Advances in Health Sciences Research, volume 38
Proceedings of the 1st International Ninevah Conference on Medical Sciences (INCMS 2021)

Spectrophotometric Determination of Metoclopramide Hydrochloride in Pharmaceutical Preparations Via Schiff 's Base Reaction

Rabee Ali [1], Nabeel Othman[1, *]

[1] *Chemistry Department / College of Science / University of Mosul / Mosul ,Iraq*

[*]*Corresponding author. Email: nsn20002004@uomosul.edu.iq*

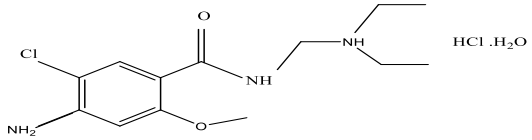
ABSTRACT

A method for determining metoclopramide (MCPD) as pure and in formulations has been suggested. The proposed method is characterized by simplicity, speed, and sensitivity. It is a spectrophotometric method based on the condensation interaction between the primary aromatic amine group in metoclopramide and the carbonyl group of aromatic aldehyde in the chromogenic reagent 2-hydroxy-1-naphthaldehyde (2HND) in an acidic medium of H_2SO_4 and absolute ethanol as the optimal medium. The Schiff base product of the condensation reaction is a yellow-colored product, which exhibits the highest absorption intensity at the wavelength of 447 nm. The proposed method is obeyed to Beer's law in the concentration range of 2 to 20 µg/ml, with a determination coefficient of 0.9960, molar absorption of 1.15 x 10^4 l/ mol.cm, while Sandell's sensitivity index has a significance of 0.0306 µg/ cm^2.The method was estimated statistically by calculating the percentage of relative error (RE%) can express the accuracy and the percentage of relative standard deviation (RSD%) can express precession. The method has been successfully applied in the determination of metoclopramide in pharmaceutical preparations in the form of tablets and injections.

Keywords: *spectrophotometric determination, metoclopramide,2-hydroxy-1-naphthaldehyde, Schiff base.*

1. INTRODUCTION

The metoclopramide is a part of the benzamide family which chemically derived from anesthetic procaine, and it has the scientific name according to IUPAC 4-amino-5-chloro-*N*-[2-(diethylamino) ethyl]-*o*-anisamide monohydrochloride monohydrate and has the chemical structure as shown in Figure 1.



$C_{14}H_{25}Cl_2N_3O_3$, M.wt. = 354.3 g/mol

Figure 1 Metoclopramide hydrochloride.

Metoclopramide hydrochloride is considered one of the compounds that are highly sensitive to light and therefore its pharmaceutical preparations are stored in opaque containers far from the light. The substance is present in the form of powder or crystals of white color and quickly dissolves in water, alcohol and moderately soluble in chloroform while insoluble in ether.

Metoclopramide is a widely used medication in outpatient treatment and hospitalization for its antiemetic properties, was also used to reduce the period of the first stage of labor in primitive women, for stomach and esophageal problems and is a dopamine-receptor blocker [1-4].

There are different spectrophotometric methods in which metoclopramide has been estimated using different reagents in various types of reactions, such as Schiff's bases[5,6],diazotization coupling [7],ion pair[8]. The spectrofluorometric method [9]. In addition, metoclopramide was determined by using other methods such as HPLC[10-13]),flow injection[14], electroch-

emiluminescence [15] and capillary electrophoresis [16].

The aim of the present work is to develop a simple and specific spectrophotometric method for the determination of metoclopramide as pure and in formulations.

2. EXPERIMENTAL

2.1. Apparatus used

All absorbance measurements were performed with a double-beam Jasco V-630uv-visible spectrophotometer, 1 cm thick glass cell, and a BP3001 professional pH meter was used.

2.2. Chemicals

All chemicals were of analytic quality. Table 1 contains chemicals used in the present investigation.

Table 1. Chemicals used.

Company	Chemical formula	M.wt g/mol	Chemicals
SDI/Iraq	$C_{14}H_{25}Cl_2N_3O_3$	354.3	Metoclopram-ide.HCl
Fluka	$C_{11}H_8O_2$	172.1	2-Hydroxy-1-naphthalde-hyde
Scharlau	H_2SO_4	98.08	Sulphuric acid
Romil	C_2H_5OH	46.07	Ethanol absolute

2.3. Reagents

Metoclopramide hydrochloride solution (100 µg / ml)

This solution was prepared by dissolving 0.0100 g of the pure MCPD (supplied to us from the General Company for Medicines and Medical Supplies Samarra - Iraq (SDI)}, and the volume was completed to 100 ml with absolute ethanol using a volumetric flask and the prepared solution was kept in a dark and sealed bottle.

Solution of 2- hydroxy-1-naphthaldehyde (0.4%)

Dissolve 0.4 g of the reagent in 100 ml of absolute ethanol, using a volumetric flask, and keep it in a dark, sealed bottle, and be stable for two days.

Pharmaceutical solutions

Tablet solution (100 µg / ml)

This solution was prepared by weighing a 10 tablets (Kraranie / Darnitsa, and each tablet contains 10 mg MCPD) exactly then grinded and mixed, then weighed amount of powder equivalent to 0.0100 g of pure MCPD and dissolved in warm absolute ethanol with stirring, then filtering and completing the volume to 100 ml with the same solvent used.

Injection solution (100 µg / ml)

2ml of injection (Emoject,10 mg / 2ml-) was taken and the volume was then supplemented to the limit with absolute ethanol using a 100 ml- volumetric flask.

3. PROCEDURE AND CALIBRATION CURVE

Preparation of the standard curve for the proposed method for estimating MCPD was performed as follows: Increasing volumes within the range 0.1 to 1.0 ml of MCPD solution at a concentration of 100 µg / ml were added to a set of 5 ml volumetric flask containing 0.5 ml of concentrated sulphuric acid and 2.5 ml of 2-HNPD reagent at concentration 0.4% then supplement the volume with ethanol to the extent of the mark. Wait for 10 minutes, and the absorbance of the colored solutions is measured at the wavelength of 447 nm against the blank solution, and as shown in Figure 2, which represents the straight standard curve that follows Beer's law with a range of concentrations 2 to 20 µg/ml, and it is observed that there is a negative deviation from Beer's law for concentrations higher than 20 µg/ml.

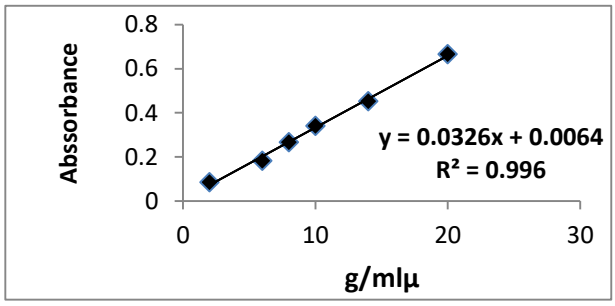


Figure 2 Standard curve for MCPD estimation according to the proposed method.

4. RESULTS AND DISCUSSION

The effect of the components of the reaction was studied separately and for several variables that affect

the absorbance of the colored product at a concentration of 100 µg / ml of MCPD and a final volume of 5ml.

4.1. Selection of acid

In this study, a number of concentrated acids, with an amount of 0.5 ml was used. The results of this investigation are shown in Table 2.

Table 2. The effect of the type of acid used.

Type of acid	Absorbance	λ_{max}
H_2SO_4	0.234	450
HCl	0.0026	452
HNO_3	0.035	494
CH_3COOH	0.055	449

According to the high absorbance of the Schiff base product achieved by using concentrated sulphuric acid so that it fixed in the next experiments.

4.2. Optimal amount of sulphuric acid

Different volumes of concentrated sulphuric acid were added, and it was found that the absorbance of the colored Schiff base product increases with the increase in the amount of acid added, but it is offset by a positive increase in the absorbance of the blank solution (Table 3), consequently, the Interference between the MCPD peak and the blank solution peak increases, and thus the color contrast decreases. Therefore, a volume of 0.5 ml of concentrated sulphuric acid was chosen.

Table 3. The effect of the amount of sulphuric acid.

Sulphuric acid (ml)	Absorbance.	
	Schiff base	Blank
0.3	0.201	0.055
0.5	0.260	0.061
0.75	0.298	0.099
1	0.306	0.144
1.25	0.332	0.187

4.3. The effect of the amount of reagent (2-HND)

The study was carried out by using different volumes 2 to 3 ml of 2-HND reagent at a concentration of 0.4% and each volume concert with different concentrations of MCPD within the range 5 to 20 µg/ml, diluting the flasks with absolute ethanol to a final volume of 5 ml, before absorbance measured waiting 15 minutes, the results are shown in Table 4.

Table 4. The effect of the amount of reagent used.

2-HND	Absorbance /µg MCP ml^{-1}				
	5	8	10	15	20
2	0.107	0.160	0.268	0.401	0.492
2.5	0.134	0.266	0.289	0.498	0.622
3	0.111	0.222	0.267	0.487	0.601

From the results in Table 4, it can be seen that the amount 2.5 ml of the 2-HND reagent solution at a concentration of 0.4% gave the highest absorbance of the Schiff base product, and the highest value of the determination coefficient (0.9837,**0.9922**,0.9839 respectively as mentioned in Table 4), therefore, this amount was adopted in the subsequent experiments.

4.4. The effect of the solvent used for dilution.

For the purpose of studying this effect on the absorption of the formed Schiff base product, 0.5 ml of each MCPD and concentrated sulphuric acid (0.5 ml) and a volume of 2.5 ml of 2-HND reagent were occupied to produce the Schiff base, then the dilution to the mark of 5 ml volumetric flasks was done by using a number of organic solvents in addition to water (Figure 3 and Table 5).

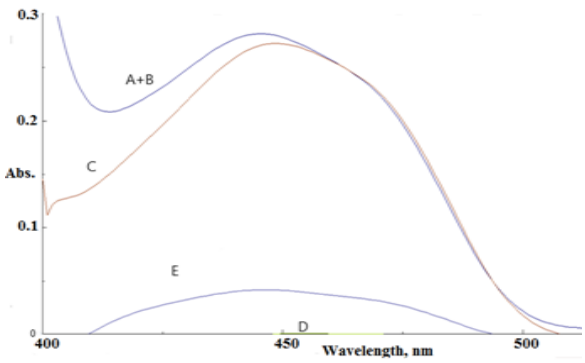


Figure 3 Effect of solvents on the resulting absorption spectrums.

Table 5. Spectral properties of the product formed in different solvents.

Solvent	A.	nm	ε,l/mol.cm)
Methanol(A)	0.281	447	$1.00x10^4$
Ethanol(B)	0.281	447	$1.00\ x10^4$
1-Propanol(C)	0.272	449	$0.97\ x10^4$
Formic acid(D)	0.0006	454	$0.002x\ 10^4$
Acetic acid(E)	0.0414	660	$0.14\ x10^4$
n-Butanol(--)	Turbid	——	——
Water(--)	Turbid	——	——

The results in Table5 indicate that the two best solvents are methanol and ethanol to give them the highest absorption value, and that water and n- butanol give turbid solutions, so the use of ethanol was kept in subsequent experiments.

4.5. The effect of the addition sequence

The effect of changing the sequence of additives of the reaction components on the absorbance of the colored compound has been studied, the results as shown in Table 6.

Table 6. Effect of additions sequence.

No.	Order	Absorbance
I	S + H^+ + R*	0.280
II	**H^+ + R + S**	**0.338**
III	R + H^+ + S	0.291
IV	R + S + H^+	0.301

* MCPD (S)+ H_2SO_4 (H^+)+ 2-HND(R)

It is noted from the results of the above Table that the sequence (II) gave the highest absorbance of the colored Schiff base product, so this sequence was followed and approved in the subsequent experiments.

4.6. Stability of the Schiff base product

The effect of time on the absorbance of the colored Schiff base product was studied with different time periods. The absorbance of the resulting compound against the blank solution was measured at the wavelength of 447 nm by adding two different concentrations of 10 and 20 µg of MCPD at the final volume of 5 ml (see Table 7).

Table 7. The stability of yellow Schiff base.

Minute	Absorbance/ µg of MCPD	
	10	20
Immediately	0.278	0.536
5	0.390	0.594
10	**0.344**	**0.634**
15	0.344	0.636
20	0.347	0.638
30	0.346	0.635
40	0.345	0.624
50	0.342	0.613
60	0.340	0.602

It is noted from the results proven in Table 7 that the Schiff base product is stable after about 10 minutes and remains stable for 50 minutes. Therefore, this time of 10 minutes was adopted in the subsequent experiments.

4.7. Accuracy and precision of the method

The accuracy and precision of the proposed method were studied by calculating the relative error (RE%) and the relative standard deviation (RSD%) of two different quantities of MCPD 10 and 20 µg in a final volume of 5 ml and five reading for each. The results, as shown in Table 8, indicate good accuracy (according to RE%) and precision (according to RSD%) of the suggested method.

Table 8. Accuracy and precision of the proposed method for estimating MCPD.

µg of MCPD		Re.*%	RE%	RSD%
Present	Measured			
10	9.58	95.8	- 4.2	0.79
20	20.52	102.6	2.6	1.07

* Average of five determinations.

4.8. The nature of the resulting product

Depending on the nature of the reagent of 2 - hydroxy-1-naphthaldehyde, which contains only one group of carbonyl and metoclopramide that contains one

group of the primary amine, the proposed mechanism for the formation of the colored Schiff base resulting from the reaction of the acidic reagent with metoclopramide as shown in Figure 4.

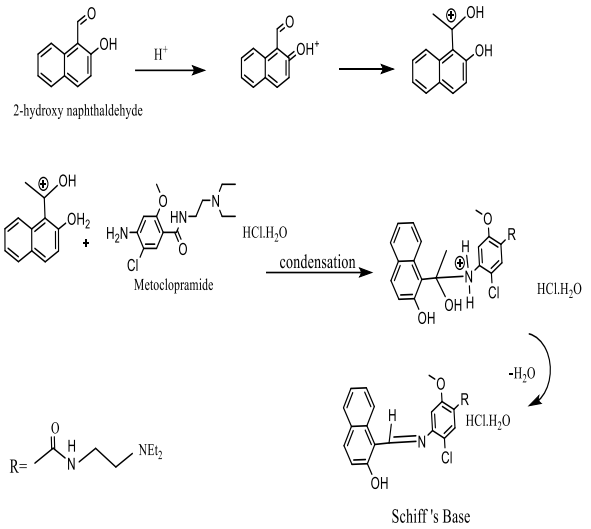


Figure 4 The proposed mechanism for the reaction between MCPD and 2-HND.

4.9.Final absorption spectrum

When adding the MCPD to the solution containing both concentrated sulphuric acid and 2-hydroxy-1-naphthaldehyde and under the optimal experimental conditions obtained, a yellow-colored compound is formed that gives the highest absorption at the wavelength of 447 nm compared to the blank solution (Figure 5).

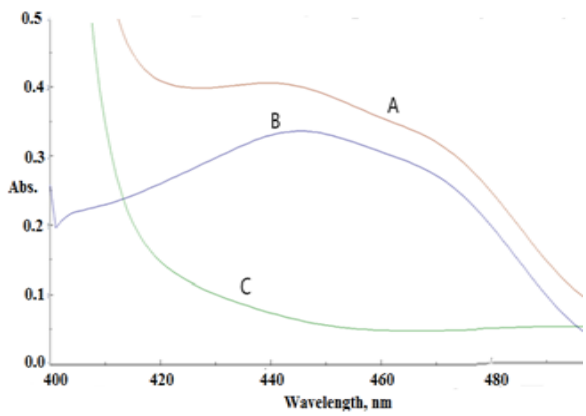


Figure 5 Absorption spectrum for 10 µg MCP / ml measured: A – Schiff base versus the solvent used, B - against the blank solution, C -the blank solution versus the solvent used.

4.10. Applications of the method

The proposed method was applied to the determination of the MCPD in the form of tablets and injections, with two different concentrations of 10 and 20 µg MCPD/ml. The results in Table 9a and b improve the success of the proposed method in estimating of MCDP in its two formulations (tablets and injection).

Table 9a. Estimation of MCPD in tablets.

µg MCPD in ml	Darnitsa 10mg Tablet Ukrainian production		
	Re.*%	RE%	RSD%
10	97.5	**-2.5**	1.33
20	96.9	-3.1	1.56

* Average of four determinations.

Table 9b. Estimation of MCPD in injection.

µg MCPD in ml	Emoject 10mg/2ml injection Turkish production		
	Re.*%	RE%	RSD%
10	101.2	1.2	1.05
20	100.9	0.9	1.24

* Average of four determinations.

The standard addition method was also used to estimate the drug content of the tablet and injection preparations to prove that there is no interference with the additives used in the pharmaceutical industries, by taking two concentrations of 3 and 6 µg of the two MCPD pharmaceutical preparations solution individually and adding different concentrations of the standard MCPD solution on the condition that the maximum extent of the estimate in the calibration curve is not exceeded (Note one of the volumetric flask contains only the unknown preparation) (Figure 6a and 6b).

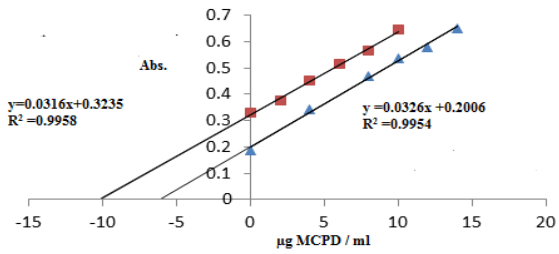


Figure 6a Plot of using standard addition method in estimation of MCPD in tablet.

Table 11. Comparison of the proposed method with author method.

Parameters	Present method	Ref.[7]
Temperature (◦C)	Room temperature	Room temperature
λ_{max} (nm)	447	513
Reagent	2-Hydroxy naphthaldehyde	Diazotised p-nitroaniline
Development time(min.)	10	10
Medium of reaction	Organic solvent	Aqueous
Beer's law range(ppm)	2-20	0.2-25
RSD(%)	≤.1.56	------
Molar absorptivity ($l.mol^{-1}.cm^{-1}$)	$1.1552x10^4$	$2.313x10^3$
color of the compound	Yellow	Red
Application of the method	Pharmaceutical preparations	Pharmaceutical preparations

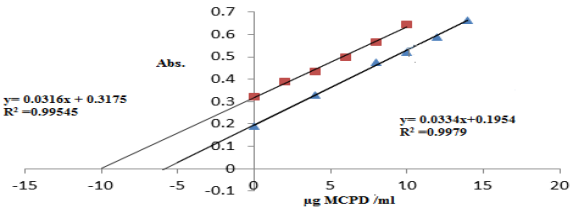


Figure 6b Plot of using standard addition method in estimation of MCPD in injection.

The results in Figure 6a and b are recorded in the Table 10, results express the trustworthiness of the method and the absence of interference with the estimation based on the percentage of recovery.

Table11. The results of Standard addition method for estimating MCPD.

Drug	µg/ml present	µg/ml found	*Re. %	RE %	RSD %
MCPD injection	6	5.85	97.5	-2.5	0.58
	10	10.05	100.5	0.5	0.85
MCPD tablet	6	6.15	102.5	+2.5	0.91
	10	10.23	102.3	+2.3	1.50

4.11. Compare the method with other methods

By comparing some analytical values of Schiff's base formation reaction for estimating MCPD (the current method) with the same values of author spectrophotometric method as shown in Table 11.

It was found that the proposed method has an acceptable sensitivity, the reaction was done at room temperature, and could be applied to quantify MCPD in it is pharmaceutical preparations.

5. CONCLUSION

The reaction of Schiff's base was applied in the proposed method for the purpose of developing a sensitive and fast spectrophotometric method for metoclopramide determination by condensing the amine group of MCPD with the reagent 2- HND in the presence of concentrated sulphuric acid to form the Schiff base of yellow color at room temperature and the colored compound has highest absorbance of 447 nm. The method has been successfully applied in the determination of metoclopramide in pharmaceutical preparations (tablet and injection).

ACKNOWLEDGMENTS

I would like to thank the Department of Chemistry /College of Science/ University of Mosul, for allowing work to complete the research in the department's laboratories.

REFERENCES

[1] Z. Zeb-un-Nisa, S.Shahnaz, M. Rizvi, R. Fatima, S. Ali, S. Shafique, et al., Estimation of equivalence for quality parameters of metoclopramide hydrochloride tablets used to treat emesis, Arch. Pharm.Pract.,12(2),2021,pp25-8.

DOI: https://doi.org/10.51847/xuwqnWXsOY.

[2] H. Taha, M. Al-Rufaie,Z.Motaweq Spectrop-hotometric determination of metoclopramide medicine in bulk form and in pharmaceuticals using orcinol as reagent. Ovidius University Annals of Chemistry, 29(2), 2018,pp 85-89

DOI: https://*doi*.org/*10.2478/auoc*-2018-0012.

[3] M. S.Mashkour, M. K. Kahlol, S. W. AL-Hasnawi, Colorimetric Determination of Metoclopramide Hydrochloride and Glutathione by using 1, 2 naphthaquinolinc-4-sulphonate-sodium reagents, Research Journal of Pharmacy and Technology, 11(8),2018,pp 3290-3294.

DOI: https://doi 10.5958/0974-360X.2018.00605.4

[4] M. Ellaithy, S.Rasheed, A. Shafik, S.Abees, Use of an antiemetic to shorten the length of labor in

nulliparous women, exploring a potential role of an old drug: A randomized controlled trial, International Journal of Gynecology and Obstetrics, Jan;148(1),2020,pp72-78.

DOI:https://doi: 10.1002/ijgo.12998. Epub 2019 Nov 8.

[5] S. Patel, C. Patel, M. Patel, Visible spectrophotometric methods for the estimation of metoclopramide hydrochloride in tablets, Indian Journal of Pharmaceutical Sciences ,68 (3) ,2006 , pp397-399.

DOI: https://doi:10.4103/0250-474X.26663.

[6] E.Thanoon, A. Saeed, Development of green spectrophotometric method for determination of metoclopramide hydrochloride. Egyptian Journal of Chemistry, Mar 9,2021.

doi: 10.21608/ejchem.2021.43857.2923.

[7] W. Abd-Alrada, I. Sulaiman, Determination of metochloropramide hydrochloride by spectrophotometric method by using diazotized p-nitro aniline reagent. international J. Drug Del. Techn, 10(1), 2020 ,pp 68-73.

DOI: doi: 10.25258/ijddt.10.1.11

[8] A. Naggar, E. Seaf, S.Mousa, A. Kotb, A. El-Sayed, Determination of metoclopramide hydrochloride in pharmaceutical formulations using three different spectrophotometric methods. Pharm. Anal. Acta, 8(2), 2017, pp1-9.
DOI: https://DOI:10.4172/2153-2435.1000538

[9] H. Elmansi, S. Mohamed, M. Fathy, Simultaneous determination of metoclopramide and aspirin by spectrofluorimetric technique: application to pharmaceutical formulations and human plasma. Analytical Methods, 8(6), 2016, pp1281-92.
DOI: https://doi.org/10.1039/C5AY02777B

[10] H. Marzouk, E. Ibrahim, M. Hegazy, S. Saad, Greenness profile assessment of selective liquid chromatographic methods for determination of a quaternary antimigraine combination along with three of their related official impurities. Biomedical Chromatography, 2021,e5132.

DOI: https:// doi: 10.1002/bmc.5132

[11] F. Ullah, Z. Iqbal, A. Raza, I. Khan, F. Ullah, M. Hassan, Simultaneous determination of methotrexate and metoclopramide in physiological fluids using RP-HPLC with ultra-violet detection; application in evaluation of polymeric nanoparticles, Journal of Liquid Chromatography and Related Technologies,40(20), 2017,pp1020-30. DOI:

https:// doi.org/10.1080/10826076.2017.1399417

[12] N. Kahali, J. Khanam, A Novel HPLC method validation based on analytical techniques of metoclopramide benzamide derivative (metoclopramide base) and its determination from solid dispersion by solvent evaporation method, J. Appl. Pharm. Sci.,2018,pp18-026.

DOI: doi: 10.7324/JAPS.2018.8203.

[13] I. Kaukab, S. Hussain Shah, G. Murtaza., Single dose pharmacokinetics of metoclopramide oral tablets utilizing HPLC-UV method. Current Pharmaceutical Analysis,15(7), 2019,pp 703-9. DOI:

https://DOI 10.2174/1573412914666180425123202

[14] M. Mohammed , S. Omar , H. Nashwan, Determination of metoclopramide in pharmaceutical commercial using flow injection chemiluminescence technique, Sys Rev Pharm, 11(3),2020,pp 503- 507.

DOI: doi:10.5530/srp.2020.3.64

[15] F. Yang, W. Zhang, B. Liu, Y. Ji, Y. Zhao, K. Zhou, Optimization of Electrochemiluminescence experimental conditions for metoclopramide determination based on response surface methodology. Int. J. Electrochem. Sci.,15, 2020,pp 8145-55.

DOI: doi: 10.20964/2020.08.79

[16] A. Mbaye, C. Foulon, M. Lecoeur, Capillary electrophoresis as a versatile tool for quality control and epidermis permeation studies of transdermal formulations, Journal of Pharmaceutical and Biomedical Analysis,193, 2021, pp 113732. DOI:

10.1016/j.jpba.2020.113732