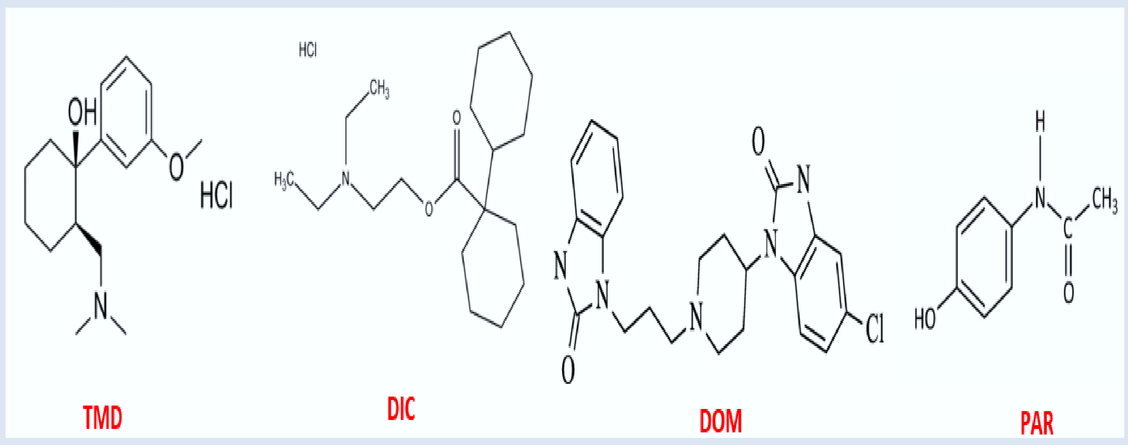
Validated HPTLC Method for the Analysis of Two Ternary Mixtures Used as Supportive Care for Cancer Patients

Mona M. Abdel Moneim[1*], Monica S. Habib[1], Rim S. Haggag[1,2], Tarek S. Belal[2]

[1] *Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Pharos University in Alexandria, Alexandria, Egypt*
[2] *Department of Pharmaceutical Analytical Chemistry, Faculty of Pharmacy, Alexandria University, Alexandria, Egypt*
[*] *Corresponding author: mona.abdelmoneim@pua.edu.eg*

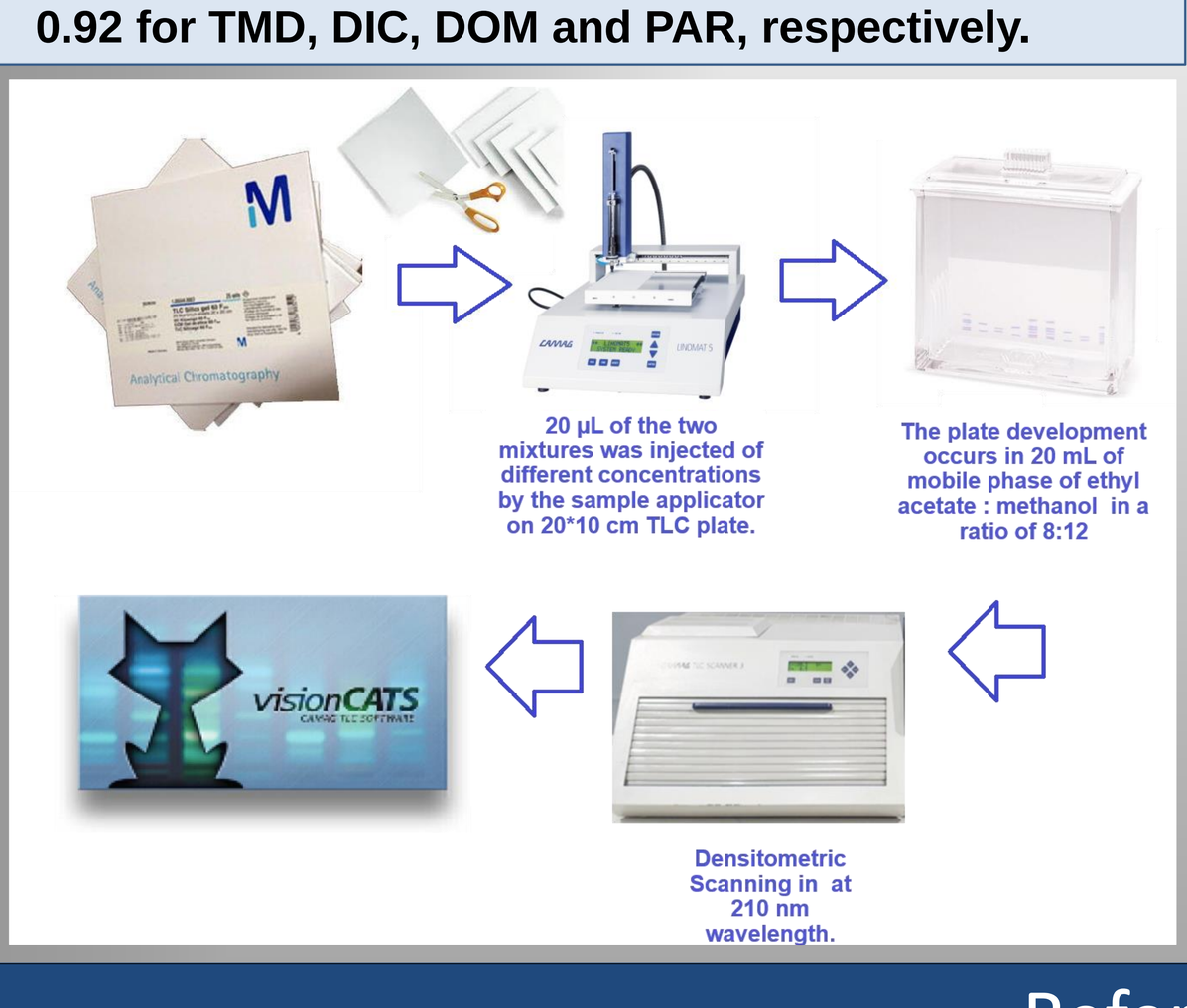
Abstract

Mild and moderate cancer associated pain is usually managed by treatment with opioids and analgesics. Thus, a reliable, simple and rapid high performance thin layer chromatographic – HPTLC method had been proposed and validated for estimation of two ternary mixtures that can be used as supportive therapy for pain relief in cancer patients. The two mixtures are composed of Tramadol (TMD), Dicyclomine (DIC) with either Domperidone (DOM) (Mixture 1) or with Paracetamol (PAR) (Mixture 2). Chromatographic separation of the two ternary mixtures was performed on aluminum plates coated with "silica gel 60 F_{254}" while the solvent system consisted of ethyl acetate: methanol 4:6 (v/v). Densitometric scanning of the separated zones was done at 210 nm for both mixtures and the retardation factor (R_f) values were 0.36, 0.60, 0.89 and 0.92 for TMD, DIC, DOM and PAR, respectively. The method validation was performed in accordance to International Conference on Harmonization guidelines covering all validation parameters as linearity, range, accuracy, precision and specificity. Linearity ranges were 0.1-2.4 µg/band, 0.2-6 µg/band, 0.2-1.2 µg/band and 0.2-3.25 µg/band for DIC, TMD, DOM and PAR, respectively. In addition, the proposed chromatographic technique was successfully applied to the assay of the three drugs in each mixture in laboratory prepared tablets to mimic the dosage forms with their excipients.



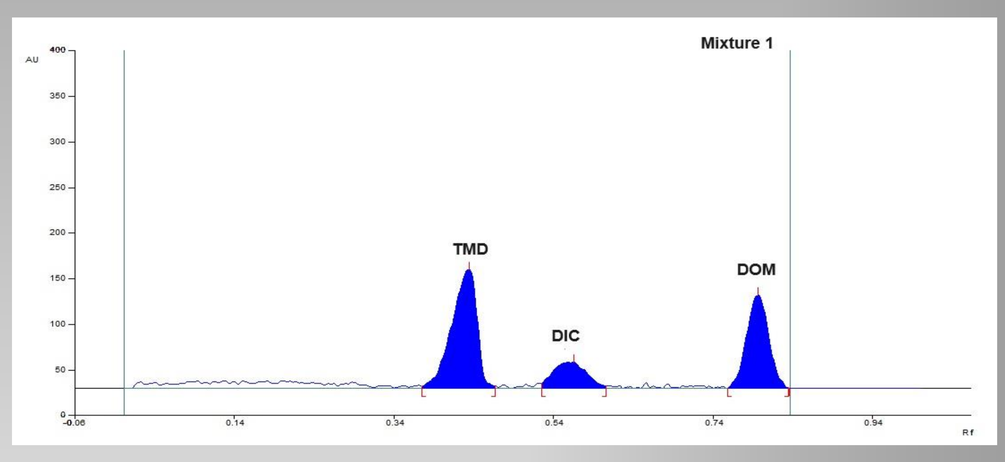
Materials and Methods

HPTLC chromatographic separation of the two ternary mixtures was performed on aluminum plates coated with "silica gel 60 F254" while the solvent system consisted of ethyl acetate: methanol 4:6 (v/v). The sample (20 µL) was injected on the plate as 5 mm bands (6 mm apart). The plates were developed in a Camag chamber (20x20 cm) after its saturation with the used mobile phase for at least 30 mins. Densitometric scanning of the separated zones was done at 210 nm for both mixtures and the retardation factor **(Rf) values were 0.36, 0.60, 0.89 and 0.92 for TMD, DIC, DOM and PAR, respectively.**



Results

Chromatograms of the two mixtures in their dosage form ratio using the proposed HPTLC method



Validation of the proposed HPTLC method.

Parameters	TMD	DIC	DOM	PAR
Linearity range (µg/band)	0.2-6	0.1-2.4	0.2-1.2	0.2-3.25
Correlation coefficient	0.9942	0.9934	0.9976	0.9965
Slope	1831.11	4662.93	2768.84	1943.64
Intercept	1040.69	-48.27	939.79	575.85
Standard error of slope	69.66	189.61	79.06	66.70
Standard error of intercept	225.45	268.25	61.24	105.55
LOD (µg/band)	0.720	0.271	0.078	0.273
LOQ (µg/band)	2.183	0.821	0.237	0.826

ICH Guidelines

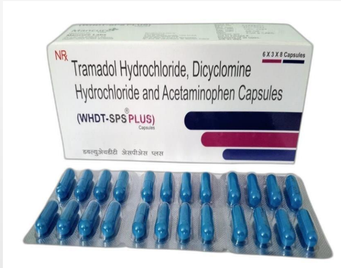
LINEARITY

Accuracy & Precision

Different synthetic mixtures of each mixture were analyzed in replicates (n = 5). The assay was repeated in the same day for accuracy and intra-day precision assessment and on different days for inter-day precision assessment. All Er (%) values and % RSD were less than 2% indicating good accuracy and precision of the proposed method

Selectivity

The successful analysis of the synthetic mixtures prepared with different ratios of both drugs including their dosage form ratio proves the method's selectivity, where good recovery and deviation results were obtained indicating that there is no interference between the two drugs



Since the tablets of both mixtures are not available in the Egyptian markets, the HPTLC method was tested for determination of both drugs in presence of ingredients commonly present by preparing laboratory prepared dosage form. All recovery % and RSD % results were acceptable.

Conclusions

The proposed HPTLC method is simple, accurate and reliable to be used for the assay of the two ternary mixtures. Thus, the proposed method could be useful for routine quality control analysis of the concerned drugs either in their bulk form or in combined drug products. The proposed HPTLC method has the advantage of low solvent consumption, high throughput by running several samples on the same plate, cheaper instrument and lower energy consumption. After being fully validated, the HPTLC method was able to analyze the ternary mixtures with their challenging dosage form ratios in their laboratory prepared tablets in presence of common tablet excipients.

References

- Leppert W. Tramadol as an analgesic for mild to moderate cancer pain. Pharmacol Rep. 2009;61(6):978-92. doi: 10.1016/s1734-1140(09)70159-8. PMID: 20081232.
- Reddy SL, Reddy PS, Reddy SG, Reddy SL. Validated Stability Indicating Liquid Chromatographic Method For Simultaneous Estimation of Paracetamol, Tramadol and Dicyclomine in Tablets. Int. J. of Pharm. Sci. and Res. 2015;6(3):1230. DOI: 10.13040/IJPSR.0975-8232.6(3).1230-40
- Ahir KB, Patelia EM, Mehta FA. Simultaneous Estimation of Tramadol HCl, Paracetamol and Domperidone in Pharmaceutical Formulation by RP-HPLC Method. J Chromat Separation Techniq. 2012;3(152):2. https://doi.org/10.1080/10826071003684224