Dedication

To whom I shall beseech, pay my thanks and prayers, to finish this tremendous work, Allah the Almighty, praise be to Him.

To the soul of my beloved father, beloved compassionate mother, beloved wife, son, brothers, sister and beloved family.

To my faithful friends

To the free souls of scientists and students who have devoted themselves to science despite their severe sickness and obstacles.

Acknowledgments

Thank Allah

I would like to express my sincere gratitude to my supervisor, **Dr. Mohmaed El Mukhtar Abdelaziz ,** for his constant support and faith that he gave me during my Ph.D work. It was his vision, patience, motivation, enthusiasm and tremendous knowledge that led me to my final destination.

My thanks are extended to my co/supervisor **Professor Zeid Abdullah Al-Othman** , Head Department of Chemistry School of science in The King Saud University for his constant motivation and support that he provided me during the entire process of my research. I am also thankful to all members of Advanced Materials Chair in The King Saud University, especially **Dr Ahmed Aquel**, **Prof A. Yacine Badjah-Hadj-Ahmed** and **Dr.Munir** for their constant motivation and support that they provided during the entire process of my research.

Furthermore, I gratefully acknowledge **Dr**. **Babiker Y. Abdulkhair** for his advice, supervision and crucial contribution in my research. His guidance helped me during all the time of my research. I could not have imagined having a better advisor and mentor for my PhD study, other than him.

List of Abbreviations

AC	Affinity chromatography
ACN	Acetonitrile
AFM	Atomic force microscopy
AIBN	Azobisisobutyronitrile
BMA	Benzyl methacrylate
BP	Bonded phase
CLC	Capillary liquid chromatography
CE	Capillary electrophoresis
CEC	Capillary electrochromatography
CLSM	confocal laser scanning microscopy
CZN	Chlorzoxazone
DMSO	Dimethylsulfoxide
DVB	Divinyl benzene
EDMA	Ethylene dimethacrylate
ESI	Electrospray ionization
FDA	Food and Drug Administration
GC	Gas chromatography
GLP	Good laboratory practice
GMA	Glycidyl methacrylate
HETP	height of effective theoretical plate
HI	Hydrophobic interaction.
HMA	Hexyl methacrylate
HPLC	High performance liquid chromatography
ICH	International Conference on Harmonization
IE	Ion-exchange
i.d.	Internal diameter
IEC	Ion-exchange chromatograph
ISEC	Inverse size-exclusion chromatography
IS	Ion suppression
ISO	International Standards Organization
IP	Ion pair
LC	Liquid chromatography
LL	Liquid liquid
LOD	Limit of detection
LOL	Limit of linearity
LS	Liquid solid
LOQ	Limit of quantification
m-HPLC	micro high performance liquid chromatography
MC	Metal complexation
MIP	Mercury intrusion porosimetry
ML	Micellar liquid
MALDI	Matrix assisted laser desorption ionization
MS	Mass spectrometry
MWCNT	Multi-walled carbon nanotubes
NP	Normal phase
ODS	Octadecylsilica

PAR	Paracetamol
PEEK	Polyether etherketone
PLOT	Porous layer open tubular
PEO	Poly ethylene oxide
PTFE	Poly tetrafluoroethylene
ROMP	Ring opening metathesis polymerization
RP	Reversed phase
RSD	Relative standard deviation
SEC	Size-exclusion chromatography
SEM	Scanning electron microscopy
SE	Size-exclusion
SFC	supercritical fluid chromatography
SCOT	Support coated open tubular
TEM	Transmission electron microscopy
TGA	Thermogravimetric analysis
THF	Tetrahydrofuran
TMOS	Tetramethoxysilane
TMSM	3- trimethoxysilyl propyl methacrylate
TPB	total pore blocking
TRIM	Trimethylolpropane trimethacrylate
UPLC	Ultra performance liquid chromatography
USP	United States Pharmacopoeia
UV	Ultraviolet
VIS	Visible
WCOT	Wall coated open tubular

ABSTRACT

This study has been focused on the development of an analytical method for the determination of Paracetamol **(PAR)** and Chlorzoxazone **(CZN)** in their combined pharmaceutical formulation using two stationary phases (home-made columns) which were prepared by two developed methods, one of which was in capillary column while the other was in conventional column.

The first preparation method was achieved using a home-made capillary column (0.10mm i.d. × 200mm length), filled with porous cross-linked hexyl polymethacrylate as monolithic stationary phase. The column morphology was characterized by scanning electron microscopy (SEM). The column showed perfect mechanical stability and permeability over the investigated flow range with regression factor R^2 0.9994. The capillary column was used to separate PAR and CZN in their pharmaceutical formulation. The method proved to be simple, fast, sensitive, efficient, cost-effective and green approach owing is the combination of the amazing properties of a monolithic material and a miniaturized liquid chromatography, which reduces the analytical costs and the effect on the environmental impact of chromatographic applications. Both components were detected using a 3-nL nano-UV cell fixed at 270nm wavelength. The optimized mobile phase was composed of 1% aqueous formic acid solution and acetonitrile at 40:60 ratio, 1.0µL/min flow rate, 4.0nL injection volume and 50°C column temperature. Under the optimized conditions, PAR and CZN were separated in about 6.5min with chromatographic resolution of 2.37. Using the prepared column, the developed method was fully validated and compared with other reported works.

The second preparation method was achieved using also a home-made, conventional column (3.2 i.d., 100 mm length) which was a glycidyl polymethacrylate as monolithic stationary phase. The column morphology was characterized by scanning electron microscopy (SEM). The permeability was evaluated using acetonitrile and water as mobile phases, and uracil as un retained substrate. A simple and economical reverse phase high performance liquid chromatography (HPLC) method has been developed for the simultaneous estimation of PAR and CZN in their pharmaceutical formulations. Components were determined using a UV detector at 270 nm. The mobile phase was composed of 1% formic acid solution and acetonitrile (65:35 v/v), 0.7 mL/min flow rate and 5.0 μL injection volume. The resolution between ingredients peaks was 1.96.

All findings proved that the both validated method using the prepared column is applicable for quality control and routine analysis of the two drugs.

ABSTRACT (IN ARABIC)

لقد ركزت هذه الدراسة على تطوير طريقة تحليلية لتعيين تركيز كل من الباراسيتامول (PAR) و كلوروزوكسازون(CZN) في تركيبتيهما الصيدلانية وذلك بعد تطوير طريقتين لإعداد طورين مونولثين ثابتين في عموديين محليي الصنع ، أحدهما كان في العمود الشعري بينما الآخر في العمود التقليدي.

تمت طريقة التحضير الأولى باستخدام عمود شعري محلي الصنع (طول 200مم-نصف قطر داخلي 0.1مم) وتمت تعبئة العمود بمركب هكسيل-ميتا كريلات المستخدم كطور ثابت على شكل طور مترابط احادي. وقد استخدام المجهر الإلكتروني (SEM) لدراسة شكل العمود الذي اظهر استقرارًا ميكانيكيا مثاليا على مدى نطاق التحقيق مع عامل الانحدار 0.9994 R^2. وقد تم استخدام العمود الشعري لفصل PAR و CZN في صيغتهما الصيدلانية. أثبتت الطريقة أنها بسيطة و سريعة و حساسة و فعالة ، واقتصادية ، وذلك لاستخدام كميات مصغرة من الطور المتحرك ، والتي تعتبر كخطوة نحو تقليل التكاليف التحليلية و التأثير البيئي للتطبيقات الكروماتوجرافية. تم الكشف عن كلا المكونين باستخدام خلية Nano-UV 3 nL عند طول موجة 270نانوميتر. حيث كان الطور المتحرك يتكون من 1٪ محلول حمض فورميك مائي و اسيتونيتريل بنسبة 40:60 ، وكان معدل الانسياب 1.0مايكروليتر/دقيقة وحجم الحقن 4.0نانوليتر ودرجة حرارة العمود 50 درجة مئوية. تحت الظروف المثلى ، تم فصل PAR و CZN في حوالي 6.5 دقيقة مع درجة فصل 2.37. ويمكن استخدام هذه الطريقة الجديدة في التحاليل الروتينية بمصانع الادوية.

تمت طريقة التحضير الثانية باستخدام عمود تقليدي مصنوع محليًا (طوله 100 مم ،نصف قطره الداخلي3.2مم) تم تعبئته بجليسيديل بوليميثاكريليت المستخدم كطور ثابت على شكل طور مترابط احادي. وقد اظهرت دراسة شكل العمود باستخدام المجهر الإلكتروني (SEM) استقرارًا ميكانيكيا وقد تم تقييم النفاذية باستخدام الأسيتونتريل والماء كمراحل متنقلة ، واليوراسيل كمادة غير مستبقية. وتم تطوير طريقة تحليليةباستخدام جهاز كروموتوغرافيا ذات الاداء العالي (HPLC) لتحليل الآني لكل من PAR و CZN في تركيباتهما الدوائية. وذلك باستخدام مكشاف الأشعة فوق البنفسجية عند 270 نانومتر. و طور متحرك يتألف من 1٪ من محلول حمض الفورميك وأسيتونيتريل (65:35 (v / v)، ومعدل تدفق 0.7 مل / دقيقة وحجم الحاقن 5.0 ميكرولتر. كانت درجة الفصل بين قمتي المكونين 1.96. ويمكن استخدام هذه الطريقة الجديدة في التحاليل الروتينية بمصانع الادوية.

List of publications

Some of the results presented in this thesis were published or accepted for publication as follows:

1- Salih, M. E., Aqel, A., Abdulkhair, B. Y., Alothman, Z. A., Abdulaziz, M. A., & Badjah-Hadj-Ahmed, A. Y. (2018). Simultaneous Determination of Paracetamol and Chlorzoxazone in Their Combined Pharmaceutical Formulations by Reversed-phase Capillary Liquid Chromatography Using a Polymethacrylate Monolithic Column. Journal of Chromatographic Science, 56(9), 819-827.. (Thomson and Reuters – ISI)

2- **Salih, M. E**., Aqel, A., Abdulkhair, B. Y., Obbed M.S., Alothman, Z. A., Abdulaziz, M. A., & Badjah-Hadj-Ahmed, A. Y., Preparation and characterization of glycidyl polymethacrylate monolith column and its application for simultaneous determination of paracetamol and chlorzoxazone in their combined pharmaceutical formulations, accepted and under publication on Journal of Analytical Chemistry. (Thomson and Reuters –ISI)

Table contents

List of Tables

List of Figures