository bases. MDZ contains a predominant negative charge and the effect the different types of surfactants had on its release from the base was mixed depending on the predominant charge carried by the surfactant. It is evident from the graphical representation that T-65 had the highest enhancement of the release of MDZ. This is not surprising as it is a non-ionic surfactant. Release profile was consistent with increase in concentration of T-65 as the highest concentration of T-65 produced the highest release (Fig. 2). The concentrations of the surfactants used were below their various critical micelle concentrations (CMC) because surfactants are able to cause increase in drug release only at concentrations below their CMC. At concentrations higher than CMC, complexation and entrapment of drug in the micelle formed may retard drug release. Release of MDZ from T-65 containing suppositories was faster than the suppositories formulated without surfactants. This also lends further confirmation to the superiority of the surfactant containing base. The effect of CTM on the release followed an inverse relationship as the highest concentration of CTM produced the slowest release rate. This could be attributed to a complexation phenomenon between MDZ and CTM due to their opposite charges, with consequent reduction of the emulsifying or wetting power of CTM. SLS also produced a significant increase in the release of MDZ from the suppositories, with more than 50 % increase achieved at 0.02 % concentration. This is likened to the high HLB value of SLS and the retention of the wetting ability due to non-interaction with the active. However, electrostatic repulsion between the negative charges of MDZ and SLS resulted in lower release compared with T-65.

To understand the release mechanisms of MDZ from the suppositories, the release

rate was described using the following equations:

$$\frac{M_t}{M} = Kt^n \quad \ldots\ldots\ldots\ldots\ldots\ldots \quad (1)$$

$$Log\frac{M_t}{M} = \log K + n\log t \quad \ldots\ldots \quad (2)$$

where $\frac{M_t}{M}$ is the fraction of released drug at time *t*, *K* is a characteristic constant which incorporates the structural and geometric configuration of suppositories and n is indicative of release mechanism. As the *K* value becomes higher, the drug is released faster. An *n* value of 1 corresponds to zero-order release kinetics. $0.5 < n < 1$ means a non-Fickian (anomalous) release model and $n = 0.5$ indicates Fickian diffusion (Peppas, 1985). From the plots of log ($\frac{M_t}{M}$) versus log *t*, (Figs. 4- 6) the kinetic parameters *n* and *k* were calculated, and presented in Table 3. Table 3 shows that the *n* values of all the batches except batches 1 and 7 were between 0.5 and 1. This indicated that release of MDZ from those suppositories was anomalous (non-Fickian). The release of MDZ from suppositories formulated with 1:3 blend of PKO:GF (batch 1) and those containing 30 mg CTM per suppository (batch 7) was found to follow almost zero-order kinetics since their values were about 1.

## Conclusion

The result of the study indicated that metronidazole suppositories with desirable release characteristic could be prepared by incorporating different surfactants into the 1:3 blend of PKO:GF. Application of diffusion model to the release of metronidazole showed release from most of the suppository batches was anomalous.

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