الجـــمـــهوريـــــة الجزائريـــة الديمقـــــراطيـــة الشـــعـــبية
République Algérienne Démocratique et Populaire

MINISTERE DE L'ENSEIGNEMENT SUPERIEUR
ET DE LA RECHERCHE SCIENTIFIQUE

UNIVERSITE ABOU BEKR BELKAÎD
FACULTE DE MEDICINE
DR. B. BENZERDJEB - TLEMCEN

وزارة الـــــــتعـليم العـــالـــــــي
والبـحث العـــــــلمـــــي
جـامعة أبو بكـر بلقـا يد
كـليـة الطب
د. ب. بن زرجـب – تلمسـان

DÉPARTEMENT DE PHARMACIE

MÉMOIRE DE FIN D'ÉTUDES POUR
L'OBTENTION DU DIPLÔME DE DOCTEUR EN PHARMACIE

THÈME:

Synthèse Chimique et Identification des dérivés Bioactifs du Diclofenac Sodique

Présenté par:

ASKRI Ahmed Mounir
BRAHMA Mohammed Ghazali

Soutenu le: 16 Octobre 2021

Le Jury

Président:

Pr. BENSAID Okkacha, Professeur en Chimie Organique

Membres:

Dr. HAMZI Imane, Maître de conférence B en Chimie Organique

Dr. LOUZIM Habiba, Maître assistante hospitalo-universitaire en Chimie Therapeutique

Encadreur:

Dr. BEGHDADI Sara El Mansouria, Maître assistante hospitalo-universitaire en Chimie Therapeutique

ACKNOWLDGMENTS

This work was carried out, for the most part, at the the Laboratory of Therapeutic Chemistry and Organic Chemistry, at the Tlemcen Faculty of Medicine [Abou BekrBelkaid University]

First of all, we thank ***Allah*** *the Almighty for giving us the strength and the means to be able to carry out this work.*

At the end of this work, we would first like to respectfully thank ***Dr. BEGHDADI SARA EL MANSOURIA****, for welcoming us into the laboratory, for having directed this work, for her advice and guidance, as well as for her scientific rigor.*

Prof. BENSAID Okkacha*, We thank you for the honor you have done us by accepting to judge this End of Study dissertation. Your competence and your kindness have always been highly regarded. Please find here the expression of our sincere thanks.*

We present our sincere thanks to ***Dr. HAMZI Imane,*** *as well as* ***Dr. LOUZIM Habiba****, for agreeing to review and discuss this modest work.*

Dedication

I dedicate this modest work first to the memory of my beloved **father** رحمة الله تعالى عليه without whom I would've never made it to this point, you were, you are & you will always be my ultimate role model.

I dedicate this work to my cherished **mother** who was always there for me, who has always encouraged me & was my unsurpassed fan, for her enormous, unconditional love that I could never repay.

To my dear brothers: **Gherissi**, **Habib** and **Hamada** as well as their wives, for having my back and supporting me throughout my hardest times.

To my dear sisters: **Fatima**, **Khaira** as well as their husbands, for being there for me no matter the circumstances.

To my lovely little sister **Doaa**, who never fails to put the smile on my face.

To the priceless **Bachir** and his wife, for taking care of me for as long as I can remember.

To my cute nieces and nephews: **Ahmed**, **Djawed**, **Meriem**, **Haitham**, **Maram**, **Habiba**, **Moataz**, **Nihal**, **Adem**, **Sliman**, **Ratil** and **Yacine**, who brought joy and delight to the family and made it grow larger.

To my dear duo **Ghazali**, with whom I have spent amazing, memorable six years.

To all my friends wherever they are, for the precious memories that can never be forgotten.

And to all those that love me, and wish me happiness.

Mounir

Dedication

Allow me to express here my thanks to a whole small world of people who made this study possible and who contributed to its development in any form whatsoever.

I dedicate this milestone in my life to the memory of my father Hadj Mohammed who died too soon. I hope that, in the world that is his now, he appreciates this humble gesture as a sign of gratitude from a son who has always prayed for the salvation of his soul. May the Almighty God have him in his holy mercy!

TO MY VERY DEAR MOTHER, Zahra

So many phrases, however expressive they may be, cannot show the degree of love and affection I feel for you. You have filled me with your tenderness and affection throughout my journey. You never stopped supporting and encouraging me during all the years of my studies, you were always there by my side to console me when needed. On this memorable day, for me as well as for you, receive this work as a sign of my deep gratitude and my deep esteem. May the almighty give you health, happiness and long life so that I can fulfill you in return.

To my self

You sacrificed a lot during these years, you were able to handle a lot of things at once, something no student in your class could, you are unique and you deserve this humble job.

To you my brothers Boudjemaa, Abdelghani, Brahim, Hadj Messaoud, Hadj Kada and Mhammed. To you my sisters Karima and Samira, Houria, Souad and Meghnia who have always supported and encouraged me during these years of study, thank you for your delicious cakes that you have baked for me, may God give you health, happiness, courage and above all success.

To my adorable nephews Zakaria, Ishak, Anes, Yasser, Yakoub and little Youcef. To my nieces Hadil, Ilham, Wafaa, Mariem and Farah. I dedicate this work to you, hoping that in the future you will choose a specialty that fascinates you and which will allow you to express your maximal creativity.

To my dear partner Mounir, to my dear friends: Abdelhak, Anes, Hakim, Reda. To my dear colleagues; with whom I shared unforgettable moments during my studies: Sidahmed Twil, Affif, Karim, Khaldi, Brahim Rahmane, Youcef and Brahim. To everyone I love and who loves me.

Finally, I hope from the bottom of my heart that all these little people, my own world, find a word of gratitude here, and that everyone recognizes themselves in their own way.

I also hope that the effort expended in this work meets everyone's expectations. However, I am solely and solely responsible for the oversights, gaps and weaknesses that this study may contain. However, the words contained in this thesis are solely my own responsibility.

Ghazali

SUMMARY

RESULTS AND DISCUSSION

LISTE OF FIGURES

LIST OF TABLES

LIST OF ABBREVIATIONS

AA2AR: Adenosine A2 receptor.

AIDS: Acquired immunodeficiency syndrome.

BP: Boiling point.

C: Celcius.

CAS: Chemical Abstracts Service.

CNS: Central nervous system.

COMT: Catechol-o-methyl transferase.

COVID-19: Coronavirus disease 2019.

DCF: Diclofenac.

DDC: Dopa-decarboxylase.

DNA: Deoxyribonucleic acid.

E: Ethambutol.

EOPD: Early-onset Parkinson's disease.

EPTB: Extrapulmonary tuberculosis.

GBD: Global burden of disease.

H: Isoniazid.

HIV: Human immunodeficiency virus.

IR: Infrared spectroscopy.

Ks: Constant f solubility.

m: Middle

MAO-B: Monoamino-oxidase B.

MAOI: Monoamine-oxidase inhibitor.

MAPK: Map kinase.

MDR-TB: Multidrug resistant tuberculosis.

MIC: Minimum inhibitory concentration.

MP: Melting point.

M.tubercuosis: Mycobacterium tuberculosis.

MW: Molecular weight.

NSAID: Nonsteroidal anti-inflammatory drug.

PD: Parkinson's disease.

pH: Hydrogen potential.

Pt: Platinum.

PTB: Pulmonary tuberculosis.

R: Rifampicin.

Rf: Frontal ratio.

S: Streptomycin.

s: Strong.

SDI: Sociodemographic index.

SPPTB: Smear positive pulmonary tuberculosis.

str: Stretching.

TB: Tuberculosis.

TLC: Thin Layer Chromatography.

UV: Ultra-Violet spectroscopy.

w: Weak.

WHO: World health organization.

Z: Pyrazinamide.

µm: micrometer.

GENERAL INTRODUCTION

GENERAL INTRODUCTION

Diclofenac Sodium is the sodium salt form of diclofenac, a benzene acetic acid derivative and nonsteroidal anti-inflammatory drug (NSAID) with analgesic, antipyretic and anti-inflammatory activity. Diclofenac sodium is a non-selective reversible and competitive inhibitor of cyclooxygenase (COX), subsequently blocking the conversion of arachidonic acid into prostaglandin precursors. This leads to an inhibition of the formation of prostaglandins that are involved in pain, inflammation and fever.

The huge efficiency of this core molecule encouraged researchers into developing new derivative compounds of diclofenac. Enormous advances in molecular biology and high throughput techniques allowed us to achieve a better comprehension of the biological processes and to make trials on new targets using new techniques for the diagnosis and treatment of numerous illnesses. Much of this progress is due to the increase in investments in pharmaceutical research and development.

Diseases like tuberculosis, Parkinson and cancer are widespread and require highly monitored medications to prevent their complications. Despite the development of new drugs and regimes for the treatment of the mentioned diseases, they are still spreading and causing more deaths every year.

The commercialized drugs in 2021 still have limited therapeutic activities in combining with targets, their complex chemical structures are causing many serious side effects and in some cases that causes therapeutic failure.

Take for example, Levodopa. At first, can cause a dramatic improvement in the symptoms, but its effects can be less long-lasting over the following years - as more nerve cells in the brain are lost, there are few of them to absorb the medicine. Therapeutic regimes used for the treatment of tuberculosis in another hand do cause serious hepatic dysfunction and the need to go for another regime become mandatory. Lastly, cancer chemotherapy until this day could not differentiate healthy cells from cancer cells, this lead to various consequences during the process of the treatment.

These problems were the starting point of researching new targets and compounds in the aim of discovering a new diclofenac derivative drug used for the treatment for at least one of the mentioned diseases with a better risk-benefit ratio.

The main objective of our work is the synthesis and identification of some bio-active derivatives of diclofenac sodium.

Our work covers two main parts:

- The first is devoted to a bibliographic study subdivided into three chapters:

- The first chapter dealing with a generalized study on tuberculosis disease and the description of the chemotherapy regimens used in Algeria since 2011, followed then by a study of the synthesis and the structure-activity relationship of a new developed drug from diclofenac sodium.

- The second chapter focuses on Parkinson's disease, its epidemiology, physiopathology and the description of the commercialised drugs for this illness, followed then by a study of the synthesis and the structure-activity relationship of a new developed drug from diclofenac sodium.

- The third and the last chapter focuses on cancer disease, its epidemiology, physiopathology and the description of the commercialised drugs for this illness, followed then by a study of the synthesis and the structure-activity relationship of a new developed drug from diclofenac sodium.

- The second part is subdivided into three chapters:

- The first describes the equipments and experimental techniques followed for the synthesis, identification and purity testing of the synthesized product.

- The second chapter deals with the results and their discussion.

- The third one recapitulates the entire work and discuss future perspectives.

BIBLIOGRAPHIC STUDY

CHAPTER I:

Antituberculosis properties of Diclofenac derivatives

I.1. Definition

Tuberculosis (TB) is a transmitted disease that is the leading cause of death from a single infectious agent (ranking above human immunodeficiency virus "HIV" that causes Acquired immunodeficiency syndrome "AIDS").(1)

A year into the beginning of the pandemic, Coronavirus disease 2019 "COVID-19" has been responsible for roughly the same number of deaths caused by TB annually. As of January 2020, TB caused 2 times more deaths than the pandemic did in one year.(2)

There is no effective vaccine against TB infection,(3) and current drug therapies are fraught with problems such as multidrug-resistant tuberculosis (MDR-TB) outbreaks caused by organisms that are resistant to at least two of the first-line antituberculosis medications, isoniazid and rifampin.(4)

I.2. Epidemiology and Clinical Characteristics of Tuberculosis

215 countries and territories have reported TB data annually to the world health organization "WHO" and were reviewed and validated in collaboration with reporting entities.

Overall, the collected data shows that:

- TB illness developed in an estimated 10 million persons in 2019, 8.2% of whom were HIV positive.
- TB incidence declined 2.3% from 2018 and 9% from 2015.
- The total number of TB deaths declined by 7% from 2018 to 2019 and by 14% since 2015.(4)

In Algeria, between 1982 and 2019, the notification rate per 100,000 population of smear-positive pulmonary tuberculosis (SPPTB) has dropped 62.2%, while that of extrapulmonary tuberculosis (EPTB) has risen 91.3%.(5)

For the last decade, the mean detection rate of pulmonary tuberculosis (PTB) was 82.6%. Of all 654,658 reported TB cases over the 38-year period, 271,661 (41.5%) were SPPTB, 301,567 (46.1%) were EPTB cases as described in figure 1.(5)

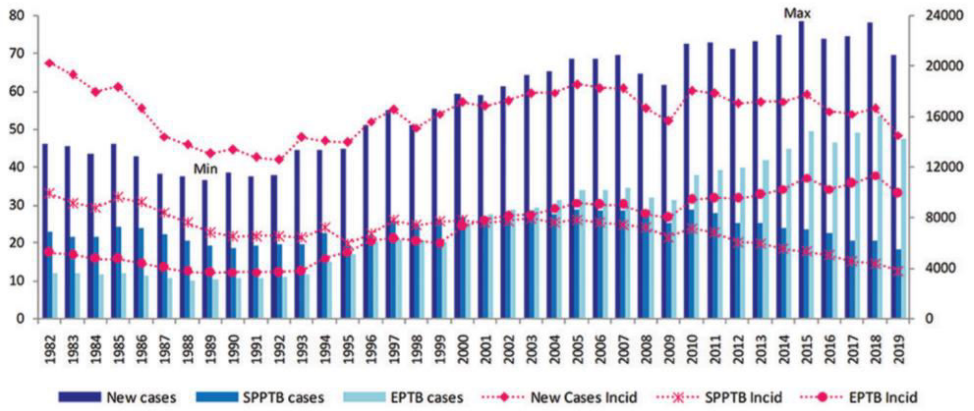


Figure 1: Yearly evolution of overall tuberculosis, smear-positive pulmonary tuberculosis, and extrapulmonary tuberculosis notification cases and notification rates in Algeria from 1982 to 2019.(5)

A study from 2009 to 2019 used to profile the epidemiology of TB was extracted from the patient medical records at Leon Bernard TB unit in Algiers, Algeria.(6)

The overall population covered by this care unit was estimated at 237661 inhabitants in 2019. The extracted data included sex, age, year, month, address, TB classification and outcome and all the patients enrolled in this study received anti TB treatment.(7)

The analysis (Cf. figure 2) revealed that EPTB was more prevalent among women, while PTB was more prevalent among men. Children under the age of fifteen years are most likely to be infected with EPTB. Focusing on PTB among the male population and EPTB among the female population and children to document the epidemiology in Algeria and to examine the risk factors, the diagnostic modalities, the treatment strategies, and the outcomes to carry out preventive and control measures are recommended.(6)

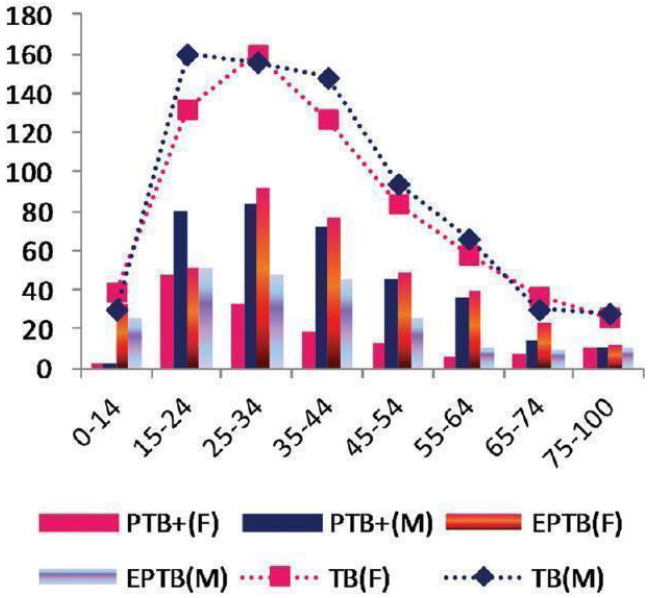


Figure 2: Tuberculosis, extrapulmonary tuberculosis, and pulmonary tuberculosis cases stratified by age group and gender at Leon Bernard tuberculosis unit from 2009 to 2019.(6)

I.3. Pathophysiology and transmission

TB is a lung disease caused by the bacterial pathogen Mycobacterium tuberculosis "*M.tubercuosis*", transmitted from a person with active tuberculosis through coughing, talking and singing.(8,9)

A better knowledge of the mechanisms of drug resistance of *M.tubercuosis* and the relevant molecular mechanisms involved will improve the available techniques for rapid drug resistance detection and will help to explore new targets for drug activity and development.(10) Series in figure 3 shows that a simple cascade for tuberculosis transmission is proposed in which a source case of tuberculosis « 1 » generates infectious particles « 2 » that survive in the air « 3 » and are inhaled by a susceptible individual « 4 » who may become infected « 5 » and who then has the potential to develop tuberculosis « 6 ».(3)

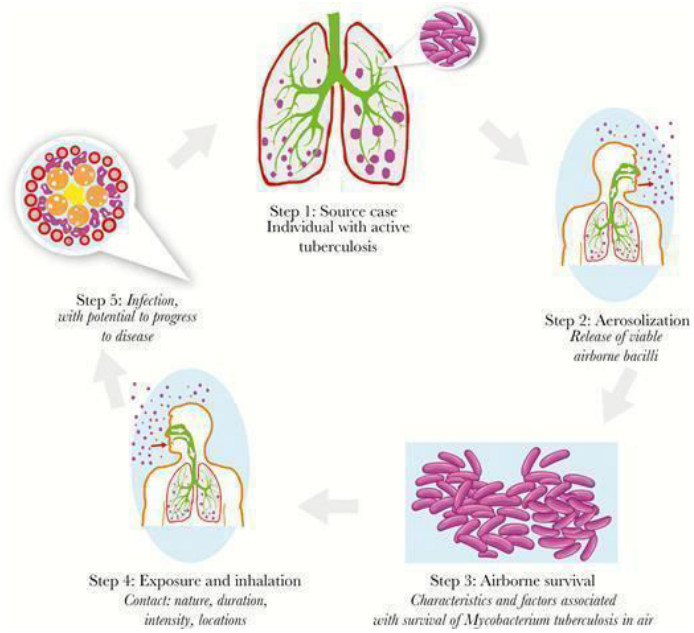


Figure 3: The cascade of tuberculosis transmission.(3)

Exposure to this bacterium can result in a spectrum of infection outcomes, mostly asymptomatic, known as latent TB. However, re-exposure to those with active disease occurs frequently, particularly in crowded conditions.(9)

In a latent TB infection, persistent bacilli are present in a non-replicating dormant state within host granulomas. During reactivation, bacilli start replicating again leading to an active TB infection that can be highly contagious. Mycobacterial lipids and lipolytic enzymes are thought to play important physiological roles during dormancy and reactivation, as summarised in the figure 4.(3)

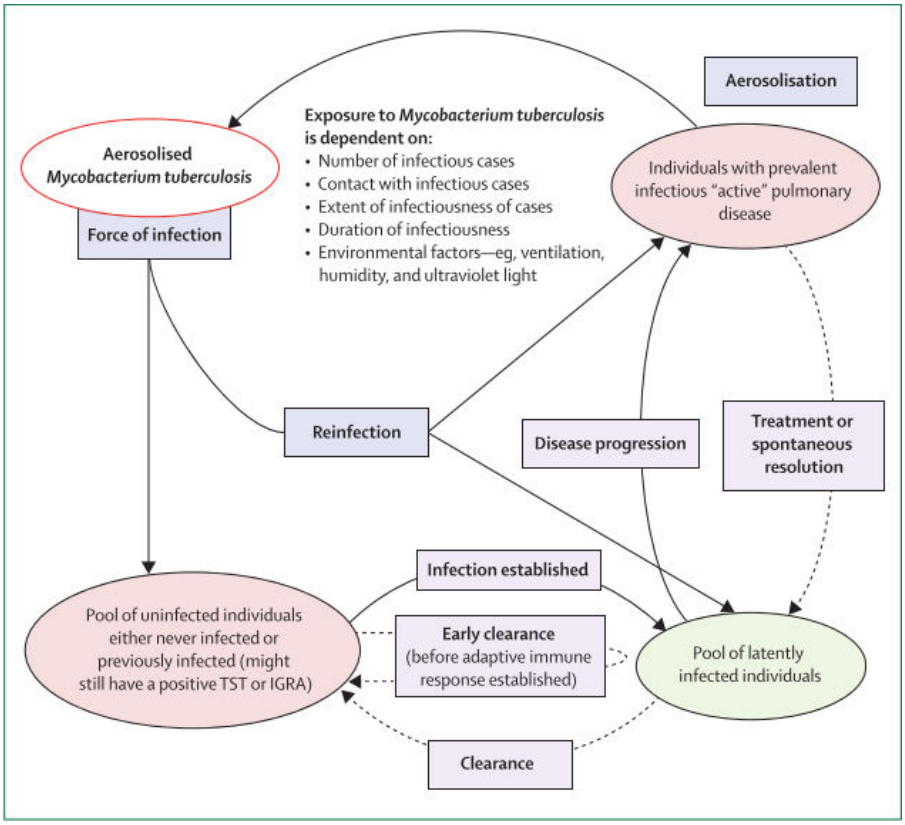


Figure 4: The cascade steps that involve tuberculosis reinfection.(8)

The TB infection process and its remarkable ability to avoid the attacks of the host's immune system make TB an interesting subject for the study of the host–pathogen interplay. *M.tubercuosis* has developed an arsenal of molecular effectors and sophisticated strategies for the success of the infective process.(11)

Figure 5 offers a panoramic overview of a selection of key molecular determinants of TB infection and discusses the molecular targets that could be exploited for the development of new antitubercular agents.(11)

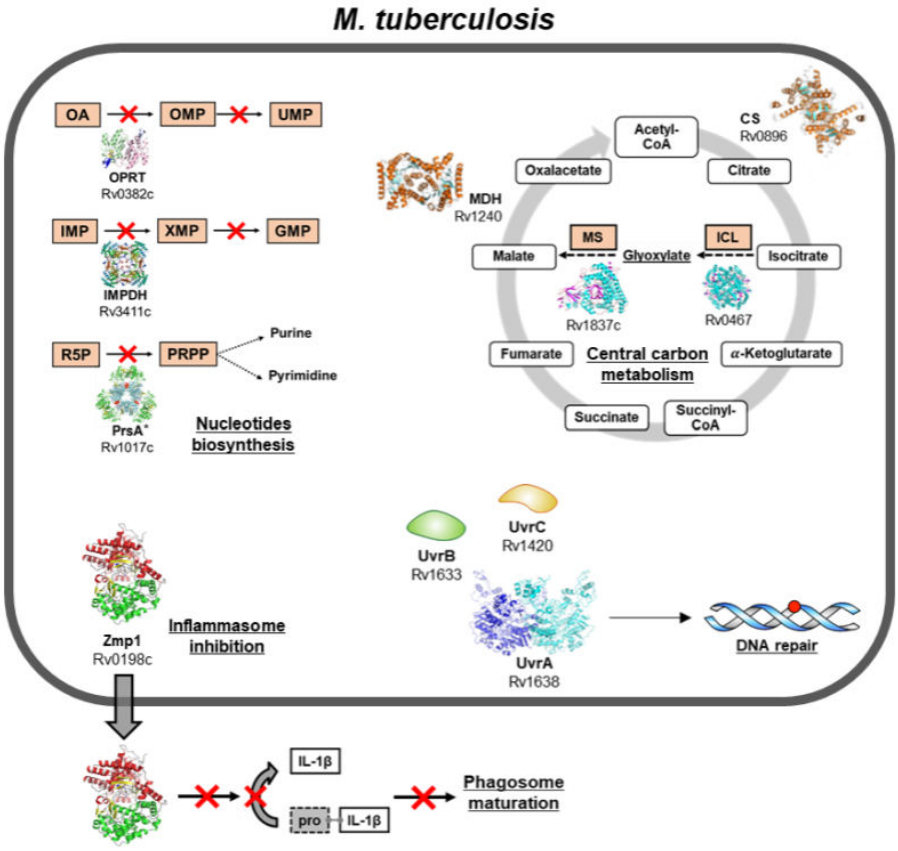


Figure 5: Visual summary, gene names, and structures of the molecular determinants of *M.tuberculosis*.(11)

I.4. Treatment

The treatment of tuberculosis is based on specific chemotherapy and has a dual objective, individual and collective:

- On an individual level, it cures patients with tuberculosis.
- Collectively, it prevents the transmission of the disease in the collectivity and contagiousness of healthy populations, by sterilizing the sources of infections; we prevent the emergence and amplification of resistance of the bacilli to antibiotics.

As such, specific chemotherapy represents the best measures of prevention of tuberculosis.(12)

I.4.1. Tuberculosis chemotherapy

Anti-tuberculosis drugs fall into two groups:

1. Essential drugs.
2. Reserve drugs.

I.4.1.1. Essential drugs

There are five essential drugs (Table I) used in Algeria:

- Isoniazid (H): highly bactericidal and sterilizing.
- Rifampicin (R): highly bactericidal and sterilizing.
- Streptomycin (S): very active on extracellular bacilli that multiply rapidly.
- Pyrazinamide (Z): very active on intracellular bacilli that multiply rapidly.
- Ethambutol (E): bacteriostatic.

Table I: Essential drugs used to treat Tuberculosis.(12)

Essential drugs	Abreviation	Daily dosage (mg/kg)	Form and dosage	Route of administration
Isoniazid	H	5 (4-6)	100/300 mg tabs	Oral
Rifampicin	R	10 (8-12)	150/300 mg tabs/caps	Oral
Pyrazinamide	Z	25 (20-30)	400 mg Tabs	Oral
Ethambutol	E	15 (15-20)	400 mg tabs	Oral
Streptomycin	S	15 (12-18)	1 g ampoule	Parenteral

I.4.1.2 Reserve drugs

These products (Cf. table II) are less active and generally more toxic than the essential products. Reserve drugs in Algeria are in the number of four. They are reserved for the treatment of chronic cases (cases of multidrug-resistant tuberculosis, isoniazid and rifampicin at least) and are prescribed and delivered only under the control of a university hospital pneumophtisiologist. Therefore they cannot be ordered in hospitals in health sectors.

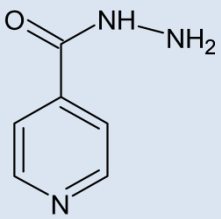
Table II: Reserve drugs used to treat Tuberculosis.(12)

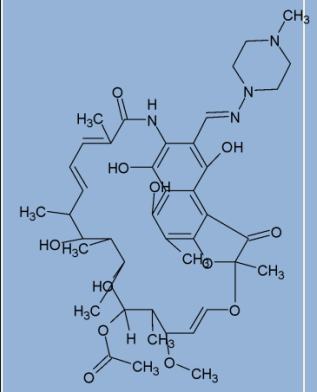
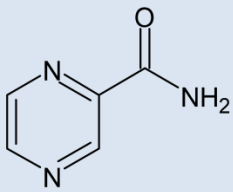
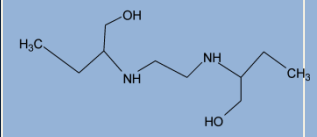
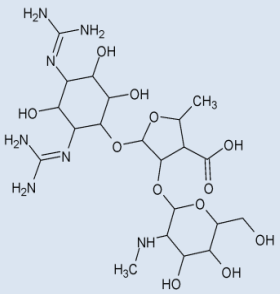
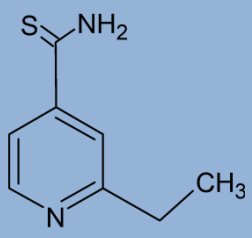
Essential drugs	Abreviation	Daily dosage (mg/kg)	Form and dosage	Route of administration
Ethionamid	ET	15 (10-20)	250 mg tabs	Oral
Ofloxacin	O	10 (8-12)	200 mg tabs	Oral
Kanamycin	K	15 (12-18)	1 g ampoule	Parenteral
Cycloserine	C	15 (10-15)	250 mg tabs	Oral

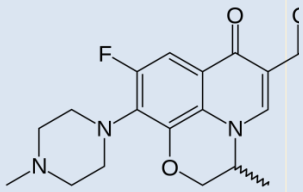
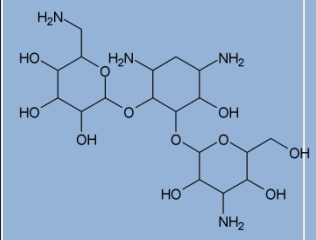
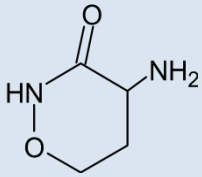
I.4.2. Medicinal chemistry of essential and reserve TB drugs

The table below describes the chemical structures and formulas, the classes and the molecular weights in g/mol for the different TB drugs used in Algeria:

Table III: Chemical properties of essential and reserve drugs used for the treatment of TB (Analysis of Approaches to Anti-tuberculosis Compounds).(13,14)

Drug	Chemical structure	Chemical formula	Class	MW (g/mol)
Isoniazid		$C_6H_7N_3O$	Mild monoamine-oxidase inhibitor (MAOI).	137.14

Rifampicin		$C_{43}H_{58}N_4O_{12}$	Ansamycins	822.95
Pyrazinamide		$C_5H_5N_3O$	Fatty acid synthase FAS I inhibitor	123.11
Ethambutol		$C_{10}H_{24}N_2O_2$	Mycolic acid inhibitor	204.31
Streptomycin		$C_{21}H_{39}N_7O_{12}$	Aminoglycosids	581.58
Ethionamid		$C_8H_{10}N_2S$	Mycolic acid inhibitor	166.24

Ofloxacin		$C_{18}H_{20}FN_3O_4$	Fluoroquinolones 2nd generation	361.36
Kanamycin		$C_{18}H_{36}N_4O_{11}$	Aminoglycosids	484.49
Cycloserine		$C_3H_6N_2O_2$	Amino acids	102.1

I.4.3. Therapeutic standardization (12)

Chemotherapy regimens have been standardized with the aim of:

- Standardize the treatment of tuberculosis according to the severity and location of the disease.
- Avoid "anarchic" treatments that generate bacterial resistance.
- Facilitate forecasting of drug consumption by the health personnel concerned, and stock management.

The standardization of the therapeutic regimen obeys the mandatory rules of administration of anti-tuberculosis chemotherapy which are as follows:

- Administration of drugs in combination.
- Optimal doses calculated according to the weight of the patients.
- Ingestion of medication on an empty stomach, or two hours after a light breakfast.

- Regularity of daily medication which must be directly supervised by a third person, at least during the initial phase of treatment.

The different treatment categories and their indications are as described in the table IV.

Table IV: The treatment categories and chemotherapy regimes indicated.(12)

Chemeotherapy regim categories	Indication	Chemeotherapy initial phase regim (months)	Chemeotherapy maintenance phase (months)
I	PTB	2 HRZE	4 HR
II	PTB category I Faliure	2 SHRZE/1HRZE	5 HRE
III	EPTB	2 HRZ	4 HR
VI	CPTB PTB MDR	Standardized regim or third line regim individualized	

I.4.4. The implementation of the treatment

Apart from the indications for hospitalization, anti-tuberculosis treatment is administered on an outpatient basis. A clinical assessment has to be made before starting the treatment, the main purpose of this assessment is to prevent the phenomena of drug intolerance or interference. It comprises:

- Weigh the patient in order to strictly adapt the dosage of drugs to his weight.
- Testing for sugar and protein in the urine.

A methodical interview to identify “patients at risk” (patients with a history of allergies, neuropsychology, liver or kidney disease or suspected of being co-infected with HIV), as well as patients treated with other drugs (contraceptives, oral hypoglycemic agents, anticoagulants, digitalics, antiretrovirals), likely to have their metabolisms modified by the anti-tuberculosis treatment.

The biological assessment of hepatic or renal functions will be reserved for "patients at risk", identified by questioning, physical examination and chemical examination of the urine. The measures to be taken in these cases are indicated in the paragraph devoted to special situations.

I.4.5. Indications for hospitalization of tuberculosis patients

The patients that follow are subjected for hospitalization:

- Diagnosis of pulmonary TB by gastric tubing (for patients who cannot spit) and of certain extra-pulmonary locations requiring a biopsy (pleura, peritoneum, bone and joint, liver) or special explorations (lumbar puncture, laparoscopy).
- Complications of tuberculosis: cachexia, acute tuberculosis, Pott's disease with paraplegia, coxalgia, hemoptysis of great abundance, pyo-pneumothorax, abundant pleurisy ...
- Complications of anti-tuberculosis treatment: erythroderma, jaundice, loss of hearing or visual acuity.
- Comorbidity (to be managed jointly with another specialist): diabetes mellitus, renal or hepatic insufficiency, psychopathy, drug addiction, HIV co-infection.
- Chronic cases and cases of PTB with multi-resistant bacilli, to be isolated in the specialized department at the start of treatment.

I.4.6. Treatment monitoring

To avoid the common side effects caused by the association of more than two drugs , patients are monitored as follows :

Check the regular intake of medication:

<u>During the initial phase of treatment</u>

For all cases with positive microscopy (Category I and II), direct supervision of the taking of the drugs must be ensured daily (or 5 days a week) in the hospital, or at home (by a member family or the person designated by the patient, monitored by a health worker), until the end of the initial phase of treatment.

For all other cases, the same mode of supervision should be provided when possible. Otherwise, indirect control can be ensured by organizing the weekly delivery of drugs on a fixed day, the intake of which is monitored on the day of delivery and self-administered during the other six days.

During the maintenance phase

Supervision of medication intake is indirect, the treatment being mostly self-administered. This control can then be ensured indirectly, at the time of delivery to the patient of the supply of drugs, which will take place every 2 or 4 weeks depending on the distance from the patient's home

In the event of extra-pulmonary tuberculosis, the effectiveness of the treatment is most often assessed according to the clinical and / or radiological progress.

I.4.7. Side effects of chemotherapy and drug interactions

Chemotherapy for tuberculosis can cause a number of side effects which usually occur during the initial phase of treatment.

These side effects are most often minor, but in 1 to 3% of cases, major side effects may appear, requiring temporary or permanent discontinuation of the offending treatment or drug.

The detection of any undesirable effects of the treatment is ensured by the personnel in charge of the treatment at each appointment or at each patient complaint. In addition to questioning, this detection must include weighing and a clinical examination of the patient. The description of these side effects and the measures to be taken are given in detail below in table V.(15)

Table V: Undesirable side effects of some anti tuberculosis drugs.(15)

Drug	Adverse effects
Isoniazid	Skin rash, Hepatitis
Rifampicin	Abdominal pain, nausea, vomiting, Hepatitis, Purpura
Pyrazinamide	Arthralgia, Hepatitis
Streptomycin	Vestibular and auditory nerve damage, Renal damage
Ethambutol	Retrobulbar neuritis, Ocular side effects
Kanamycin	Vertigo, Auditory nerve damage,Nephrotoxicity
Ethionamide	Hepatotoxicity, Abdominal pain
Cycloserine	Depression, Psychosis

I.4.8. Special measures

The treatment of tuberculosis involves some special measures based on various criterias, we describe some of them below.

I.4.8.1. Treatment of childhood tuberculosis

The treatment of childhood tuberculosis is standardized as follows in the table below:

Table VI: Treatment of childhood tuberculosis.(12)

Child weight (kg)	Initial phase (2 months)	Initial phase (2 months)	Initial phase (2 months)	Maintenance phase (4 months)
	RHZ pediatric tablets 60/30/150	1g ampoule	400 mg E tablets	RH pediatric tablets 60/30
7	1	0.15	-	1
8-9	1+0.5	0.20		1+0.5
10-14	2	0.20	-	2
15-19	3	0.25	1	3
20-24	4	0.33	1	4
25-29	5	0.50	1+0.5	5

I.4.8.2. Patients with liver failure

For patients with biologically proven hepatic insufficiency or for those who have had regressive jaundice in the first weeks of treatment, the doses of isoniazid to 4 mg/kg and the doses of rifampicin to 8 mg/kg should be reduced, while monitoring the level of serum transaminases.(16)

If signs of hepatocellular insufficiency or cytolysis worsen or recur, isoniazid administration should be suspended and treatment with rifampicin below 8 mg/kg, ethambutol at a dose below 8 mg/kg, ethionamide at a dose of 15 mg/kg, and if necessary pyrazinamide, at the normal dose.

I.4.8.3. Patients with chronic renal failure

In patients with patent chronic renal failure, with a serum creatinine greater than 12 mg/l in women and 15 mg/l in men, the dosage of anti-tuberculosis drugs whose elimination is renal should be adopted. Streptomycin, kanamycin, or ethambutol will not be prescribed. The only regimen to administer is the 2RHZ/4RH regimen.(12)

I.5. Structure activity relationship of novel anti tuberculosis derivatives of diclofenac hydrazone acids and diclofenac amides (17)

In 2006, **Dharmarajan Sriram** *and al.* prepared new substituted derivatives of diclofenac acid hydrazones and diclofenac amides. Their biological evaluation was performed to determine their antimycobacterial activities against Mycobacterium tuberculosis. The general procedures for the synthesis of diclofenac acid hydrazone compounds (4a–g) and diclofenac amides (5a–d) are described in figure 6.

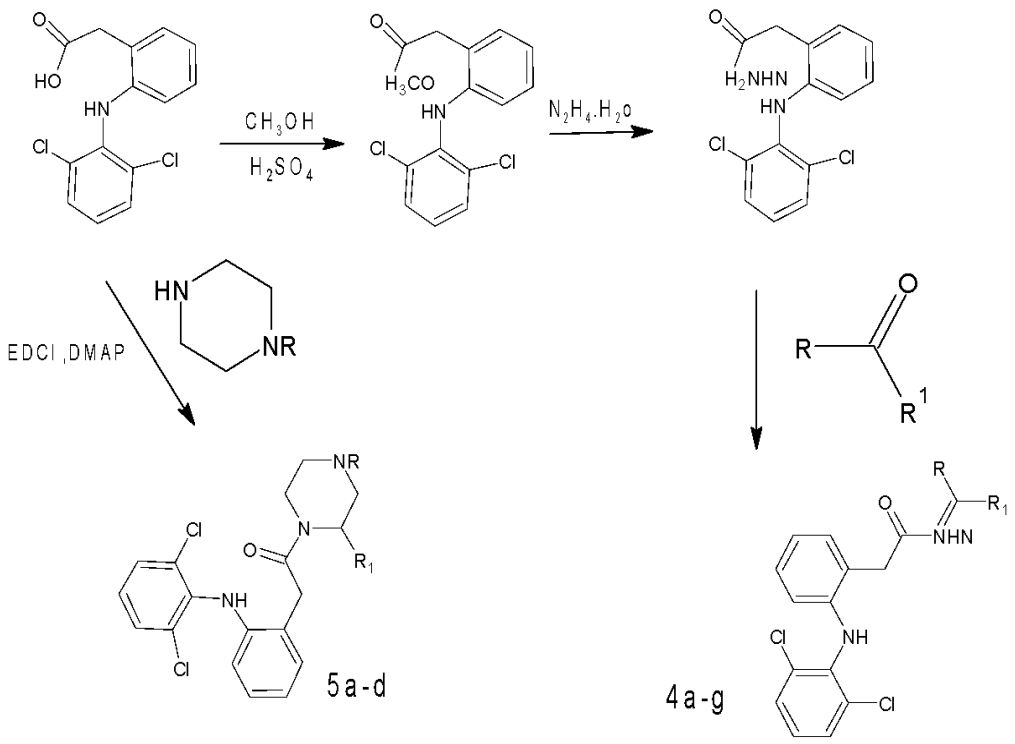


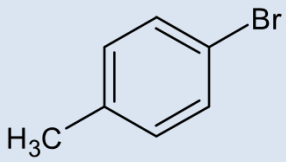
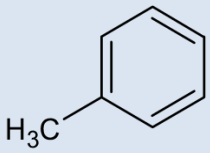
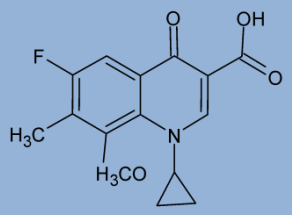
Figure 6: Synthetic protocol of diclofenac acid hydrazone compounds (4a–g) and diclofenac amides (5a–d).(17)

All compounds were screened for their, in vitro, antimycobacterial activity against *M.tubercuosis* by measuring their minimum inhibitory concentration "MIC". Among the

synthesized compounds of the diclofenac acid hydrazones, **4g** was found to be the most active substance in vitro with a MIC value of 0.3614 µm but was two times less potent than isoniazid (MIC=0.1822 µm), a first line antitubercular medication.

Among the synthesized molecules of the diclofenac amides, **5d** was found to be the most active substance in vitro with a MIC value of 0.0383 µm and was more potent than the compound of the reference "Isoniazid". These results are also described in table VII.

Table VII: In vitro antimycobacterial activities of diclofenac acid hydrazone 4g and amide 5d against *M.tuberculosis* compared to Isoniazid.(17)

Compound	R	R1	MIC (µm)
4g			0.3614
5d		-CH3	0.0382
Isoniazid	-	-	0.1822

Subsequently, compound **5d** was tested against *M.tubercuosis* at a dose of 25 mg/kg in six-week-old female CD-1 mice. Results showed that compound **5d** decreased the bacterial load in lung and spleen tissues with 6.36 ± 0.19 and 3.18 ± 0.11 log 10 protections, respectively, and was considered to be promising in reducing bacterial count in lung and spleen tissues. When compared to isoniazid at the same dose level, **5d** decreases the bacterial load equally in spleen tissues. The results are also as described in table VIII.

Table VIII: In vivo activity data of 5d and isoniazid against M.tuberculosis in mice.(10)

Compound	Lungs (logCFU ± SEM)	Spleen (logCFU ± SEM)
Control	8.78 ± 0.12	6.84 ± 0.21
5d	6.36 ± 0.19	3.18 ± 0.11
Isoniazid	5.80 ± 0.18	3.14 ± 0.12

Among the synthesized substances, 1-cyclopropyl-6-fluoro-8-methoxy-7-[[N4-(2-(2-(2,6-dichlorophenylamino)phenyl)acetyl)-3-methyl]-N1-piperazinyl]-4-oxo-1,4-dihydro-3-quinoline carboxylic acid **(5d)** was found to be the most active molecule in vitro with MIC value of 0.0383 µm and was more potent than first line antitubercular drug isoniazid (MIC: 0.1822 µm).

These results highlight the importance of increasing lipophilicity of the compound to overcome transport barriers into the cells. The presence of the fluoroquinolone moiety which might inhibit the deoxyribonucleic acid (DNA) gyrase enzymes of *M.tubercuosis* may explain the enhanced activity of amide derivatives.

CHAPTER II:

Antiparkinson properties of Diclofenac derivatives

II.1. History

Parkinson's disease (PD) has been known to humans since for hundreds of years. It was referred to in the ancient Indian medical system of Ayurveda under the name Kampavata (where “kampa” means tremor in Sanskrit) as early as 600 Before Christ. In Western medicine, it was described by the physician Galen as "shaking palsy" in 175 Anno Domini “AD”.(18)

Sylvius de la Boë wrote on animal motor activities and described the symptoms of resting tremor in 1637, and Sauvages described festination – a form of gait or walk associated with PD.(19)

It was in 1817 that a detailed medical essay was published on the subject by doctor James Parkinson after whom it was named. His essay was called “An Essay on the Shaking Palsy”. This essay established PD as a recognised medical condition in which Dr Parkinson studied and reported six cases in his own practice in London.(20,21)

Some forty three years later, Dr. Jean-Martin Charcot did a great job in refining and expanding this early description and in disseminating information overseas (particularly in France) about PD. He separated Parkinson’s from multiple sclerosis and other disorders characterized by tremor, and he recognized cases that later would likely be classified as Parkinsonism.(20)

In his “Manual of Diseases of the Nervous System”, William Gowers described his personal experience with some patients in 1886. He correctly identified that men were more prone to the disease and also detailed the joint deformities typical of Parkinson’s. Known For his descriptive prose, Gowers offered one of the most memorable similes regarding Parkinsonian tremor (Cf. figure 7).(22)

The image was drawn by Sir Gowers, in 1886 as part of his documentation of PD. The image appeared in his book, “A Manual of Diseases of the Nervous System”, still used today by medical professionals as a primary reference for this disease.(22)

Figure 7: Illustration by Gowers as part of his documentation of Parkinson's disease in his book “A Manual of Diseases of the Nervous System”.(22)

II.2. Definition

PD is a progressive nervous system disorder that affects movement, it comes second to Alzheimer's among neurodegenerative diseases in terms of prevalence worldwide. An estimated 10 million people worldwide have the condition.Although most cases are sporadic but about 10-15% of patients have a positive family history of PD.(23,24)

Parkinson’s is a chronic and progressive movement disorder that involves the malfunction and death of dopaminergic neurons in the brain. These dying neurons produce dopamine, a

chemical that sends messages to the part of the brain that controls movement and coordination. As PD progresses, the amount of dopamine produced in the brain decreases, leaving a person unable to control movement normally.(25)

Both men and women can have Parkinson's disease. However, the disease affects about one and a half times more men than women.(25,26)

One of the most important risk factors of PD is age. Although most people with Parkinson's first develop the disease at about age 60, about 5 to 10% of people with Parkinson's have "early-onset" disease (EOPD) which usually begins before the age of 45, but in some very rare cases it could appear before the age of 21.(27,28)

II.3. Epidemiology

Incidence is the number of new cases of a disorder first developed or diagnosed during a specific time interval within a predefined population at risk. In contrast, prevalence refers to the total number of persons with the disorder at a fixed point in time. Prevalence is a function of both disease incidence and duration.(29)

PD is the second most common neurodegenerative disease (after Alzheimer's) but it's growing faster than this latter. From 1990 to 2015, the number of individuals with Parkinson disease globally increased by 118% to 6.2 million. The proportion of people with PD tend to increase with the region's wealth therefore it is high in rich countries (e.g. Canada, US, Western Europe, 90–180/100,000) compared with poor ones (e.g. sub-Saharan Africa, 30–70/100,000) (Cf. figure 8).(30–32)

Incidence tends to increase with age; both genders and all ethnic groups appear to be susceptible to PD.(33) Environmental risk factors such as pesticide exposure, repeated loss of consciousness, and antidepressant drug use along with family history are all positively related to PD.(34)

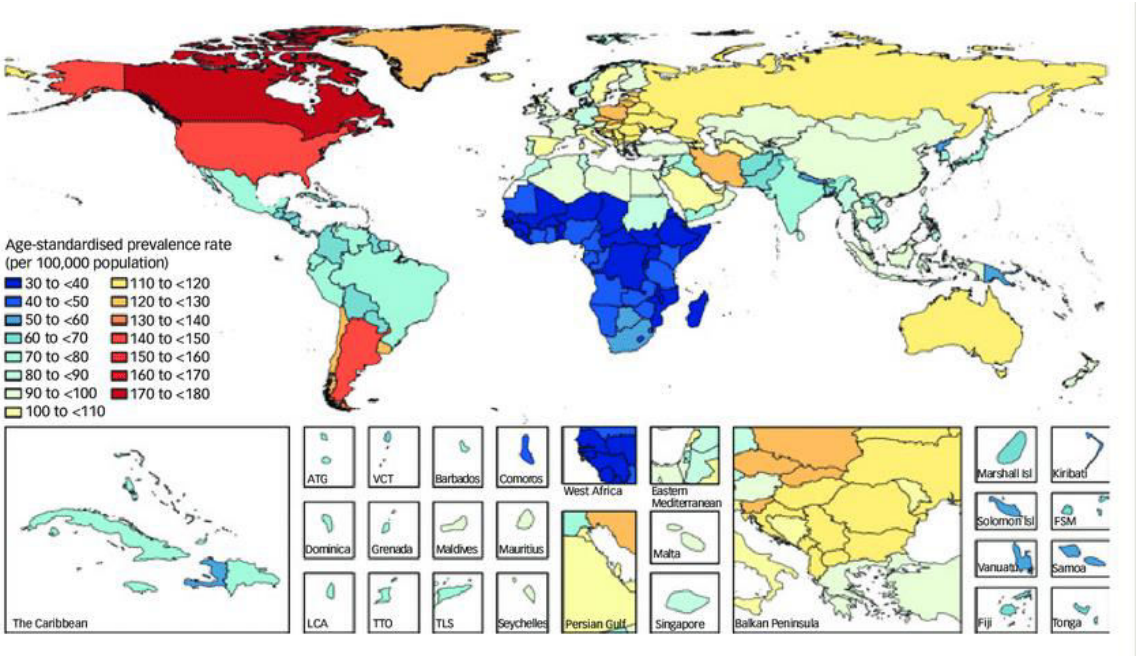


Figure 8: Age-standardised prevalence of Parkinson's disease per 100,000 population for both sexes in 2016.(30)

The Global Burden of Disease "GBD" analysis also estimated PD's prevalence according to socio-demographic index (SDI: a composite measure of income per capita, education and fertility) and reported that there were 2.1 million people with PD in high SDI regions, 1.8 million in middle SDI regions and 113,00 in low SDI regions. PD was estimated to cause 3.2 million disability-adjusted life years and this impact was greatest in high SDI countries (Cf. figure 9).(33,35)

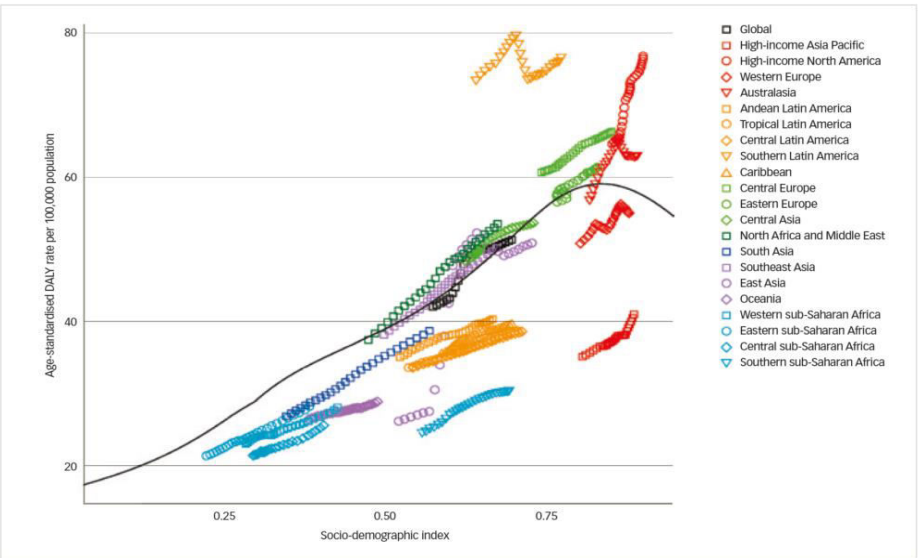


Figure 9: Age-standardised DALY rates for Parkinson's disease in 21 regions by socio-demographic index for years 1990–2016.(30)

II.4. Symptoms

Many different symptoms are associated with Parkinson's disease. But their developpement order and their severity vary for each individual. It's unlikely that a person with Parkinson's would experience all or most of these.(36)

II.4.1. Main Symptoms

The main PD's symptoms that affect movement are:

- **Tremor-shaking:** This begins in the hand or arm and is more likely to occur when the limb is relaxed.(37)
- **Slowness of movement (bradykinesia):** movements become much slower than normal, this translates in difficulties doing everyday tasks and result in a distinctive slow, dawdling walk with very tiny steps.(38)
- **Muscle rigidity:** stiffness and tension in the muscles, which can make it moving around and make facial expressions tough, painful muscle cramps can also occur.(39)

These characteristic symptoms are usually referred to by neurologists as Parkinsonism.

II.4.2. Physical Symptoms

Out of which can distinguish three principal symptoms:

- **Balance problems:** these can make someone with the condition more likely to have a fall and injure themselves.(40)
- **Loss of sense of smell (anosmia):** sometimes happens several years before other symptoms develop.(41)
- **Problems with peeing:** such as having to get up frequently during the night to pee or unintentionally peeing (urinary incontinence).(42)

II.4.3. Cognitive and Psychiatric Symptoms

Depression and anxiety, mild cognitive impairment, slight memory problems, trouble with activities that require planning and organization & dementia are also very common in PD's patients.(43)

II.5. Causes

Parkinson's disease is caused by a loss of nerve cells in the part of the brain called the substantia nigra; nerve cells in this area of the brain are responsible for generating the chemical called dopamine.(44)

For now it's not well-known why the loss of nerve cells associated with PD appears, although the studies are ongoing to identify potential causes. Nowadays, scientists are convinced that a combination of genetic changes and environmental factors may very well be responsible for the condition.(24,44)

II.5.1. Genetic Causes

A number of genetic factors have been shown to increase a person's risk of developing PD, although exactly how these make some people more susceptible to the condition is unclear. Parkinson's can run in families as a result of faulty genes being passed to a child by their parents. But it's rare for the disease to be inherited this way.(44,45)

II.5.2. Environmental Factors

Many researchers believe that environmental conditions may have some serious role in the development of Parkinson’s, it's been suggested that pesticides and herbicides used in farming and traffic or industrial pollution can have some contribution to the condition. But the evidence linking environmental factors to Parkinson's disease is still inconclusive.(24)

II.6. Pathophysiology

Even though we are learning more by the day about the pathophysiology of Parkinson’s disease, it’s still considered largely (in about 90% of cases) idiopathic (of undetermined cause). PD is likely the product of the host’s susceptibility and environmental factors. A small percentage of cases are genetically linked and genetic factors are being intensely studied.(46–48)

The disease is characterized by localized involvement in the black substance also called substantia nigra, this entity is located in the midbrain. The degree of severity of motor symptoms correlates with the severity of destruction of nerve cells.(44)

Destruction of the dopaminergic pathway leads to a decrease in the release of dopamine and an inhibitory acetylcholine overproduction of internal pallidum on the motor thalamus by the gabaergic route. The pathway through the cortex, striatum, internal pallidum and brainstem is called the activator loop.(49)

Black arrows indicate inhibitory connections and gray arrows indicate excitatory connections. The thickness of the arrows corresponds to their presumed activity. Abnormal activity in the 'motor' loop of the basal ganglia is strongly implicated in the development of Parkinsonism as shown in Figure 10 (left panel).(50)

In its simplest form, this model explains the firing rate changes in the basal ganglia as the result of disturbances of the balance of activity in the direct and indirect pathways (Cf. figure 10 right panel).(50)

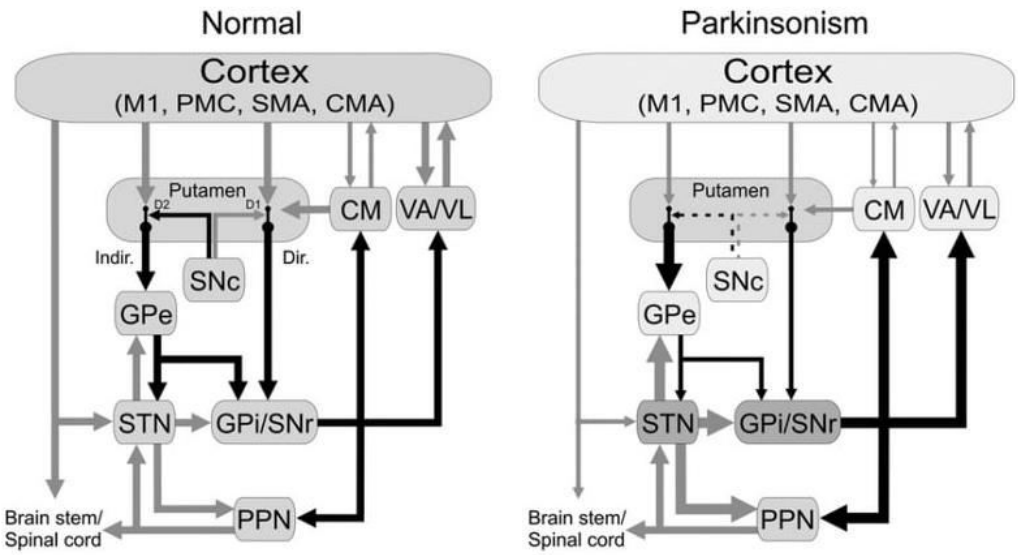


Figure 10: Parkinsonism-related changes in overall activity "rate model"in the basal ganglia.(50)

II.7. Treatments

The treatment of the parkinsonian patient aims for two main goals: on one hand, the ideal control of symptoms of the disease and on the other hand, the absence of progression of the disease, or even lesion recovery if possible, and finally, to minimize the side effects.(51)

Drugs prescribed for Parkinson's disease are intended to restore normal levels of dopamine in the brain. To do this, we have several options.We can: administer a dopamine

precursor (which will be converted into dopamine in the brain), give a substance that works like dopamine (a dopamine agonist), administer a substance that blocks the breakdown of dopamine or its precursor in the brain (to keep levels high as long as possible).(52)

II.7.1. Levodopa-Carbidopa

Levodopa is a dopamine precursor (dopaminergic drug) that is used to treat PD's patients. It is a prodrug that is converted to dopamine by DOPA-decarboxylase and can cross the blood-brain barrier therefore exercising its function thanks to its chemical structure as figure 11 shows. Carbidopa is a peripheral dopa decarboxylase (DDC) inhibitor which decreases the peripheral conversion of levodopa to dopamine, by exercising this action it makes more levodopa available to the brain. This allows a lower dose of levodopa to be used and have more beneficial effect while also reducing the amount of side effects.(53,54)

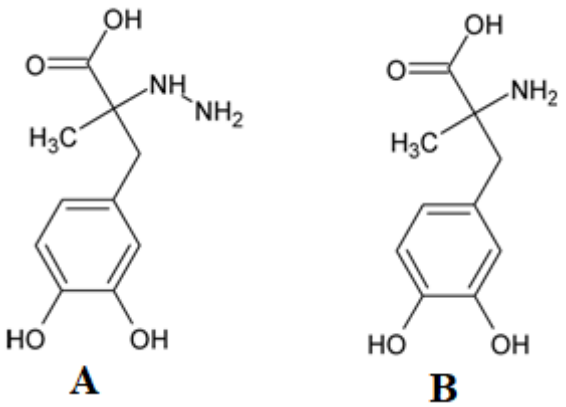


Figure 11: Carbidopa's "A" and Levodopa's "B" chemical structures.(54)

When in the brain, levodopa is converted (decarboxylated) to dopamine and stimulates the dopaminergic receptors, thereby compensating for the lost supply of endogenous dopamine found in Parkinson's disease. To assure that enough concentrations of levodopa reach the central nervous system, it is administered with carbidopa, this latter does not cross the blood-brain barrier, thereby diminishing the decarboxylation and inactivation of levodopa in peripheral tissues and increasing the delivery of dopamine to the central nervous system "CNS".(55)

II.7.2. Direct Dopaminergic Agonists

In the brain, dopamine agonists act like dopamine. They cause fewer involuntary movements than levodopa but are more frequently responsible for other side effects such as

wake-up dizziness, nausea, vomiting, drowsiness and hypnotic episodes (which make driving a vehicle absolutely contraindicated), hallucinations and delusional episodes.(53,56)

In some people high doses of dopamine agonists can cause mood swings, sexual hyperactivity, or even gambling addiction. For these reasons, it is preferable that people who take high-dose dopamine agonists need to be monitored by a psychologist, especially during the time needed for the treatment to be adjusted.(57)

Dopamine agonists come as tablets or capsules to be taken several times a day. Some prolonged-release forms allow only one dose per day. There are two dopamine agonists (Cf. figure 12) that are administered differently: Apomorphine which is injected subcutaneously and Rotigotine which is presented as a transdermal patch to be replaced every day.(58)

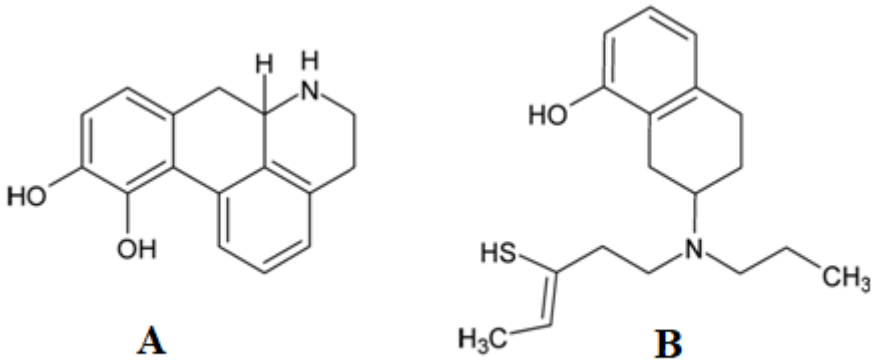


Figure 12: Apomorphine's "A" Rotigotine's "B" chemical structures.(59)

II.7.3. ICOMT

This class of PD medications has no direct effect on its symptoms but is used to prolong the effect of levodopa by blocking this latter's metabolism. COMT (catechol-o-methyl transferase) inhibitors are used primarily to help with "wearing off," in which the effect of levodopa becomes short-lived. We find in this class of medications three well-known drugs: Entacapone, Tolcapone and Opicapone, their structures are shown in figure 13.(60)

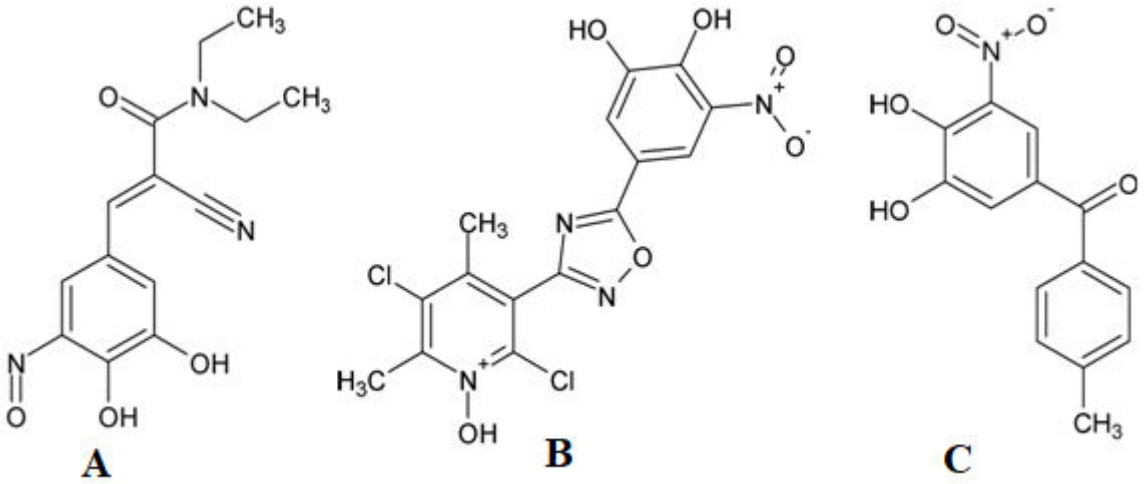


Figure 13: Entacapone's "A", Opicapone's "B" Tolcapone's "C" chemical structures.(61)

II.8. Structure activity relationship of novel antiparkinsonian derivatives of diclofenac triazin (62)

In 2018, Sudhakar P *and al*. performed the design, molecular docking and synthesis of 1,2,4-triazin analogue of diclofenac as potential ligand against PD by replacing the carboxyl group (Cf. figure 14). In this study, Dopamine receptor D3 protein, Dopa decarboxylase, Adenosine A2 receptor, and P38 map kinase were targeted, whereas MAO-B was taken from a protein data bank.

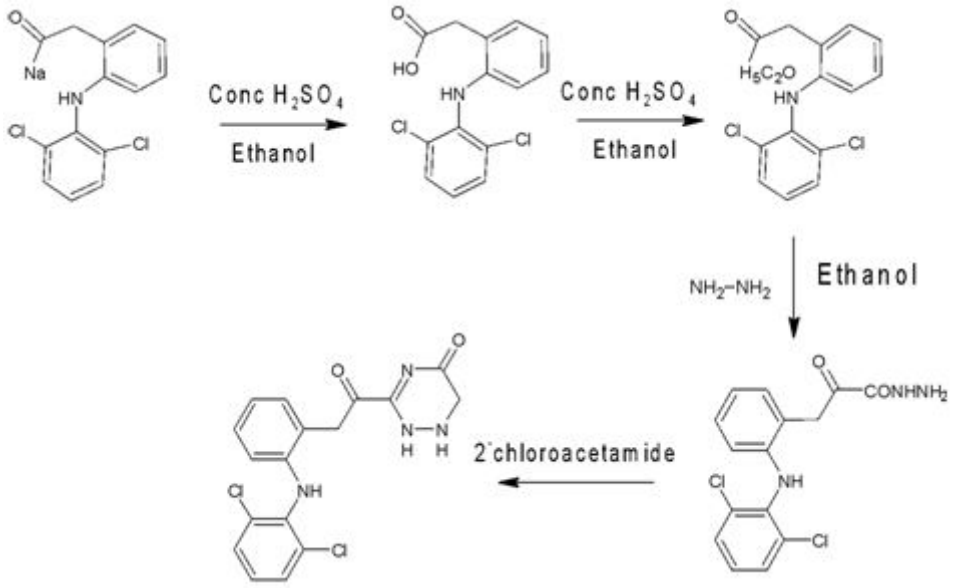
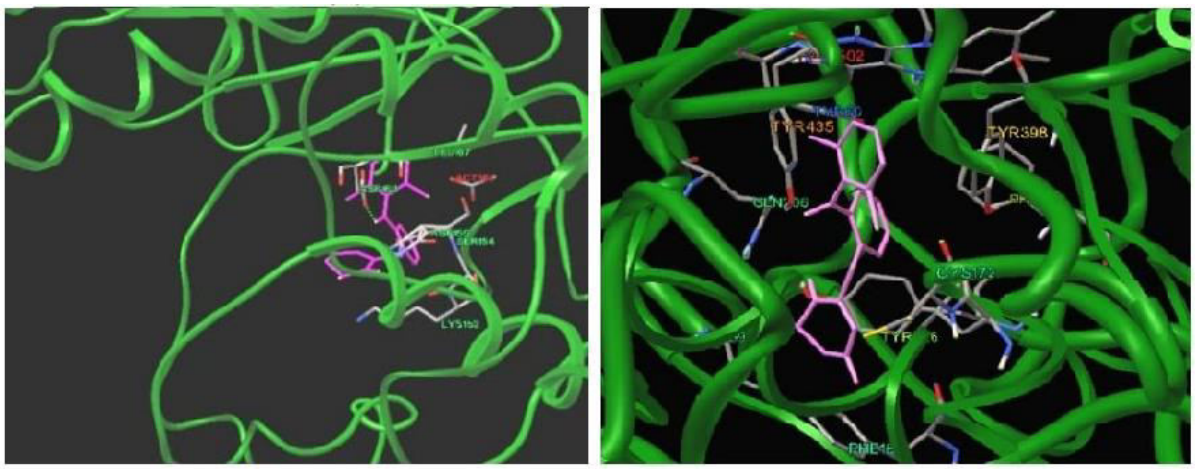


Figure 14: The synthesis scheme of 1, 2, 4-triazin analogue of diclofenac.(62)

Researchers believed that 1,2,4-triazin derivative probably has some appropriate anti Parkinsonian properties by antagonism: the Adenosine A2 receptor is expressed in the basal ganglia where it functionally opposes the actions of the dopamine D2 receptor. Inhibition of the A2 receptor leads to enhancement of D2 receptor functions. Hence in this study carboxyl group of diclofenac was replaced by addition of 2-chloroacetamide and produced 1,2,4-triazin derivative of diclofenac.

The molecular docking of 1,2,4-triazin derivative of diclofenac was studied against 5 major targets that are : 1-Dopamine receptor D3 protein (3PBL), 2 -Dopa decarboxylase-DDC (1JS3), 3-Adenosine A2 receptorAA2AR (3EML), 4-P38 map kinase (2ZAZ), 5-Monoamine oxidase-B "MAO-B" (2V5Z) enzymes targets with the help of the Autodock k4 program. In this study, MAO-B, MAPK (Cf. figure 2.9) show high affinity involved in this target and D3 protein, DDC, AA2AR, shows little binding affinity when compared to the MAO-B, MAPK (Cf. table IX).



(D) (E)

Figure 15: Docking study of 1,2,4-triazin derivative of diclofenac against (D) P38 MK and (E) MAO-B targets.(62)

Table IX: Docking score of 1,2,4–triazin derivative of diclofenac against parkinson enzyme targets.(62)

Target Name	Docking Score (Kcal/Mol)
Dopamine receptor	-6.35 to -5.64
Dopa decarboxylase	-6.86 to -5.81
Adenosine A2 receptor (AA2AR)	-6.86 to -5.81
P38 map kinase (MAPK)	-8.67 to -4.63
Monoamino oxydase-B (MAO-B)	-10.25 to -6.76

Results revealed that best fit of ligand against active site and the docking scores are P38 map kinase (-8.67) and MAO-B (-10.25). The synthesis 1,2,4-triazin analogues of diclofenac showed improvement in binding affinity and potent protective effect against the risk of PD.

CHAPTER III: Anticancer properties of Diclofenac derivatives

III.1. Definition

A tumor is a mass or lump of tissue that may resemble swelling. Not all tumors are cancerous, but it is a good idea to see a doctor if one appears. Tumors may be benign (not cancer) or malignant (cancer). Benign tumors may grow large but do not spread into, or invade, nearby tissues or other parts of the body. Malignant tumors can spread into, or invade, nearby tissues. They can also spread to other parts of the body through the blood and lymph systems.(63)

III.2. Physiopathology

Cancer is an uncontrolled proliferation of cells in the body. This proliferation leads to the formation of a mass called a tumor. It will gradually invade the organ in which it was born, affecting its functioning.(64)

In addition, cells can escape from this mass, diffuse throughout the body and lead to the formation of secondary tumors. We then speak of metastases.(64)

Cancers arise from initially healthy and functional cells that have become abnormal as a result of the build-up of alterations in their genetic makeup (DNA). These alterations (or mutations) will lead to deregulation, or even the inactivation of the systems that normally allow the control of cell division. The cell then becomes capable of proliferating in an anarchic manner leading to the formation of a tumor.(65)

Healthy cells have a system to detect DNA damage and repair it. When an abnormality is detected, cell division is momentarily stopped to allow its repair. If the lesion cannot be repaired, the cell sets off a program that leads to its death. This phenomenon called "apoptosis" is likened to cell suicide.(65)

Everything is complicated when DNA damage affects the regions themselves involved in these processes; if mutations appear in the genes necessary for the detection of abnormalities or for triggering the cell suicide program, the cell will continue to isolate itself and divide anyway. This is the first step in transforming a healthy cell into a cancer cell.(65)

The accumulation of other abnormalities will then lead this cell to lose its initial function and acquire properties allowing it to give rise to a malignant tumor, capable of developing to

the detriment of surrounding healthy cells and migrating to other regions of the cell of the body.(66)

III.3. Treatments

Anticancer drugs are a group of drugs that are effective in the treatment of malignant or cancerous disease (tumours). There are many different classes of anticancer drugs including antimetabolites, hormones and alkylating agents.(67)

Alkylating agents are a class of antineoplastic or anticancer drugs which act by inhibiting the transcription of DNA into the ribonucleic acid “RNA” and thereby stopping the protein synthesis. Alkylating agents substitute alkyl groups for hydrogen atoms on DNA, resulting in the formation of cross links within the DNA chain and thereby resulting in cytotoxic, mutagenic, and carcinogenic effects.(68)

Platinum (Pt) is a chemical element classified as a transition metal with several degrees of oxidation, the most common of which are 0, +2 and +4. The complexes are differentiated by the number of ligands surrounding the metal.(69)

The two major types of platinum complexes are :

- Pt-II complexes: they have a plane geometry: the central platinum atom is surrounded by 4 ligands (L1 to L4), arranged according to a square plan. This is the structure found in most platinum complexes used in clinics.(70)
- Pt-V complexes: are characterized by 6 ligands and have an octahedral structure. Relative to Pt-II complexes, the two additional ligands (L5 and L6) are arranged along an axis perpendicular to the plane square defined by the first 4 ligands.(71)

Platinum complexes are well-known for their antitumor activities. Cisplatin, Carboplatin and Oxaliplatin (Cf. figure 16) have entered clinical application and nedaplatin, lobaplatin, and heptaplatin have evolved into clinical drugs regionally. Ideally, platinum drugs should kill cancerous cells exclusively without harming the normal ones. Practically, however, the tumor selectivity of platinum drugs is quite poor. Thus, systemic toxicity and drug resistance become the major defects of platinum drugs.(72)

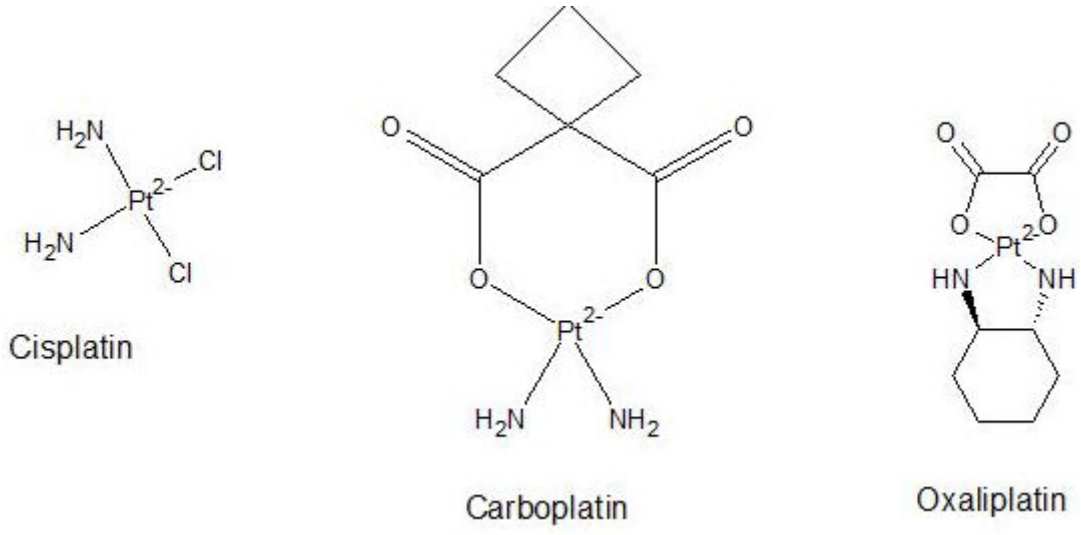


Figure 16: Chemical structures of Cisplatin, Carboplatin and Oxaliplatin.(72)

The development of cisplatin as a successful antitumor drug is often seen as the prototypical success story for platinum complexes. The large number of patients who have been cured after cisplatin treatment of cancer is impressive. However, the fact that the precise mechanism of action remains elusive has resulted in great interest in metal DNA binding generally and cisplatin and its analogs' binding properties particularly. As a consequence, cisplatin chemistry has provided a fertile ground for exciting bioinorganic chemistry research.(73)

Oxaliplatin is a platinum-based chemotherapy drug in the same family as cisplatin, it is used together with other cancer medications to treat colon and rectal cancer. Carboplatin is a key therapy for ovarian and breast cancers, and is generally used in combination with other medicines. Carboplatin can be given to patients who are intolerant to cisplatin due to kidney failure, nausea and vomiting, hearing loss, or neuropathy.(74)

III.4. Structure Activity relationship of novel anticancer derivatives of diclofenac-platinum complexes (75)

In 2017, Francesco Paolo Intini *and al*. have reported the synthesis of DCF-Pt-II derivatives consists in the derivatization of diclofenac with a diamine-linker to obtain a primary amine able to coordinate to the electrophilic Pt-II center. For this purpose, they selected 1,2-ethylenediamine. The reactionnal scheme of derviatives synthesized from Diclofenac is detailed in figure 17 as follows: DCF-en, compound 1: cis-[PtCl2 (DCF-en) (NH3)], compound 2: trans [PtCl2 (DCF-en)(DMSO)], compound 3: cis-[Pt(DCF) 2(NH3)2].

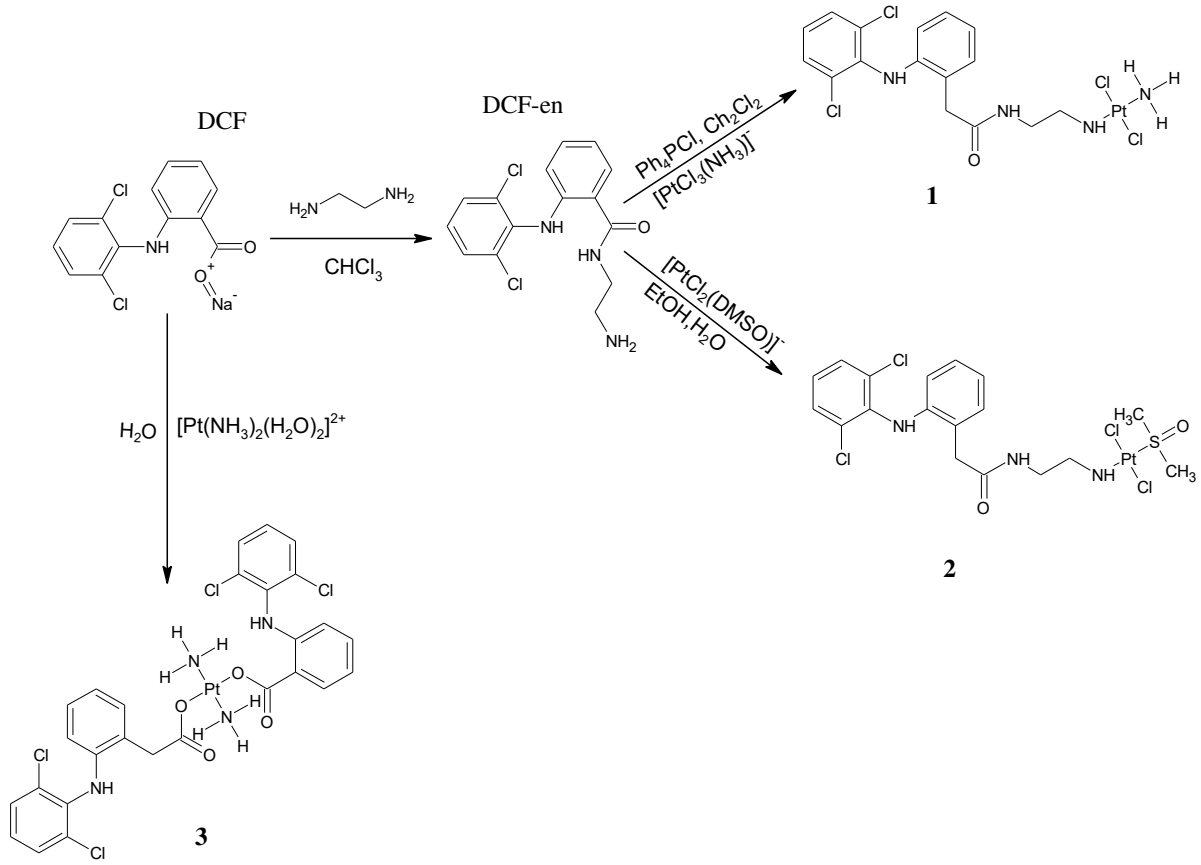


Figure 17: Chemical synthesis of compounds «1 », « 2 » and « 3 » from diclofenac.(75)

After that, the cytotoxic activity quantified by measuring the half maximal inhibitory concentration « IC50 » of these four compounds was tested on normal human cells (MRC-5 pd30), on cisplatin-sensitive cell line (cancer cells) and on cisplatin-resistant cell line (A2780cisR) while being compared to the molecule of reference (cisplatine).

The IC_{50} value of compound number 3 in normal human cells (MRC-5 pd30) is more than four times greater than the one found for the sensitive cancer cell line (A2780), it is also significantly more potent than cisplatin in a cisplatin resistant tumor cell line A2780cisR (cisplatin-resistant variant of A2780 cells). Thus, compound 3, in comparison with cisplatin, reveals selectivity toward tumor cells relative to non tumorigenic normal cells.(Cf. table X).

Table X: Cytotoxicity (IC50 Mean Values a, μM) of compound 3 and cisplatin.(75)

	Cancer cell line					Normal cell line	Resistance factor
	HeLa	HT-29	A2780	A2780cisR	SW480	MRC-5 pd30	RF
3	3.6±0.1	4.4±0.1	1.61±0.03	3.16±0.03	2.8±0.2	6.61±0.09	1.96
Cisplatin	22.8±0.9	13.3±0.5	3.1±0.1	16.7±2.2	2.6±0.2	3.07±0.03	5.39

The capability of compound 3 to affect tumor cells resistant to cisplatin (A2780cisR) when compared to this latter is remarkable. In addition, the humble cytotoxicity of 3 in noncancerous cells derived from human lung tissue (MRC-5 pd30) deserves special watchfulness. The resistance factor in DCF-containing complexes (namely compound 3) was also significantly lower compared to that of cisplatin.

Finally, we conclude that compound 3 may be soon recognized as a promising approach to improving the therapeutic index of platinum anticancer agents in contrast to free cisplatin.

PRACTICAL SECTION

Materials and Method

I.1 Introduction

The aim of the experimental study that was done in the laboratory of Therapeutic Chemistry at the faculty of medicine of Tlemcen from December 2020 to Septmber 2021.

The experimental study concerns the chemical synthesis of**:**

- Diclofenac ethyl acetate.
- Diclofenac phenyl hydrazine.

I.1.1. The Identification and the estimation of the purity of the synthesized products; using the following methods:

- Solubility measurement.
- Melting point measurement.
- Thin Layer Chromatography « TLC » (for monitoring).
- UV spectroscopy (Laboratory of Toxicology – faculty of Medicine Tlemcen)
- Infrared (IR) spectroscopy (Laboratory of Macromolecules – faculty of Science Tlemcen).

The following figure sum up the general chemical synthesis reaction:

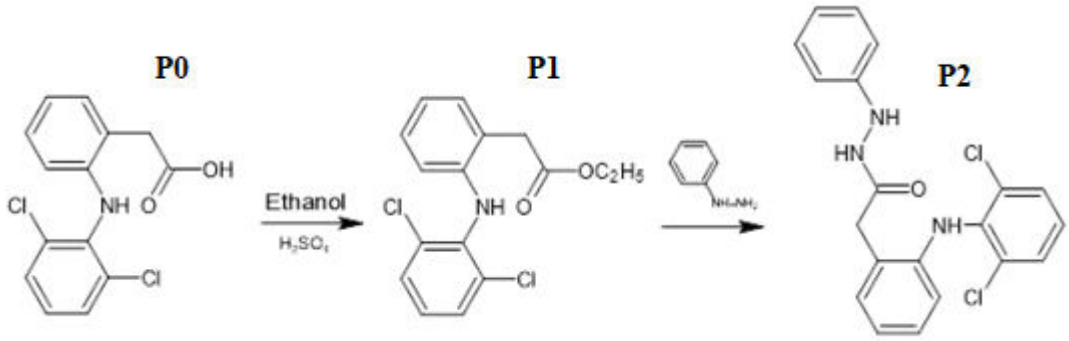


Figure 18: General scheme of the synthesis of diclofenac ethyl acetate and diclofenac phenylhydrazine.

I.2. Chemical synthesis

I.2.1. Chemical synthesis of diclofenac ethyl ester

To synthesize diclofenac ethyl acetate (ester) from the powder of diclofenac sodium, a preliminary reaction of acidification is required to transform diclofenac sodium to diclofenac acid.

The acidification reaction (Step1) was achieved by the dissolution of diclofenac sodium in ethanol. This reaction was followed by the esterification (Step2) of diclofenac acid to obtain **P1:** diclofenac ethyl ester.

Diclofenac ethyl ester appears as a pink crystalline powder due to its previous presence in an acidic medium Hence the need to operate purification by recrystallization in vacuum.

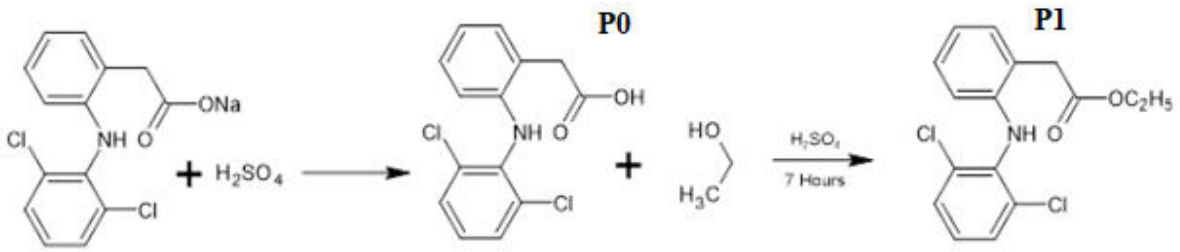
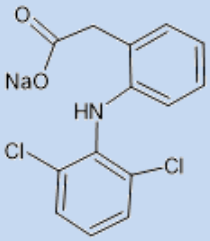
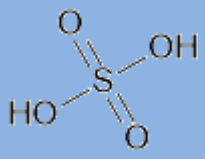
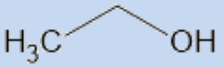
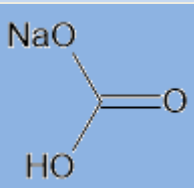


Figure 19: General scheme of synthesis of diclofenac ethyl ester.

I.2.1.1. Reagents

The following table reports the reagents used for the synthesis, isolment and purification of the synthesized powder:

Table XI: The reagents used for the synthesis, isolment and purification of diclofenac ethyl ester.

Steps	Reagents	Chemical structure	Laboratory	Physico-chemical characteristics
Synthesis	Diclofenac sodium salt CAS: 15307-79-6		SIGMA LIFE SCIENCE	MW=318.13 g/mol MP=275-277°C
	Sulfuric acid CAS 7664-93-9		HONEY WELL FLUKA	MW=98.08g/mol d=1.840g/ml in 25°C
	Ethanol 96° CAS : 64-17-5		EMSURE	MW=46.07g/mol MP=78,5°C
Isolement	Sodium bicarbonate CAS 144-55-8		SIGMA ALDRICH	MW=84.01
Purification	Methanol CAS 67-56-1		SIGMA ALDRICH	MW = 32.04 BP= 64.5 °C
	Distilled water			

I.2.1.2. Materials

Table XII reports the materials used in the purification of the synthesized powder.

Table XII: The materials used for the synthesis and purification of diclofenac ethyl ester.

Steps	Materials	Tools
Weighing	Counting scale Gibertini Model Max :1010 g E =0.01 g T = - 1010 +15°C/+30°C	Measuring cylinder 50ml Beaker 50 ml Spatula
Synthesis	**Reflux heating :** Refrigerant Heating mantle: Wise Therm brand Pipes gallows **Thermometer** : ZEAL , -10°C/ +150°C, 76 mm Beaker 250 ml	Pipette 2 ml Pipette pump 2 ml Magnetic bar pH paper
Purification	ARE Heating **Magnetic Stirrer/** VELP SCIENTIFICA Ice bath **Oven** : Model JOUANSA 240±10 V 1000W 50/60Hz	Measuring cylinder Crystallizer Schlenk flask Büchner 80mm Conical seal Water pump Beaker 250 ml Spatula Filter paper

I.2.1.3. Operating method

This method was described and applied in 2006 by Dharmarajan Sriram *and al.* (10) and Sudhakar P *and al*. in 2018.(62)

Like any ordinary synthesis, P1 is obtained first chemically following the step 1 of acidification to obtain P0 and step 2 of esterification, then isolated from the reaction mixture and finally purified.

I.2.1.3.a. Synthesis of diclofenac acid

Figure 20 illustrates the pictures that of the synthesis of diclofenac acid P0.

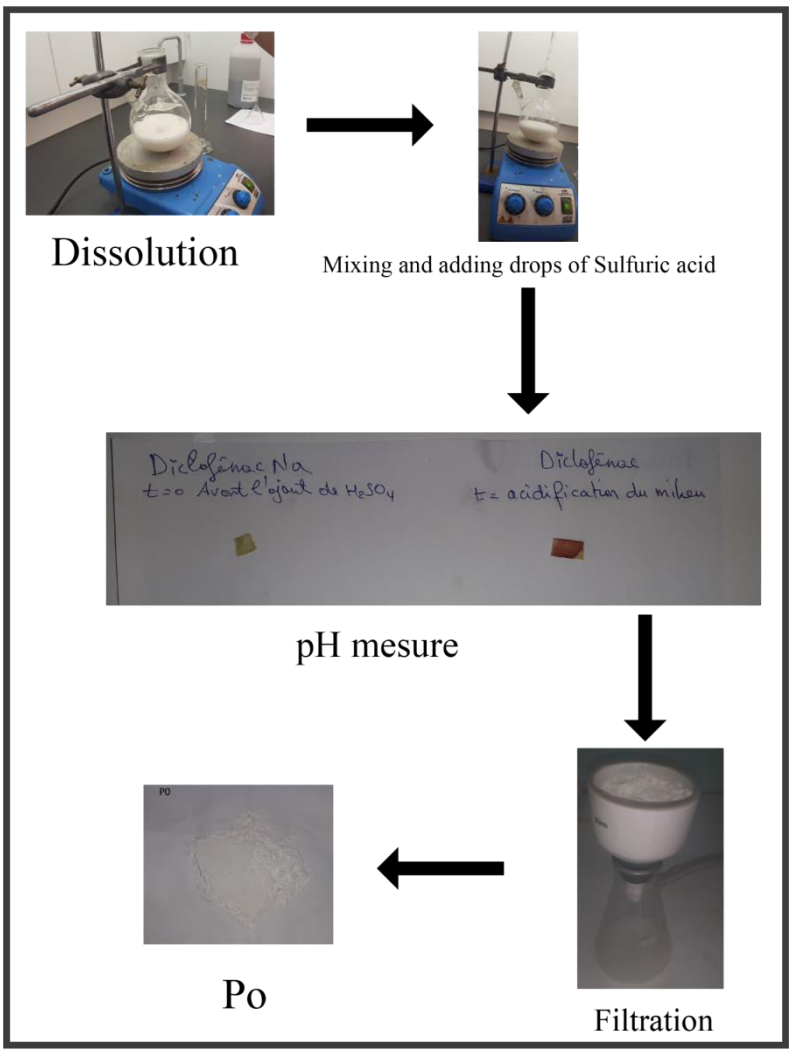


Figure 20: Pictures illustrating the synthesis of diclofenac acid (P0).

Method

This method was described and applied in 2006 by Dharmarajan Sriram *and al.*(10) and Sudhakar P *and al*. in 2018.(62)

- In a 500 ml schlenk flask, completely dissolve 32.13 g (0.101 mol) of diclofenac sodium salt, in 150 ml of 96° ethanol (2.5mol).
- 3.85 ml were versed in the reaction mixture gradually.
- The solution was then mixed for 2 hours using the ARE heating magnetic stirrer at room temperature.
- The pH was measured to ensure the mixture is acid.
- The acid obtained was filtered in vacuum.
- The powder was then dried for 90 minutes in the oven at 90°C.

I.2.1.3.b. Synthesis of diclofenac ethyl acetate

Figure 21 illustrates the pictures that of the synthesis of diclofenac ethyl ester (P1).

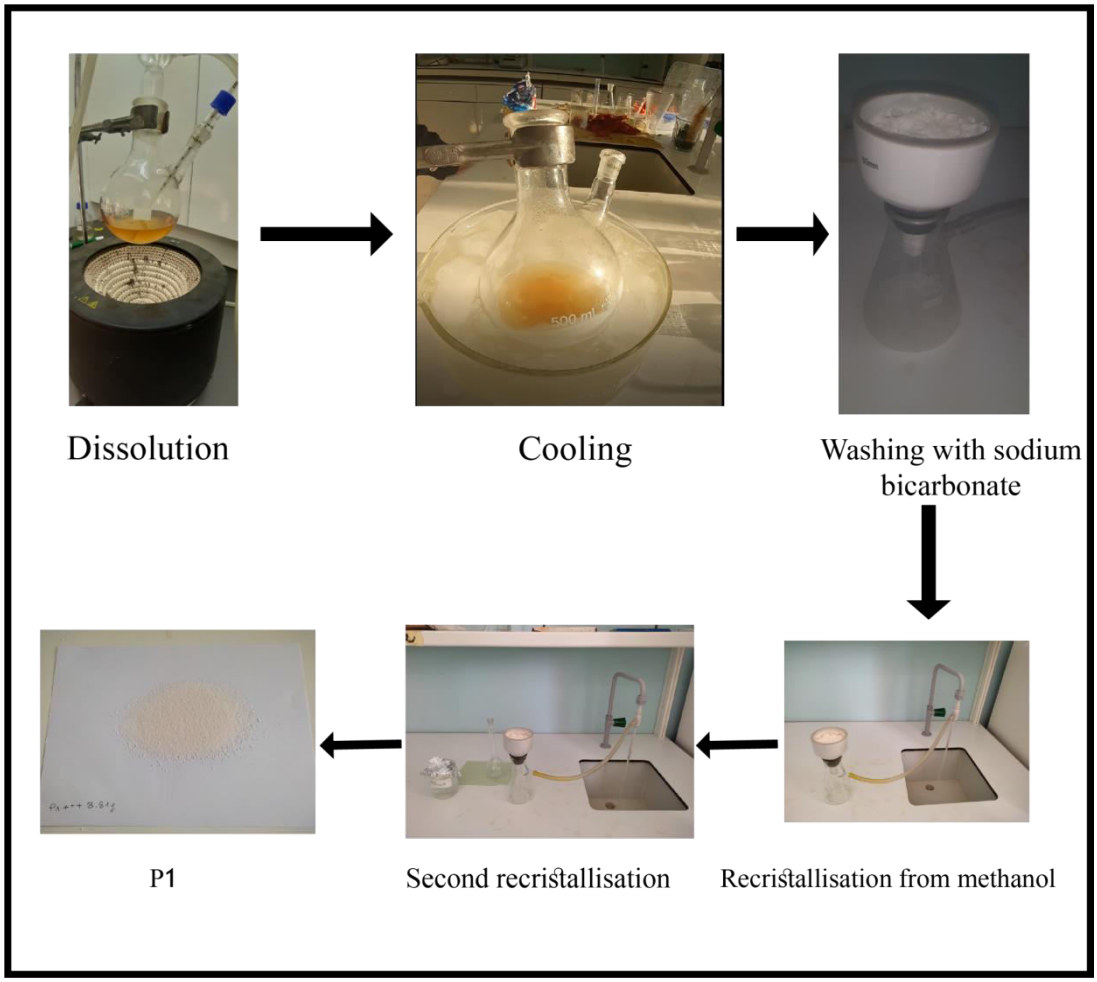


Figure 21: Pictures illustrating the synthesis of diclofenac ethyl acetate (P1).

Method

This method was described and applied in 2006 by Dharmarajan Sriram *and al.*(10) and Sudhakar P *and al*. in 2018.(62)

1. Synthesis

- In a 500 ml schlenk flask, add 29.61 g (0.1 mol) of diclofenac acid to 90 ml of 96° ethanol.
- 2 ml of sulfuric acid was additionned to the solution.
- The mixture was heated under reflux using heating mantle for 7 hours.
- The reaction procedure was followed with TLC.
- Reaction mixture gave on processing ethyl ester.

2. Isolement

The solution of P1 was cooled using tap water, then placed in an ice bath and filtered in vacuum, washed with a solution of sodium bicarbonate 10% (150ml) and 5% (200ml) respectively then dried for 30 min in the oven at 40°C.

Preparation of 150 ml of sodium bicarbonate 10%

- 15 g of sodium bicarbonate was weighed and placed in a graduated flask, distilled water was additioned until reaching the 150 ml line. The solution was then mixed until complete dissolution.

Preparation of 200 ml of sodium bicarbonate 5%

- 10 g of sodium bicarbonate was weighed and placed in a graduated flask, distilled water was additioned until reaching 200 ml line. The solution was then mixed until complete dissolution.

3. Purification

The retrieved powder appeared somewhat pink hence the need to recrystallize it.

Recristallisation from methanol

- 10g of impure P1 was mixed with 40 ml of methanol.
- The mixture was heated using the heating magnetic stirrer.
- The solution obtained was after that cooled using tap water, and then placed in an ice bath.
- The resulting mixture was filtered in vacuum to give pure P1.

- A second recristallisation of 8g of impure P1 was performed of the first recristallisation filtrate.

I.2.2. Chemical synthesis of diclofenac phenylhydrazine (P2)

In order to obtain diclofenac phenylhydrazine P2, the ester ought to be replaced with phenylhydrazine group as described in figure 22.

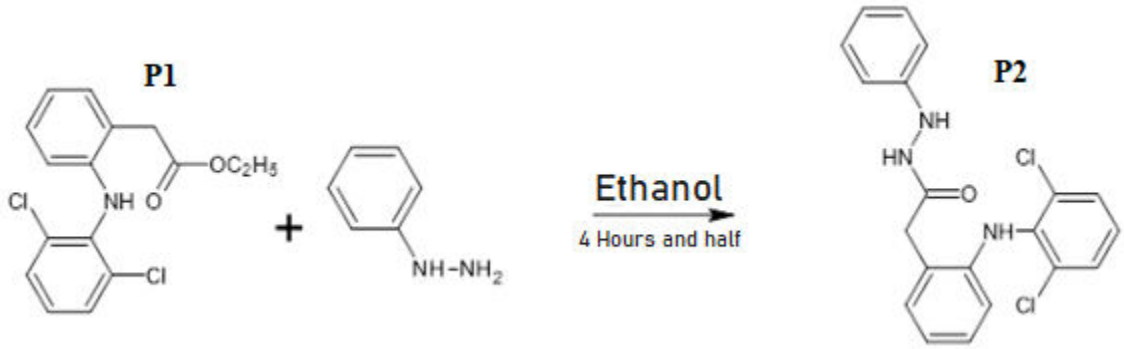
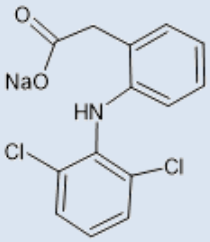
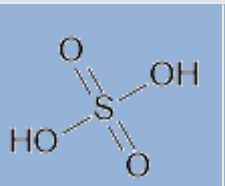


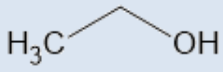
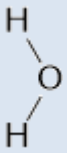
Figure 22: Scheme of chemical synthesis of diclofenac phenylhydrazine.

I.2.2.1. Reagents

The following table summarizes the reagents used in the synthesis and the purification of the synthesized powder.

Table XIII: The reagents used for the synthesis and purification of diclofenac phenylhydrazine.

Steps	Reagents	Chemical structure	Laboratory	Physico-chemical characteristics
Synthesis	Diclofenac ethyl ester		Synthesized	Synthesized MW= 324 g/mol
	Phenylhydrazine		HONEY WELL FLUKA	MW=98.08 g/mol d=1.840g/ml in 25°C

	Ethanol 96° CAS 64-17-5		EMSURE	MW= 46.07 g/mol MP=78,5°C
Purification	Methanol CAS 67-56-1		SIGMA ALDRICH	MW = 32.04 g/mol BP= 64.5 °C
	Distilled water			

I.2.2.2. Materials

Table XIV summarizes the materials used for the synthesis and the purification of P2:

Table XIV: Table of tools and materials used for the synthesis and purification of diclofenac phenylhydrazine.

Steps	Materials	Tools
Weighing	Counting scale Gibertini Model Max :1010 g E= 0.01 g T = - 1010 +15°C/+30°C	Measuring cylinder 50ml Beaker 50 ml Spatula
Synthesis	**Reflux heating :** Refregirant Heating mantle: Wise Therm brand Pipes gallows **Thermometer** : ZEAL , -10°C/+150°C, 76 mm Beaker 250 ml	Pipette 2 ml Pipette pump 2 ml Magnetic bar pH paper

Purification	ARE Heating **Magnetic Stirrer/** VELP SCIENTIFICA Ice bath **Oven** : Model JOUANSA 240±10 V 1000W 50/60Hz	Measuring cylinder Crystallizer Schlenk flask Büchner 80mm Conical seal Water pump Beaker 250 ml Spatula Filter paper

Method

To perform this reaction, 0.02 mol of phenylhydrazine is needed. Meanwhile, in the laboratory the phenylhydrazine 97° is the only available. Thus, the rate conversion is mandatory (Cf. figure 23).

0.02 mol is equivalent to 1.97 ml of absolute phenylhydrazine

Phenylhydrazine 97° → 1.97 ml

Phenylhydrazine 100°→ V

$V=1.97\times100/97 = 2.03$ ml

- **Synthesis**

- In a 500 ml schlenk flask, add 3.24 g (0.01 mol) of diclofenac ethyl ester in 55 ml of 96° ethanol.
- 2 ml of phenyl hydrazine 97° was added to the solution.
- The mixture was heated under reflux using the heating mantle for 4 and half hours at a temperature between 62 and 75°C.
- The reaction procedure was followed with TLC.
- The mixture was concentrated to half using the heating stirrer.
- In a 250ml beaker, the concentrated mixture was cooled with tap water and poured in ice cold distilled water.

➢ **Purification**

The solid thus precipitated out was filtered in vacuum, dried in the oven for 45 minutes at 50°C and recrystallized twice from ethanol 96° after that due to its bright yellow coloration.

Recristallisation from ethanol

- 6 grams of impure P1 were mixed with 40 ml of ethanol.
- The mixture was heated using the heating magnetic stirrer.
- The solution obtained was after that cooled using tap water, and then placed in an ice bath.
- The resulting mixture was filtered in vacuum to give pure P2.
- A second recristallisation was performed on 5g of P2 this time from 35ml of ethanol.

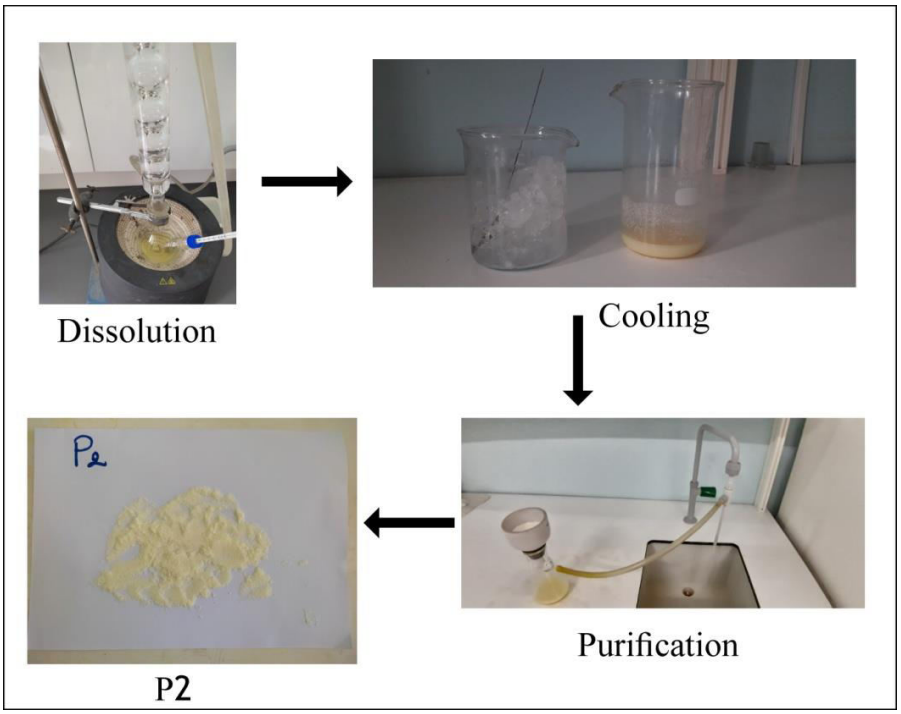


Figure 23: Chemical synthesis and purification of P2.

I.3. Identification

The identification of synthesized products is carried out by several methods including solubility measurement, melting point measurement, thin layer chromatography (TLC), UV spectroscopy and infrared spectroscopy.

In this section, the two products P2 and P1 are identified by these methods.

I.3.1. Solubility measurement

Solubility is a crucial physico-chemical parameter affecting the whole process of drug discovery and development.

I.3.1.1. Principle

Solubility is defined as the amount of substance that passes into solution to achieve a saturated solution at constant temperature and pressure. Solubility is expressed in terms of maximum volume or mass of the solute that dissolves in a given volume or mass of a solvent. It is expressed in $mol.L^{-1}$.

The solubility product is - in the case of an ionic solid compound - the equilibrium constant of the dissolution reaction. This constant is denoted Ks (T). It depends only on the temperature T and, in general, it increases with this one for the solids and decreases for the gases.

Solubility can be measured quantitatively or qualitatively with the naked eye on different liquids.

I.3.1.2. Reagents

Three liquids were used to test the solubility of P1 and P2:

- Distilled water.
- Ethanol.
- Acetic acid.

I.3.1.2. Method

Solubility of P1 and P2 was measured qualitatively.

In a series of hemolysis tubes, under constant pressure and temperature, mix the synthesised products with every solution and observe the results with the naked eye.

I.3.2. Melting point measurement

The two synthesized products (P1 and P2) have been identified by melting point measurement at the laboratory using the Kofler bench.

I.3.2.1. Principle

The temperature at which a solid substance transitions to the liquid state is called the melting point.

The presence of impurities in the compound results in a melting point's changement. Although this value is characteristic of a compound and makes checking its purity possible, it's not sufficient for the identification to take place.

Kofler bench

A Kofler bench is a metal strip with a temperature gradient (that ranges from room temperature to 260°C). Any substance can be placed on a section of the strip inorder to reveal its thermal behaviour at this exact temperature.

The gradient is engineered to be approximately linear.

I.3.2.2. Material

- Kofler bench: brand: HEIZBANK. Power: 100w. Power supply: 220V-50 / 60Hz. Temperature: + 50 ° C to + 260 ° C.
- Micro spatula.

I.3.2.3. Method

- Calibrate the device beforehand with the standard substances provided with it.
- The bench must be switched on sufficiently in advance so that its temperature in all points is stable.
- Clean the device with a cotton ball soaked in acetone.
- Place the synthesized product at the cold end of the bench and induce it, thanks to the micro-spatula, towards the hot zone until a marking line is observed from which the fusion begins.
- Drag the cursor and read the indicated temperature.

I.3.3. Thin Layer Chromatography

Thin Layer Chromatography (TLC) is a technique used to isolate non-volatile mixtures.

I.3.3.1. Principle

The experiment is conducted on a sheet of aluminium foil, plastic, or glass which is coated with a thin layer of adsorbent material. The material usually used is aluminium oxide, cellulose, or silica gel.

This separation is based on the difference in affinity of the different constituents between the two phases: stationary (fixed) and mobile one.

The stationary phase is fixed on a plate, and the liquid mobile phase, called the eluent. This eluent represents a solvent or a mixture of solvents. Each constituent migrates from one certain height characteristic of the substance, which is called frontal ratio (Rf).

Rf =Spot height/Solvent height

Each stain (spot) corresponds to a constituent, it can appear:

- To the naked eye: if the product is colored.
- Under a UV lamp (aromatic nuclei for example).
- With a chemical developer: $KMnO_4$, KOH ... if the product is not colored**.**

I.3.3.2. Objective

Execute a separate analysis of the products of the two synthesized produced compounds.

- P1: Diclofenac ethyl ester.
- P2: Diclofenac phenyl hydrazine.

I.3.3.3. Material

For the TLC identification method, we used the materials described in Table XV below:

Table XV: Materials used for the chromatographic separation.

Material	UV Lamp	TLC
Description	**VILBER LOURMAT :** 4 W-365 nm tube, 4W- 254 nm tube, 8W	TLC Silica gel 60 F254
	Chromatographic tank	Hemolysis tubes Pasteur pipette
Tools	-	Rack Spatula Beaker

I.3.3.4. Method

Eight TLC experiments were carried out for the monitoring during multi-phases (T0, T1, T2 and T3) of the synthesis of P1 and P2.

Results with eluents A and B were not satisfying for P1, consequently they were not implemented at T1 and T2.

Results with eluent A were not satisfying for P2, consequently they were not implemented at T1 and T2 and replaced with eluents D and E.

Preparation of eluents

Add other eluents

Eluent **A**: Ethyl acetate/petroleum ether 7:3 (v/v) for the synthesis of P1 and P2.
Eluent **B**: Ethyl acetate/petroleum ether 5:5 (v/v) for the synthesis of P1 and P2.
Eluent **C**: Ethyl acetate/petroleum ether 3:7 (v/v) for the synthesis of P1 and P2.
Eluent **D**: Ethyl acetate/petroleum ether/Acetic acid 3:6:1 (v/v/v) for the synthesis of P2.
Eluent **E**: Ethyl acetate/petroleum ether/Acetic acid 3:5:2 (v/v/v) for the synthesis of P2.

- **Protocol**

- Fill the beaker with the eluent; cover it, so that the atmosphere in the beaker is saturated with eluent vapors.
- Draw a fine line with a pencil at 1 cm on the chromatographic plate from the bottom edge: this is the deposit line.
- Using Pasteur pipettes, take a drop of P1 and P2 and lay it on the bottom edge. Allow to dry in the open air.
- Place the plate vertically in the beaker and cover it.
- After good elution, remove the plate from the beaker and let it dry.
- Observe the plates in the open air and then under the UV lamp (254 nm).
- Circle the spots then calculate the frontal ratios Rf.

I.3.4. Ultraviolet spectroscopy

Ultraviolet-visible (UV-Vis) spectroscopy is one of the most popular analytical techniques because it is very adaptable and able to detect roughly every molecule.

I.3.4.1. Principle

The Principle of UV-Visible Spectroscopy is based on the absorption of ultraviolet light or visible light by chemical compounds, which results in the production of distinct spectra.

When matter absorbs UV radiation, the electrons present in it go through excitation. This gets them to jump from a ground state (an energy state with a proportionately diminutive amount of energy associated with it) to an excited one (an energy state with a relatively enormous amount of energy associated with it). It is important to note that the difference in the energies of the ground state and the excited state of the electron is always equivalent to the amount of ultraviolet radiation or visible radiation absorbed by it.

With UV-Vis spectroscopy, the UV-Vis light is passed through a sample and the transmittance of light by a sample is measured. From the transmittance (T), the absorbance can be calculated as A= -log (T). An absorbance spectrum is obtained that shows the absorbance of a compound at different wavelengths. The amount of absorbance at any wavelength is due to the chemical structure of the molecule.

I.3.4.2. Material and reagents

- Biochrom WPA Lightwave II UV/Visible Spectrophotometer.
- Analytical balance.
- Distilled water.
- Ethanol 96°

I.3.4.3. Method

- Use the four arrow keys to navigate around the display and select the required setting from the active (highlighted) option.
- In a plastic cell, we poor, each time, the amount of product as mentioned in table XVI in the cell holder.
- Press Enter.

- Before each try, we use a cell filled with the solvent alone and press the reference button inorder set reference.

Table XVI: UV-Vis essays of P1 and P2.

Essays	Product experimented	Solvent	Concentration
E1	P1	Ethanol	21.67 mg/ml
E2		Ethanol	9 mg/ml
E3		Ethanol	33.67 mg/ml
E4		Distilled water	24.67 mg/ml
E5	P2	Ethanol	16.67 mg/ml

I.3.5. Infrared spectroscopy

Infrared spectroscopy is one of the most widely used spectroscopic tools for characterizing a molecule.

I.3.5.1. Principle

This analysis technique (non-destructive) is particularly fine: it allows us to characterize the bonds between atoms and their mode of vibration. We can thus perform the functional analysis of a molecule by determining all the groups of chemicals that constitute it.

However, this process does not make it possible to distinguish between them the enantiomers, nor the diastereomers.

The infrared range concerns wavelengths between 800 nm and 50 µm approximately. In practice, we work on a wavelength band between 2.5 and 25 µm. Infrared spectra do not use the wavelength λ but the wavenumber which is the reverse of it. The study area is therefore between 400 and 4000 cm-1.

The area of the spectrum between 1500 and 600 cm-1 contains several difficult bands to interpret: it is called the region of fingerprints. It is however useful for determine the identity of the compound. When the bands of this zone are identical, we have deal with the same molecule.

I.3.5.2. Material

- The machine used for generating the Infrared spectrum: Agilent Technologies Cary 600 series FTIR spectrometer.

I.3.5.3. Method

- The sample is placed on top of the machine's prism.
- The software coupled with it is launched.
- We press Enter to start the analysis.

Results and Discussion

II.1. Chemical synthesis

II.1.1. Synthesis of diclofenac ethyl ester P1

II.1.1.1. Aspect

- **Results**

After recrystallization, the purified diclofenac ethyl ester is in the form of a bright pink crystalline powder as figure 24 illustrates.

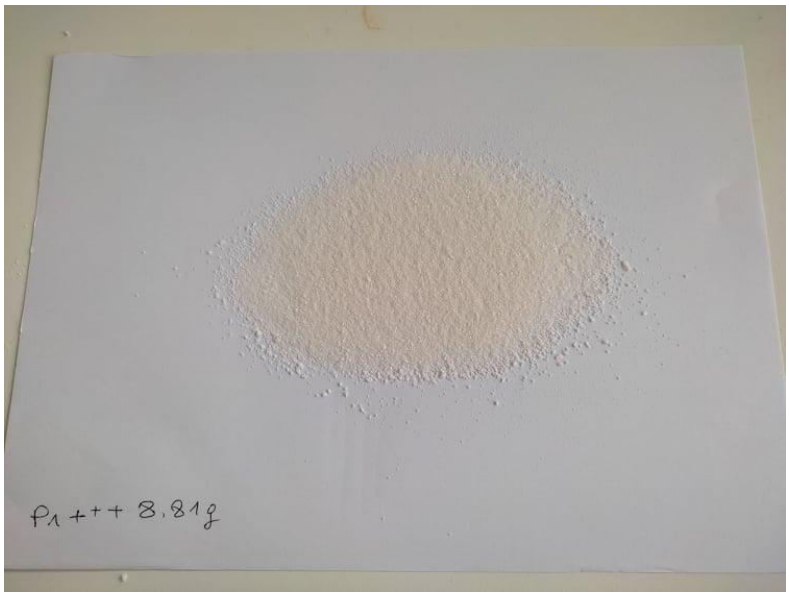


Figure 24: Aspect of recrystallised diclofenac ethyl ester.

- **Discussion**

The purification of diclofenac ethyl ester gave a white crystalline powder, which approximates the color of pure diclofenac ethyl ester.(78)

The initial (before recrystallisation) appearance was a pinkish powder, which explains the need for purification.

II.1.1.2. Yield calculation

- **Results**

Yield (Y) = (Practical measured mass of diclofenac ethyl ester / Theoric mass calculated of diclofenac ethyl ester) ×100

- **Weighing**

The measured mass of diclofenac ethyl ester after purification is 24.10 g.

- **Theoric mass calculation**

To calculate the theoric mass, we need to define the limiting reagent. The reaction of acidification of diclofenac sodium is defined as follows:

Diclofenac acid + Ethanol → Diclofenac ethyl ester + H_2O

1n 1n → 1n + 1n

29.61/296.13 71.01/46.07

0.1 mol 1.54 mol

Conclusion: diclofenac acid is the limiting reagent.

Calculating the theoric mass of diclofenac ethyl ester

0.1= m / 324.13 → m= 32.4 g

Yield (Y) = $\frac{24.1}{32.4} \times 100$= 74 %

➢ **Discussion**

Sudhakar *and al.* (2018) synthesized diclofenac ethyl ester with the same reagents (Diclofenac acid, Ethanol and sulfuric acid) for 22 hrs and recrystallized with the same solvent (methanol) and resulted a yield of 94 %.(62)

The synthetic yield of product P1 is 74%, it is less than that resulting from the work described above. This is probably due to:

- The reaction lasted for 7 hours.
- Temperature (62°-72°C).
- The nature of Büchner.
- The second phase of washing and recrystallising the powder.
- The amount of sodium bicarbonate used for rinsing the product.

II.1.2. Synthesis of diclofenac phenyl hydrazine P2

II.1.2.1. Aspect

➢ **Results**

After recrystallization, the purified diclofenac phenyl hydrazine is in the form of a yellowish crystalline powder (Cf. figure 25).

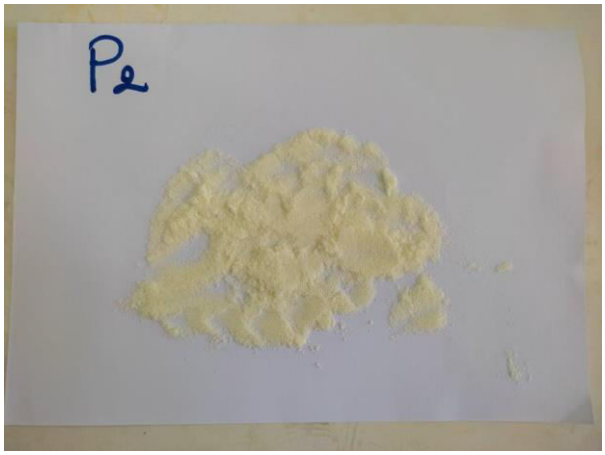


Figure 25: Aspect of recrystallised diclofenac phenylhydrazine.

➢ **Discussion**

The purification of diclofenac phenyl hydrazine gave a bright yellowish crystalline powder; the initial appearance was a darker yellowish powder, which explains the need for purification.

The yellow color is due to phenyl hydrazine substituent.(1)

II.1.2.2. Yield calculation

➢ **Results**

Yield (y) = (Practical mass measured of diclofenac phenyl hysrazine / Theoric mass calculated of diclofenac phenyl hydrazine) × 100

- **Weighing**

The measured mass of diclofenac phenyl hydrazine after purification is 6.9 grams.

- **Theoric mass calculation**

To calculate the theoric mass, we need to define the limiting reagent. The reaction of acidification of diclofenac sodium is defined as follows:

Diclofenac ethyl ester + phenyl hydrazine → Diclofenac phenylhydrazine + ethanol.

1n + 1n → 1n + 1n

3.24/324 3.68/98.08

0.1 mol 0.04 mol

Conclusion: phenyl hydrazine is the limiting reagent.

- **Calculation of the theoric mass of diclofenac phenyl hydrazine**

0.04= m/393.21 → m=15.75 g

Yield (Y) = $\frac{6.9}{15.75} \times 100 = 43\%$

➢ **Discussion**

The synthetic yield of product P2 is 43%, which is low. This is probably due to:

- Reactivity of phenyl hydrazine is less than reactivity of hydrazine hydrate, because of the conjugaison of the free double in the aromatic cycle, resulting in the diminution of its nucleophile reactivity, and then the reaction of addition of nucleophile did not happen.
- The amount of distilled water used for cooling the product.
- The length of the reaction 4 hours and half (the duration of the raction is 20 hours)
- Temperature (62°-75°C).

II.2. Identification

II.2.1. Synthesis of diclofenac ethyl ester

II.2.1.1. Solubility measure

➢ **Results**

The qualitative results of solubility of P1 are mentioned in the table and the figure below.

Table XVII: Solubility of P1 in different solvents.

Solvent	Distilled water	Ethanol	Acetic acid
Solubility	-	+	+

(-): insoluble (+): soluble (+/-): more or less soluble

Figure 26: Solubility results of P1 in different solvents (right).

➢ **Discussion**

Diclofenac ethyl ester is insoluble in distilled water because it's an organic impolar product. Diclofenac ethyl ester is soluble in ethanol because it's an organic impolar product that explains the choice of this solvent of the sythesis of P2.

Diclofenac ethyl ester is a basic product, it is then soluble in acid solvents such acetic acid.

II.2.1.2. Melting temperature measure

➢ **Results**

The measured melting point of pure diclofenac ethyl ester in the laboratory of therapeutic chemistry is 63° C.

The melting point of the starting reagent diclofenac sodium is from 272°C to 275°C (Cf. table XI).

➢ **Discussion**

The melting point of 95% pure diclofenac ethyl ester of J and K scientific LTD laboratory (CAS:15307-77-4) is in between 67-69°C.(62)

The melting point of the starting reagent diclofenac sodium is from 272°C to 275°C (Cf. table XI).

The vast transition of the melting point from around 275°C to around 63°C explains the satisfying results of the reaction of acidification followed by the reaction of esterification.

Experimental MP (63°C) approximative the reference MP (67-69°C), thus product P1 is probably diclofenac ethyl ester, subject to IR results.

II.2.1.3. Thin Layer Chromatography

- **Results**

Figure 27 illustrates the TLC results for diclofenac ethyl ester (P1) synthesis, observed on naked eye then under UV in eluents A, B and C at T1= 1h30mins.

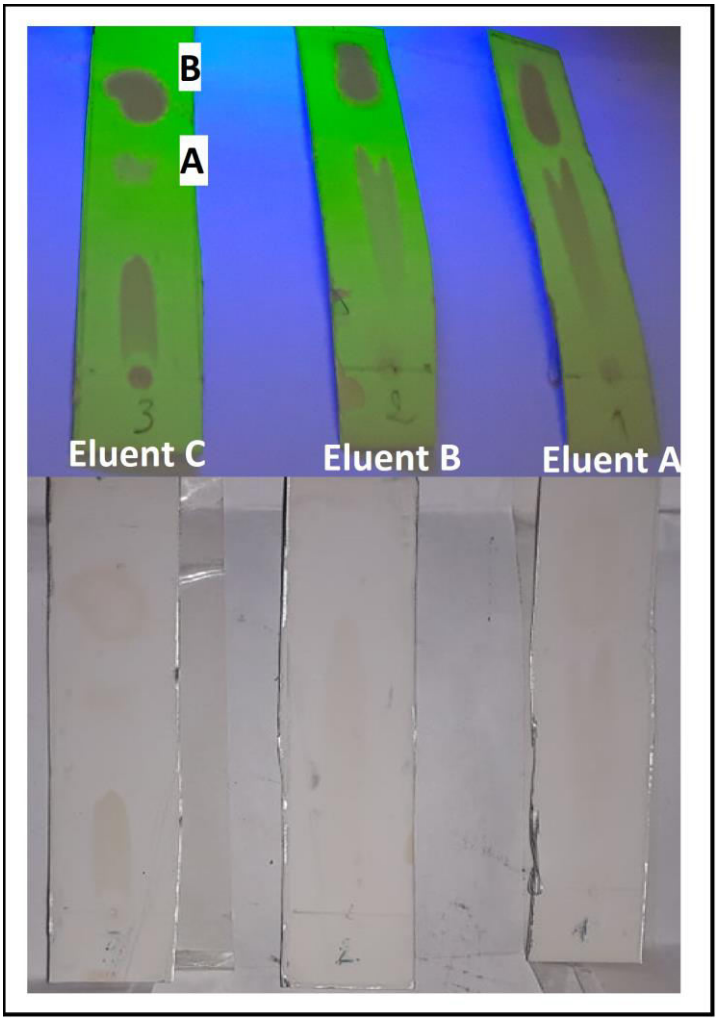


Figure 27: TLC results for diclofenac ethyl ester (P1) synthesis, observed on naked eye and under UV light in eluents A, B and C at T1.

Figure 28 illustrates the TLC monitoring for diclofenac ethyl ester (P1) synthesis results, observed on naked eye then under UV light in eluent C at T2, T3 and T4.

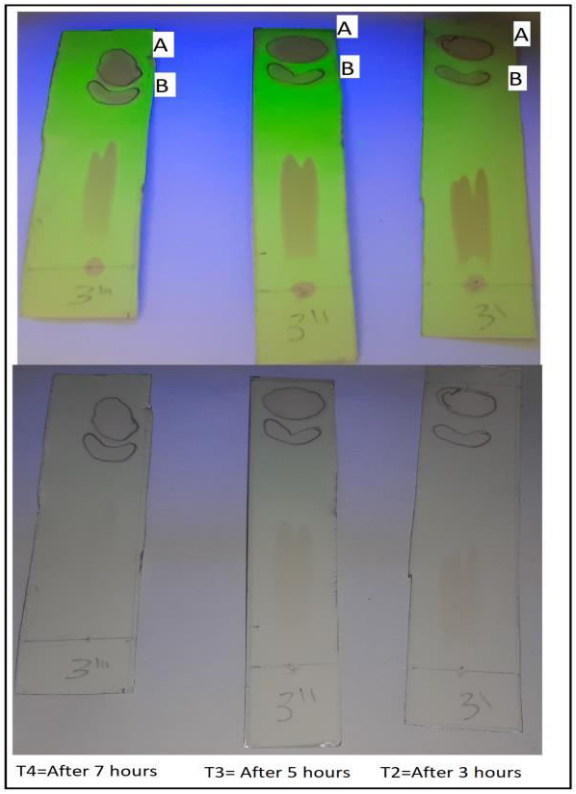


Figure 28: TLC results for diclofenac ethyl ester (P1) synthesis, observed on naked eye and under UV light in eluents A, B and C at T2, T3 and T4.

➢ **Solvent front**

Eluent C: The height of the solvent front is equal to 5.3 centimeters.Table XVIII explores the Rf results of the TLC issued at T1, T2, T3 and T4.

We define A as diclofenac acid and B as diclofenac ethyl ester (P1).And Rf results are expressed in the table below.

Table XVIII: Rf results from TLC monitoring in the eluent C during multiple phases.

Rf	A	B
T1 = After 1 hour and half	0.9	0.77
T2 = After 3 hours	0.916	0.783
T3 = After 5 hours	0.916	0.78
T4 = After 7 hours.	0.905	0.7

- **Discussion**

At T1

In eluent C results, we observe two separate spots: "A" of the starting reagent and "B" of the other product. Eluents A and B showed only one spot, it is a bad separation that's why we choose eluent C as the ideal one for monitoring the rest of the reaction. Rf results show two different values, each one corresponds to a different product.

At T2

We can still observe two spots A and B. But the intensity of B was increasing while that of A was decreasing. We conclude that a new product is being generated and the starting reagent is being consumed.

At T3

A spot is more intense and B spot is lighter than before. The Rf results of A are approximately the same, they are different from B results and correspond to the new generated product (P1).

II.2.1.4. Ultraviolet spectroscopy

- **Results**

The spectrums below illustrate the UV absorption of P1 in ethanol (Cf. figure 29) and in distilled water (Cf. figure 30). The results were as follows:

- **E1:** The maximum absorption was reached at 222 nm wavelength.
- **E2:** The maximum absorption was reached at 244 nm wavelength.
- **E3:** There were no wavelengths available on the table.
- **E4:** The maximum absorption was reached at 271 nm wavelength.

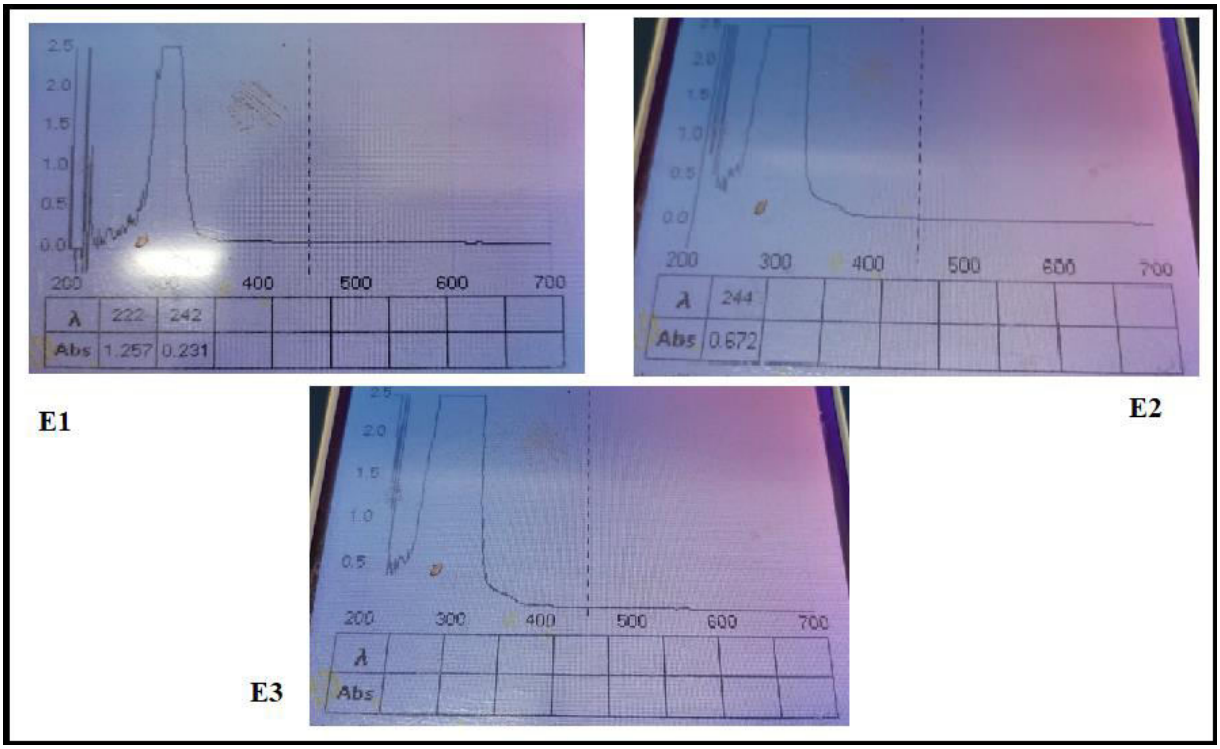


Figure 29: UV experimental spectrums for the first, second and third essays (E1, E2 and E3).

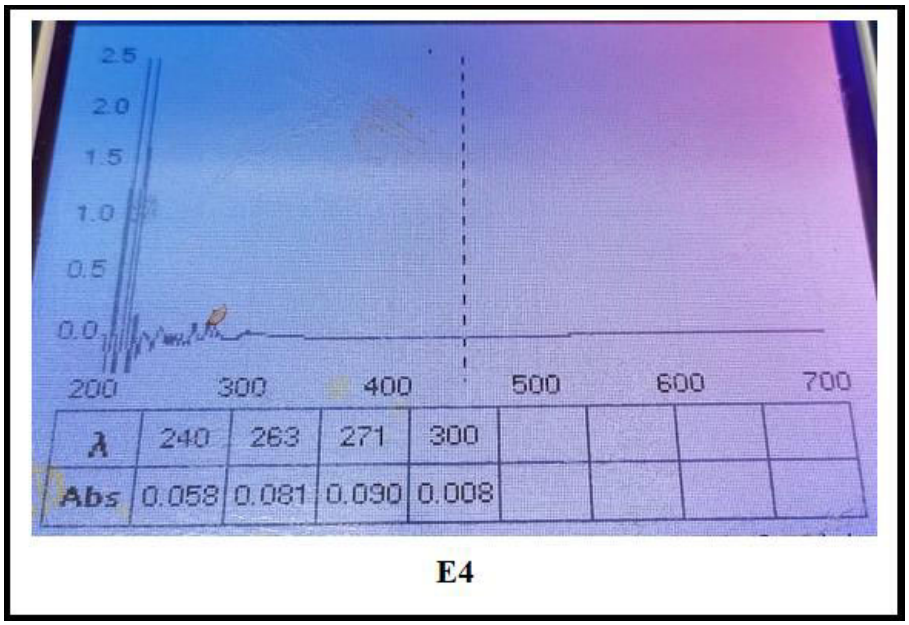


Figure 30: UV experimental spectrums of the fourth essay (E4).

- **Discussion**

Since all maximum absorption correspondant wavelengths values were between 200 and 300 nm, we are pretty sure there is a conjugated system in our product (phenyl ring).

II.2.1.5. Infrared spectroscopy

➢ **Results**

Figure 31 represents the practical IR spectrum of purified P1 (diclofenac ethyl ester).

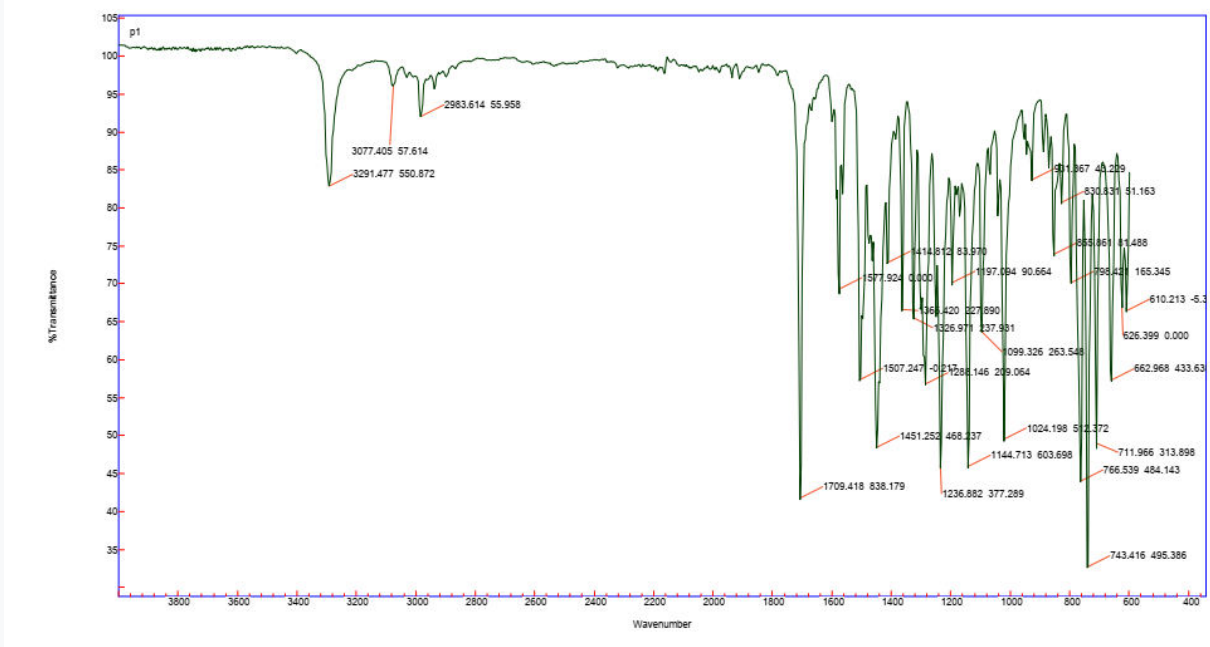


Figure 31: Practical IR spectrum of diclofenac ethyl ester (P1).

➢ **Discussion**

The allocation of the peaks of the IR spectra was made for the P1 based on infrared tables taken from the literature. The use of this latter and our practical spectrum made it possible to establish the following comparative table:

Table XIX: Comparative analysis of the IR experimental spectrum of P1 with reference spectrum..

	Reference spectrum	Practical frequency	Vibration mode
Functional Groups	3270	3291.477	N-H stretching
	2981	2983.614	-CH2-
	1720	1709.418	C=O

	1577	1577.924	C=C stretching
Digital Prints	1326	1326.971	C-N
	1238	1236.882	H-C-O
	1458	1451.252	-CH2-
	600-800	626.399	C-Cl
	600-800	610.213	C-Cl

The presence of certain characteristic bands on the practical spectrum confirms the existence of functional groups present in diclofenac ethyl ester such as: C=O (1709.418), -CH2- (1451.252) , N-H (3291.477).

II.2.2. Synthesis of diclofenac phenylhydrazine

II.2.2.1. Solubility measure

- **Results**

The qualitative solubility results of P2 are shown in figure 32 and table XX:

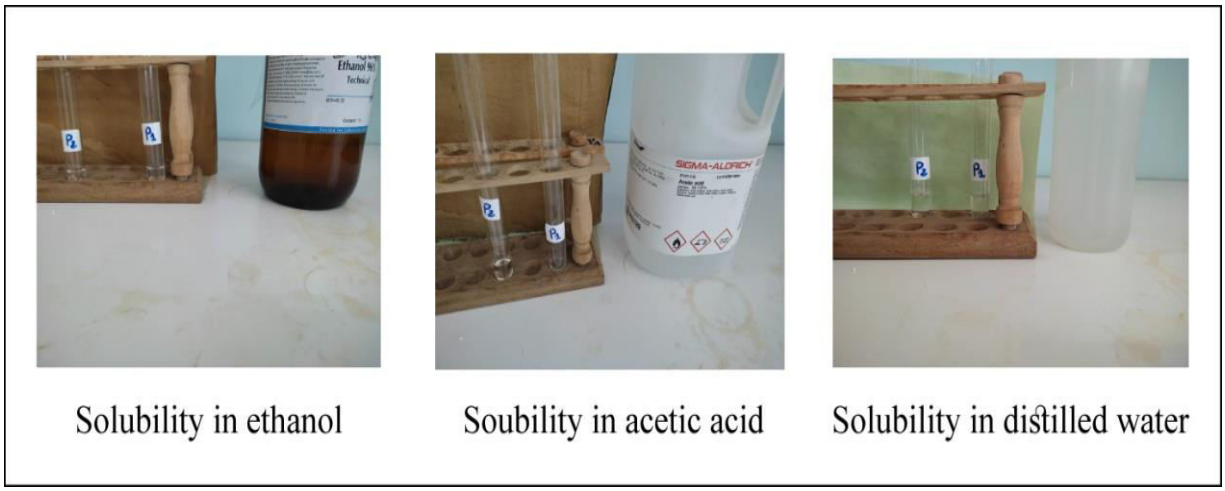


Figure 32: Solubility results of P2 in different solvents (left).

Table XX: Qualitative solubility results of P2 in different solvents.

Solvent	Distilled water	Ethanol	Acetic acid
Solubility	+/-	+	+

(-): insoluble (+): soluble (+/-): more or less soluble

- **Discussion**

P2 is more or less soluble in distilled water; it is an organic impolar product.

P2 is soluble in ethanol because it's an organic impolar product.

It is soluble in acid solvents such acetic acid, then it is a basic product.

II.2.2.2. Melting point measure

- **Results**

The measured melting point of pure P2 ester after drying is 67° C.

The melting point of the starting reagent diclofenac ethyl ester P1 was 63°C.

- **Discussion**

The melting point of the starting reagent diclofenac ethyl ester was 63°C.

There was a transition of only four degrees in the melting point, which gets us to suspect that the reaction is incomplete. The reagent P1 is probably still present.

II.2.2.3 Thin Layer Chromatography identification

- **Results**

TLC was carried out using different eluents A, B, C, D and E and observed under UV and on naked eye. The figure 33 illustrates the observed results in eluent C at T0 that were used to calculate the Rf (Cf. table XXIII).

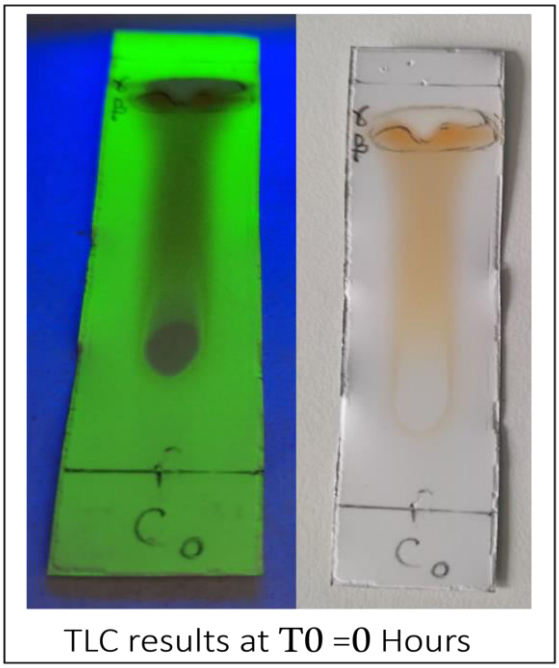


Figure 33: The results of the TLC monitoring for P2 diclofenac phenyl hydrazine synthesis, observed on naked eye then under UV using the eluent C at T0.

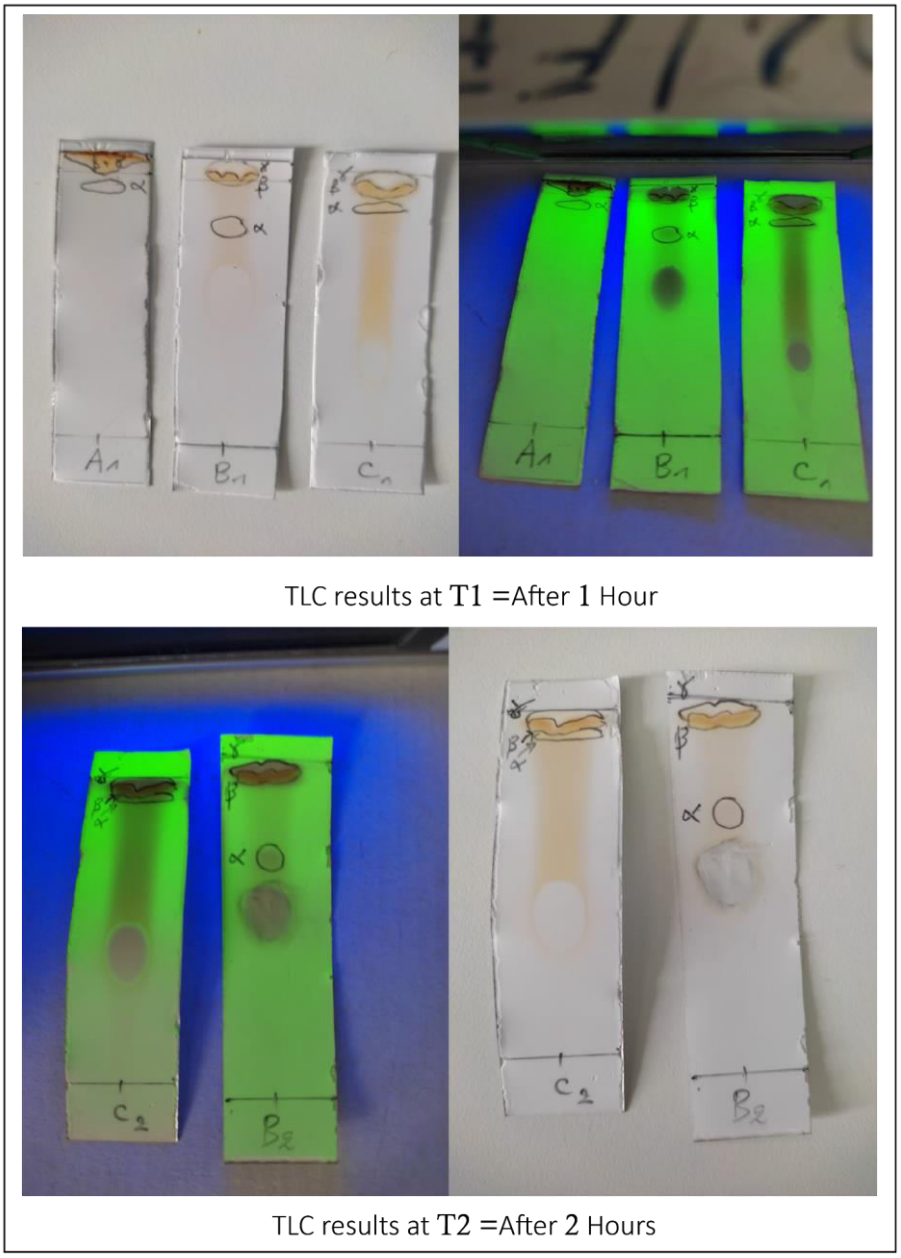


Figure 34: The results of the TLC monitoring for P2 diclofenac phenylhydrazine synthesis, observed on naked eye then under UV using the eluent A, B and C at T1 and T2.

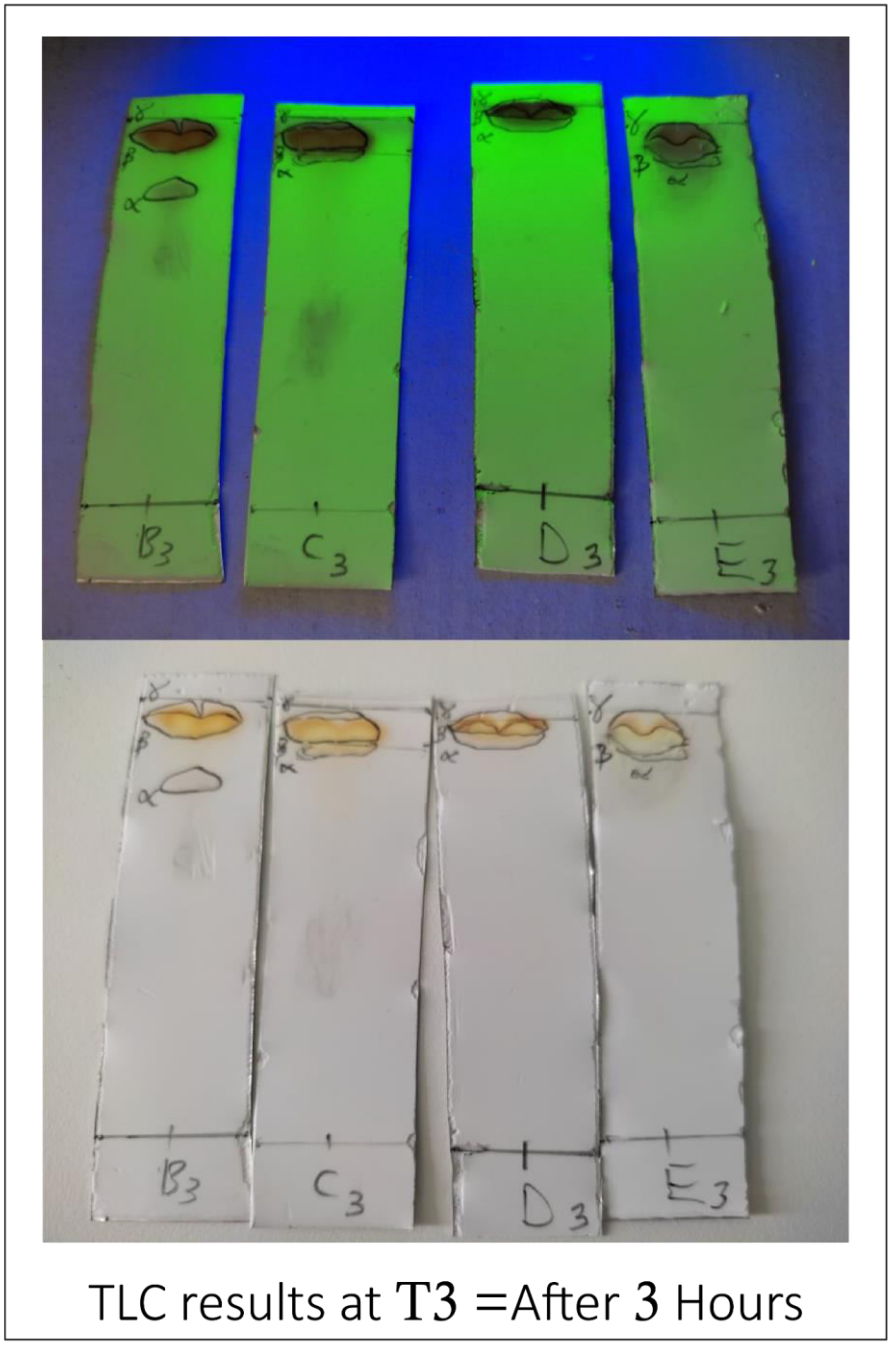


Figure 35: The results of the TLC monitoring for P2 diclofenac phenylhydrazine synthesis, observed on naked eye and under UV light using the eluent B,C,D and E at T3.

The height of the solvent front for the different eluents is equal to 5.9 centimeters. The result of Rfs of TLC is shown in the following table:

The height of the solvent front for the different eluents is equal to 5.9 centimeters. The result of TLC's Rfs is shown in the following table:

Table XXI: Rf results from TLC monitoring in the eluent A,B,C,D and E during multiple phases.

TLC Results	Product	Eluent				
		A	B	C	D	E
T0	P2	-	-	0	-	
	Phenyl hydrazine			0.87		
	P1			0.92		
T1 (After 1 hour)	P2	0.88	0.78	0.87	-	
	Phenyl hydrazine	Bad separation	0.96	0.88		
	P1		0.97	0.92		
T2 (After 2 hours)	P2	-	0.71	0.83	-	
	Phenyl hydrazine		0.94	0.86		
	P1		0.98	0.89		
T3 (After 3 hours)	P2	-	0.78	0.86	0.94	0.86
	Phenyl hydrazine		0.91	0.91	0.98	0.93
	P1		0.96	0.94	1	0.98

(-): non-realised TLC

- **Discussion**

At T0

We observe the presence of only two spots, one visible on naked eye (β), which corresponds to phenylhydrazine, above it, the other spot γ which corresponds to P1. The Rf results show two different values, each corresponding to a separate reagent.

At T1

We observe the appearance of a third spot (α) in eluents A, B and C that was unexisting in T0. The Rf results showed a value for α spot, while, showed a bad separation for the other two spots. Thus, eluent A was not used furthermore to monitor the progress of the substitution.

At T2

We always observe three spots α, β and γ. The intensity of α spot was increases, while that of β and γ was decreases. We conclude that a new product is being generated and the starting reagents are being consumed.

At T3

We observe the intensification of α spot and the weakening of the two other spots. Eluents D and E were used at this period to refine the results by adding acetic acid, the results were not satisfying.

The Rf results of α spot at T1, T2 and T3 are approximately the same, they are different from β and γ Rf results and correspond to the new generated product that was missing in T0.

II.2.2.4. UV characterisation

- **Results**

The spectrum below (Cf. figure 37) illustrates the UV absorption of P2 in ethanol. E5's maximal absorption was reached at 248 nm wavelength.

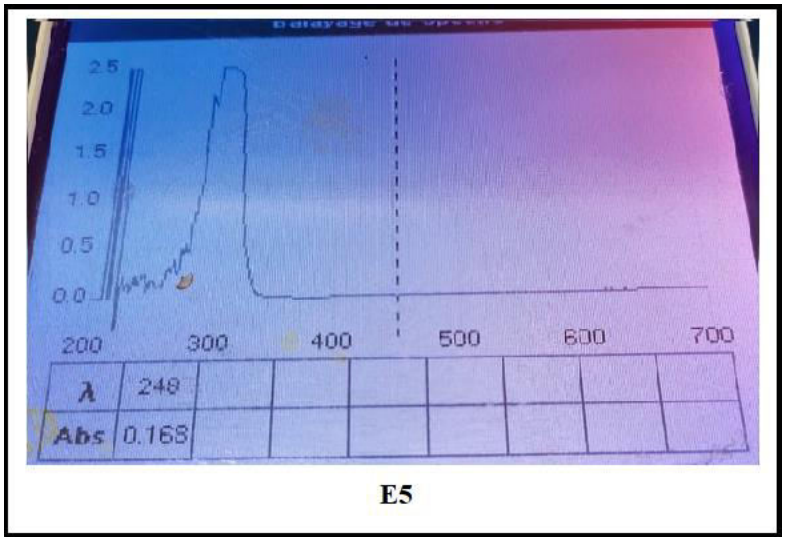


Figure 36: UV experimental spectrum of the fifth essay (E5).

- **Discussion**

Since all maximum absorption correspondant wavelengths values were between 200 and 300 nm, we are pretty sure there is a conjugated system in our product (phenyl ring).

E5 can be compared best with E1 (same solvent: ethanol, same volume: 3ml and comparable amounts of products used: 65 mg of P1, 50 mg of P2).
E1's maximal absorption was reached at 222 nm wavelength, whereas E5's was reached at 248 nm, these values are close which can lead us to suspect that these two products are the same (the reaction of phenylhydrazine with diclofenac ethyl ester did not occur), this hypothesis is to be confirmed with Infrared spectroscopy analysis.

II.2.2.5. Infrared spectroscopy

- **Results**

Figure 37 represents the practical IR spectrum of purified P2 (diclofenac phenylhydrazine).

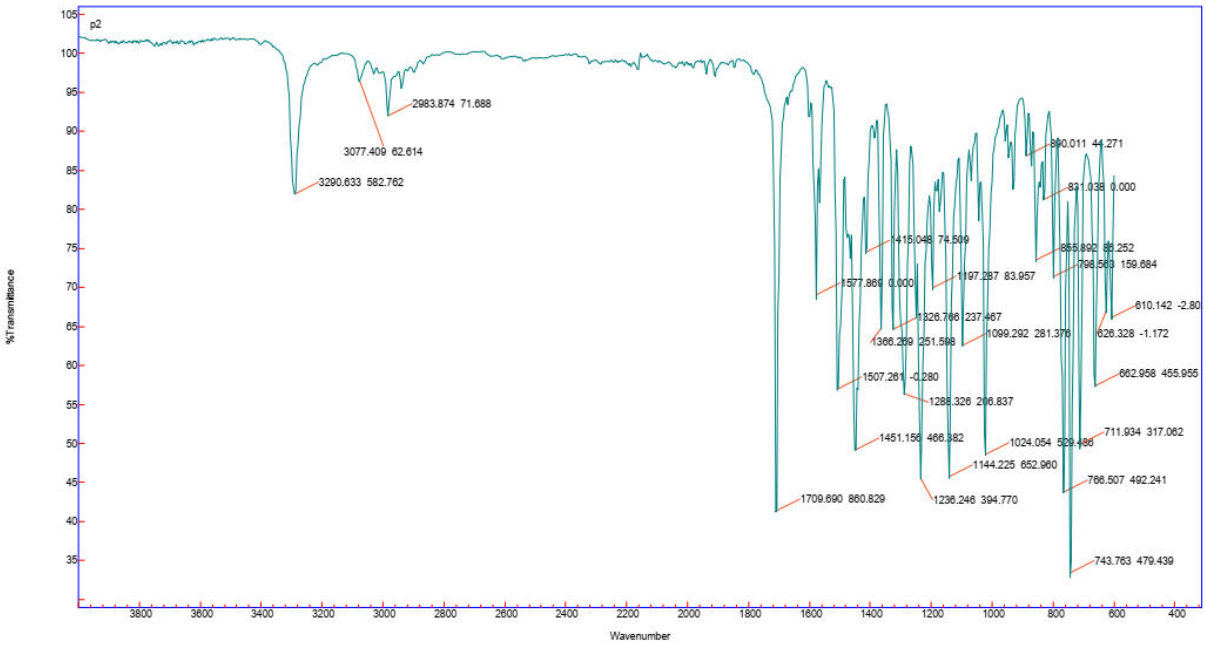


Figure 37: IR experimental spectrum of diclofenac phenylhydrazine (P2).

➢ **Discussion**

The allocation of the peaks of the IR spectra was made for the P1 based on infrared tables taken from the literature. The use of this latter and our practical spectrum made it possible to establish the next table:

Table XXII: Comparative analysis of the IR experimental spectrum of P2 with the reference spectrum.

	Reference spectrum	Practical frequency	Vibration mode
Functional Groups	3270	3290.633	N-H stretching
	2981	2983.874	-CH2-
	1720	1709.690	C=O
	1577	1577.869	C=C stretching
Digital Prints	1326	1326.766	C-N
	1238	1236.246	H-C-O
	1458	1451.156	-CH2-

	600-800	626.328	C-Cl
	600-800	610.142	C-Cl

The presence of certain characteristic bands on the practical spectrum confirms the existence functional groups present in diclofenac ethyl ester such as: N-H (3290.633), C-N (1326.766), C=C aromatic (1577.869).

For the two practical superpositioned spectrums P1 and P2 (Cf. figure 38), the comparative study of the position, the shape and the intensity of the bands indicates a superposition of these two spectrums. Analysis of the two IR spectra led us to conclude that P1 and P2 are the same product.

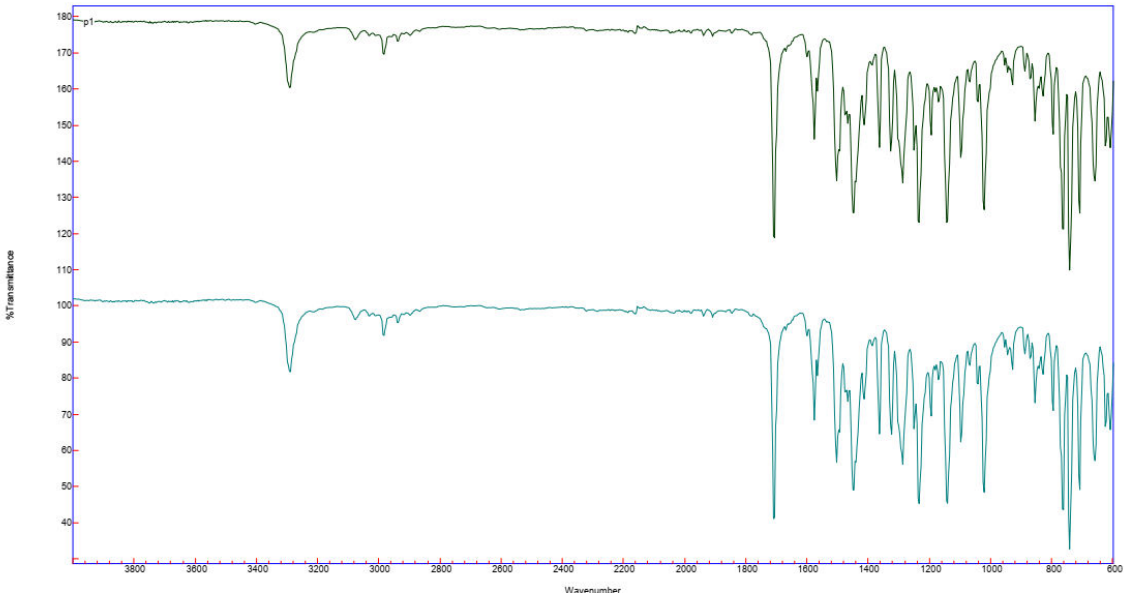


Figure 38: Th experimental superpositioned IR spectrums of P1 and P2.

CONCLUSION AND PERSPECTIVES

CONCLUSION AND PERSPECTIVES

Since Diclofenac was patented in 1965 by Ciba-Geigy, it was commonly used as an NSAID; its wide efficiency has then oriented research towards the discovery of new flexible and modular synthesis methodologies of biologically active derivative molecules of diclofenac. There are so many synthetic methods and techniques that the question is no longer whether the preparation of a molecular target is achievable, but rather with what perfection will it be.

The aim of this work is to synthesize biologically active compounds of diclofenac sodium. Given the importance of phenyl hydrazine substituents in the treatment of tuberculosis, Parkinson and cancer diseases, we were interested in synthesizing the compound: Diclofenac phenyl hydrazine.

First, we synthesized diclofenac acid. The use of concentrated sulfuric acid was unachievable because of the required volume, we then used 1/10 sulfuric acid to proceed the acidification reaction. The Ph bend confirmed the progress towards an acidic solution.

Next, diclofenac ethyl ester was synthesized with good efficiency (yield 74%). Modulating the duration or temperature of the heating or the use of another recrystallization solvent could have improved it. Analytical control of the product issue confirmed the structure of the compound in question.

Then, we wanted to synthesize diclofenac phenyl hydrazine, but this could not be achieved. The analytical methods did not identify the structure of this compound. We obtained an intermediary with different structural characteristics than what diclofenac phenyl hydrazine should be. The use of a catalyst or the modulation of the duration and the temperature could make the reaction complete.

The identification was carried out by measuring the melting point and TLC Rfs, the results of which the synthesis of P1 was satisfactory. On the other hand, that of synthesis of P2 was not.

The identification was also carried out by a spectral study: infrared analysis. This analysis has proven its importance in identifying functional groupings of the structure of the different synthesized products, the results showed no difference between P1 and P2.

HPLC analysis of the synthesized products could improve the results of their purity by identifying the presence of specific impurities and determining their content. It also allows them to be dosed with active substances. Only it requires availability standards.

Due to the pandemic situation we could not synthesize other diclofenac derivatives. Testing the biological activity on *M.tuberculosis* strains was not possible due to the complexity and the danger that can cause such operation.

The lack of cell banks in Algeria resulted in unachieving the testing the biological activity on Parkinson targeted ligand and cancer cells.

SOURCES

1. World Health Organization. Global tuberculosis report 2020 [Internet]. Geneva: World Health Organization; 2020 [cité 21 sept 2021]. Disponible sur: https://apps.who.int/iris/handle/10665/336069
2. Duarte R, Aguiar A, Pinto M, Furtado I, Tiberi S, Lönnroth K, et al. Different disease, same challenges: Social determinants of tuberculosis and COVID-19. Pulmonology. juill 2021;27(4):338-44.
3. Churchyard G, Kim P, Shah NS, Rustomjee R, Gandhi N, Mathema B, et al. What We Know About Tuberculosis Transmission: An Overview. The Journal of Infectious Diseases. 3 nov 2017;216(suppl_6):S629-35.
4. Griffith DE, Kerr CM. Tuberculosis: Disease of the past, disease of the present. Journal of PeriAnesthesia Nursing. août 1996;11(4):240-5.
5. Selmane S, L'hadj M. Spatiotemporal Analysis and Seasonality of Tuberculosis in Algeria. International Journal of Mycobacteriology. 2021; 10(3):9.
6. Selmane S, L'Hadj M. Epidemiology and clinical characteristics of tuberculosis in leon bernard tuberculosis unit in algeria. Int J Mycobacteriol. 2020;9(3):254.
7. Fukunaga R, Glaziou P, Harris JB, Date A, Floyd K, Kasaeva T. Epidemiology of Tuberculosis and Progress Toward Meeting Global Targets — Worldwide, 2019. MMWR Morb Mortal Wkly Rep. 26 mars 2021;70(12):427-30.
8. Yates TA, Khan PY, Knight GM, Taylor JG, McHugh TD, Lipman M, et al. The transmission of Mycobacterium tuberculosis in high burden settings. The Lancet Infectious Diseases. févr 2016;16(2):227-38.
9. Cadena AM, Hopkins FF, Maiello P, Carey AF, Wong EA, Martin CJ, et al. Concurrent infection with Mycobacterium tuberculosis confers robust protection against secondary infection in macaques. Salgame P, éditeur. PLoS Pathog. 12 oct 2018;14(10):e1007305.
10. Palomino J, Martin A. Drug Resistance Mechanisms in Mycobacterium tuberculosis. Antibiotics. 2 juill 2014;3(3):317-40.
11. Ferraris D, Miggiano R, Rossi F, Rizzi M. Mycobacterium tuberculosis Molecular Determinants of Infection, Survival Strategies, and Vulnerable Targets. Pathogens. 1 févr 2018;7(1):17.
12. Agadir F, Halassa SA, Ali Pacha S, Baough L. Manuel de la lutte Antituberculeuse: a l'usage des personnels medicaux. 2011.
13. Lancini G, Parenti F, Gallo GG. The Antibiotics: An Overview. In: Antibiotics [Internet].

Boston, MA: Springer US; 1995 [cité 23 sept 2021]. p. 1-14. Disponible sur: http://link.springer.com/10.1007/978-1-4757-9200-3_1

14. Shi R, Itagaki N, Sugawara I. Overview of Anti-Tuberculosis (TB) Drugs and Their Resistance Mechanisms. MRMC. 1 nov 2007;7(11):1177-85.
15. Addington WW. The Side Effects and Interactions of Antituberculosis Drugs. Chest. déc 1979;76(6):782-4.
16. Weyer K, Falzon D, Jaramillo E. Towards all-oral and shorter treatment regimens for drug-resistant tuberculosis. Bulletin of the World Health Organization. 1 oct 2018;96(10):667-667A.
17. Sriram D, Yogeeswari P, Devakaram RV. Synthesis, in vitro and in vivo antimycobacterial activities of diclofenac acid hydrazones and amides. Bioorganic & Medicinal Chemistry. mai 2006;14(9):3113-8.
18. Ovallath S, Deepa P. The history of parkinsonism: Descriptions in ancient Indian medical literature: History of Parkinsonism. Mov Disord. mai 2013;28(5):566-8.
19. Parent A. Franciscus Sylvius on Clinical Teaching, Iatrochemistry and Brain Anatomy. Can J Neurol Sci. juill 2016;43(4):596-603.
20. Goetz CG. The History of Parkinson's Disease: Early Clinical Descriptions and Neurological Therapies. Cold Spring Harbor Perspectives in Medicine. 1 sept 2011;1(1):a008862-a008862.
21. Palacios-Sánchez L, Torres Nupan M, Botero-Meneses JS. James Parkinson and his essay on "shaking palsy", two hundred years later. Arq Neuro-Psiquiatr. sept 2017;75(9):671-2.
22. Lewis PA, Plun-Favreau H, Rowley M, Spillane J. Pierre D. and the First Photographs of Parkinson's Disease. Mov Disord. mars 2020;35(3):389-91.
23. Sveinbjornsdottir S. The clinical symptoms of Parkinson's disease. J Neurochem. oct 2016;139:318-24.
24. Ball N, Teo W-P, Chandra S, Chapman J. Parkinson's Disease and the Environment. Front Neurol. 19 mars 2019;10:218.
25. Kalia LV, Lang AE. Parkinson's disease. The Lancet. août 2015;386(9996):896-912.
26. Wooten GF. Are men at greater risk for Parkinson's disease than women? Journal of Neurology, Neurosurgery & Psychiatry. 1 avr 2004;75(4):637-9.
27. Ferguson LW, Rajput AH, Rajput A. Early-onset vs. Late-onset Parkinson's disease: A Clinical-pathological Study. Can J Neurol Sci. janv 2016;43(1):113-9.
28. Vela L, Martínez Castrillo JC, García Ruiz P, Gasca-Salas C, Macías Macías Y, Pérez Fernández E, et al. The high prevalence of impulse control behaviors in patients with

early-onset Parkinson's disease: A cross-sectional multicenter study. Journal of the Neurological Sciences. sept 2016;368:150-4.

29. Abbas MM, Xu Z, Tan LCS. Epidemiology of Parkinson's Disease-East Versus West. Mov Disord Clin Pract. janv 2018;5(1):14-28.
30. Chaudhuri KR, National Parkinson Foundation Centre of Excellence, King's College Hospital, Denmark Hill, London, UK, King's College London, Department of Neurosciences, Institute of Psychiatry, Psychology & Neuroscience, London, UK, Titova N, Department of Neurology, Neurosurgery and Medical Genetics, Pirogov Russian National Research Medical University, Moscow, Russian Federation. Societal Burden and Persisting Unmet Needs of Parkinson's Disease. European Neurological Review. 2019;14(1):28.
31. Dorsey ER, Sherer T, Okun MS, Bloem BR. The Emerging Evidence of the Parkinson Pandemic. Brundin P, Langston JW, Bloem BR, éditeurs. JPD. 18 déc 2018;8(s1):S3-8.
32. Setianingsih DA, Abdullah S. Identification of risk factors for impulse-control disorder symptoms in patients with Parkinson's disease. J Phys: Conf Ser. janv 2021;1722:012099.
33. Khan MAB, Grivna M, Nauman J, Soteriades ES, Cevik AA, Hashim MJ, et al. Global Incidence and Mortality Patterns of Pedestrian Road Traffic Injuries by Sociodemographic Index, with Forecasting: Findings from the Global Burden of Diseases, Injuries, and Risk Factors 2017 Study. IJERPH. 23 mars 2020;17(6):2135.
34. Hirsch L, Jette N, Frolkis A, Steeves T, Pringsheim T. The Incidence of Parkinson's Disease: A Systematic Review and Meta-Analysis. Neuroepidemiology. 2016;46(4):292-300.
35. Alonso A, Rodriguez LAG, Logroscino G, Hernan MA. Use of antidepressants and the risk of Parkinson's disease: a prospective study. Journal of Neurology, Neurosurgery & Psychiatry. 1 juin 2009;80(6):671-4.
36. Weintraub D, Moberg PJ, Duda JE, Katz IR, Stern MB. Effect of Psychiatric and Other Nonmotor Symptoms on Disability in Parkinson's Disease: NONMOTOR CAUSES OF DISABILITY IN PD. Journal of the American Geriatrics Society. mai 2004;52(5):784-8.
37. Louis ED, Wise A, Alcalay RN, Rao AK, Factor-Litvak P. Essential tremor-Parkinson's disease: A double whammy. Journal of the Neurological Sciences. juill 2016;366:47-51.
38. Opara JA, Małecki A, Małecka E, Socha T. Motor assessment in Parkinson`s disease. Annals of Agricultural and Environmental Medicine. 2017;24(3):5.
39. Erro R, Stamelou M. The Motor Syndrome of Parkinson's Disease. In: International

Review of Neurobiology [Internet]. Elsevier; 2017 [cité 13 sept 2021]. p. 25-32. Disponible sur: https://linkinghub.elsevier.com/retrieve/pii/S0074774217300041

40. Rascol O, Payoux P, Ory F, Ferreira JJ, Brefel-Courbon C, Montastruc J-L. Limitations of current Parkinson's disease therapy. Ann Neurol. 2003;53(S3):S3-15.
41. Kranick SM, Duda JE. Olfactory Dysfunction in Parkinson's Disease. Neurosignals. 2008;16(1):35-40.
42. Winge K, Skau A-M, Stimpel H, Nielsen KK, Werdelin L. Prevalence of bladder dysfunction in Parkinsons disease. Neurourol Urodyn. mars 2006;25(2):116-22.
43. Landau S, Harris V, Burn DJ, Hindle JV, Hurt CS, Samuel M, et al. Anxiety and anxious-depression in Parkinson's disease over a 4-year period: a latent transition analysis. Psychol Med. févr 2016;46(3):657-67.
44. Balestrino R, Schapira AHV. Parkinson disease. Eur J Neurol. janv 2020;27(1):27-42.
45. Bonam SR, Muller S. Parkinson's disease is an autoimmune disease: A reappraisal. Autoimmunity Reviews. déc 2020;19(12):102684.
46. Di Maio R, Hoffman EK, Rocha EM, Keeney MT, Sanders LH, De Miranda BR, et al. LRRK2 activation in idiopathic Parkinson's disease. Sci Transl Med. 25 juill 2018;10(451):eaar5429.
47. Collins LM, Toulouse A, Connor TJ, Nolan YM. Contributions of central and systemic inflammation to the pathophysiology of Parkinson's disease. Neuropharmacology. juin 2012;62(7):2154-68.
48. Nguyen TN, Padman BS, Lazarou M. Deciphering the Molecular Signals of PINK1/Parkin Mitophagy. Trends in Cell Biology. oct 2016;26(10):733-44.
49. Plotkin JL, Goldberg JA. Thinking Outside the Box (and Arrow): Current Themes in Striatal Dysfunction in Movement Disorders. Neuroscientist. août 2019;25(4):359-79.
50. Galvan A, Wichmann T. Pathophysiology of Parkinsonism. Clinical Neurophysiology. juill 2008;119(7):1459-74.
51. Moldovan A-S, Groiss S, Elben S, Südmeyer M, Schnitzler A, Wojtecki L. The treatment of Parkinson′s disease with deep brain stimulation: current issues. Neural Regen Res. 2015;10(7):1018.
52. Cools R. The costs and benefits of brain dopamine for cognitive control: The costs and benefits of brain dopamine for cognitive control. WIREs Cogn Sci. sept 2016;7(5):317-29.
53. Kujawa K, Leurgans S, Raman R, Blasucci L, Goetz CG. Acute Orthostatic Hypotension When Starting Dopamine Agonists in Parkinson's Disease. Arch Neurol [Internet]. 1 oct

2000 [cité 13 sept 2021];57(10). Disponible sur: http://archneur.jamanetwork.com/article.aspx?doi=10.1001/archneur.57.10.1461

54. Javaraju S, Ahmed H, Alsinai A, Soner N. Domination topological properties of carbidopa-levodopa used for treatment Parkinson's disease by using φp-polynomial. Eurasian Chem Commun [Internet]. sept 2021 [cité 13 sept 2021];3(9). Disponible sur: https://doi.org/10.22034/ecc.2021.295039.1203
55. Olanow CW, Stern MB, Sethi K. The scientific and clinical basis for the treatment of Parkinson disease (2009). Neurology. 26 mai 2009;72(Issue 21, Supplement 4):S1-136.
56. Boyle A, Ondo W. Role of Apomorphine in the Treatment of Parkinson's Disease. CNS Drugs. févr 2015;29(2):83-9.
57. Bearn J, Evans A, Kelleher M, Turner K, Lees A. Recognition of a dopamine replacement therapy dependence syndrome in Parkinson's disease: a pilot study☆☆Supplementary. Drug and Alcohol Dependence. 7 déc 2004;76(3):305-10.
58. Brennan K, Genever R. Managing Parkinson's disease during surgery. BMJ. 1 nov 2010;341(nov01 2):c5718-c5718.
59. Subramony JA. Apomorphine in Dopaminergic Therapy. Mol Pharmaceutics. 1 août 2006;3(4):380-5.
60. Zeuner KE, Schäffer E, Hopfner F, Brüggemann N, Berg D. Progress of Pharmacological Approaches in Parkinson's Disease. Clin Pharmacol Ther. mai 2019;105(5):1106-20.
61. Di Giovanni S, Eleuteri S, Paleologou KE, Yin G, Zweckstetter M, Carrupt P-A, et al. Entacapone and Tolcapone, Two Catechol O-Methyltransferase Inhibitors, Block Fibril Formation of α-Synuclein and β-Amyloid and Protect against Amyloid-induced Toxicity. Journal of Biological Chemistry. mai 2010;285(20):14941-54.
62. Sudhakar P, Pushkalai SP, Sabarinath C, Priyadharshini S, Haripriya S. Molecular docking and synthesis of 1, 2, 4 -triazin analogue of diclofenac as potential ligand for parkinson's. Rese Jour Pharmacol and Pharmacod. 2018;10(1):8.
63. Arneth B. Tumor Microenvironment. Medicina. 30 déc 2019;56(1):15.
64. Fadaka A, Ajiboye B, Ojo O, Adewale O, Olayide I, Emuowhochere R. Biology of glucose metabolization in cancer cells. Journal of Oncological Sciences. juill 2017;3(2):45-51.
65. Chaffer CL, Weinberg RA. A Perspective on Cancer Cell Metastasis. Science. 25 mars 2011;331(6024):1559-64.
66. Bacac M, Stamenkovic I. Metastatic Cancer Cell. Annu Rev Pathol Mech Dis. févr 2008;3(1):221-47.

67. Masood I, Kiani MH, Ahmad M, Masood MI, Sadaquat H. Major Contributions towards Finding a Cure for Cancer through Chemotherapy: A Historical Review. Tumori. janv 2016;102(1):6-17.
68. Ralhan R, Kaur J. Alkylating agents and cancer therapy. Expert Opinion on Therapeutic Patents. sept 2007;17(9):1061-75.
69. Soriano CM. Platinum. IEEE Potentials. 1998;16(5):29-32.
70. Chen X, Wu Y, Dong H, Zhang C, Zhang Y. Platinum-Based Agents for Individualized Cancer Treatment. CMM. 31 nov 2013;13(10):1603-12.
71. Ali I, A. Wani W, Saleem K, Haque A. Platinum Compounds: A Hope for Future Cancer Chemotherapy. ACAMC. 1 janv 2013;13(2):296-306.
72. Jamieson ER, Lippard SJ. Structure, Recognition, and Processing of Cisplatin−DNA Adducts. Chem Rev. 8 sept 1999;99(9):2467-98.
73. Trimmer EE, Essigmann JM. Cisplatin. Essays Biochem. 1999;34:191-211.
74. Pujade-Lauraine E, Wagner U, Aavall-Lundqvist E, Gebski V, Heywood M, Vasey PA, et al. Pegylated Liposomal Doxorubicin and Carboplatin Compared With Paclitaxel and Carboplatin for Patients With Platinum-Sensitive Ovarian Cancer in Late Relapse. JCO. 10 juill 2010;28(20):3323-9.
75. Intini FP, Zajac J, Novohradsky V, Saltarella T, Pacifico C, Brabec V, et al. Novel Antitumor Platinum(II) Conjugates Containing the Nonsteroidal Anti-inflammatory Agent Diclofenac: Synthesis and Dual Mechanisms of Antiproliferative Effects. Inorg Chem. 6 févr 2017;56(3):1483-97.
76. Ramachandran E, Ramukutty S. Growth, morphology, spectral and thermal studies of gel grown diclofenac acid crystals. Journal of Crystal Growth. mars 2014;389:78-82.
77. Cary R, Dobson S, Brooke I. Phenylhydrazine. World Health Organization. 2000;

Abstract

The carboxylic acid function of Diclofenac is essential for anti-inflammatory activity, its esterification leads to prodrugs. On the other hand, its transformation into hydrazine leads to derivatives endowed with anti-tuberculosis activity.

This end-of-study project is based on one of the methods of synthesizing hydrazine-functional diclofenac from diclofenac sodium. These compounds will subsequently be identified by the different methods of characterization and analysis such as solubility measurement, melting point measurement, UV spectroscopy and IR spectroscopy. This reaction was monitored during multiple phases using TLC.

The monitoring showed the appearance of a new resulting product while that of IR expressed approximatively the same results between the two synthesised products. This might be due to the use of phenylhydrazine instead of hydrazine hydrate which have less nucleophilic properties due to the presence of the phenyl function.
Keywords: Diclofenac,Phenylhydrazine-Tuberculosis-Cancer-Parkinson

Résumé

La fonction acide carboxylique du Diclofénac est essentielle pour l'activité anti-inflammatoire, son estérification conduit à des prodrogues. En revanche, sa transformation en hydrazine conduit à des dérivés doués d'activité antituberculeuse.

Ce projet de fin d'étude est basé sur l'une des méthodes de synthèse du diclofénac à fonction hydrazine à partir du diclofénac sodique. La mesure de ces composés sera notamment identifiée par les différentes méthodes de caractérisation et d'analyse, mesure du point de fusion, spectroscopie UV et spectroscopie IR. Cette réaction a été surveillée pendant plusieurs phases en utilisant la CCM.

Le suivi a montré l'apparition d'un nouveau produit résultant alors que celui de l'IR exprimait approximativement les mêmes résultats entre les deux produits synthétisés. Cela pourrait être dû à l'utilisation de phénylhydrazine à la place de l'hydrate d'hydrazine qui a moins de propriétés nucléophiles en raison de la présence de la fonction phényle.
Mots clés: Diclofénac-Phénylhydrazine-Tuberculose-Cancer-Parkinson

ملخص

تعتبر وظيفة حمض الكربوكسيل للديكلوفيناك ضرورية للنشاط المضاد للالتهابات ، وتؤدي الأسترة إلى عقاقير أولية. من ناحية أخرى ، يؤدي تحوله إلى الهيدرازين إلى مشتقات تتمتع بنشاط مضاد لمرض السل.

يعتمد مشروع نهاية الدراسة هذا على إحدى طرق تصنيع ديكلوفيناك الهيدرازين الوظيفي من ديكلوفيناك الصوديوم. سيتم تحديد هذه المركبات لاحقًا من خلال الطرق المختلفة للتوصيف والتحليل مثل قياس القابلية للذوبان وقياس نقطة الانصهار والتحليل الطيفي للأشعة فوق البنفسجية والتحليل الطيفي للأشعة تحت الحمراء. تمت مراقبة هذا التفاعل خلال مراحل متعددة باستخدام TLC.

أظهر الرصد ظهور منتج جديد ناتج بينما عبر IR عن نفس النتائج تقريبًا بين المنتجين المركبين. قد يكون هذا بسبب استخدام فينيل هيدرازين بدلاً من هيدرات الهيدرازين التي لها خصائص أقل للنواة بسبب وجود وظيفة فينيل.

الكلمات المفتاحية: ديكلوفيناك - فينيل هيدرازين - السل - السرطان - باركنسون