The importance of Ethnobotany and Herbarium studies as the first step in Phytochemistry and Biological activity research

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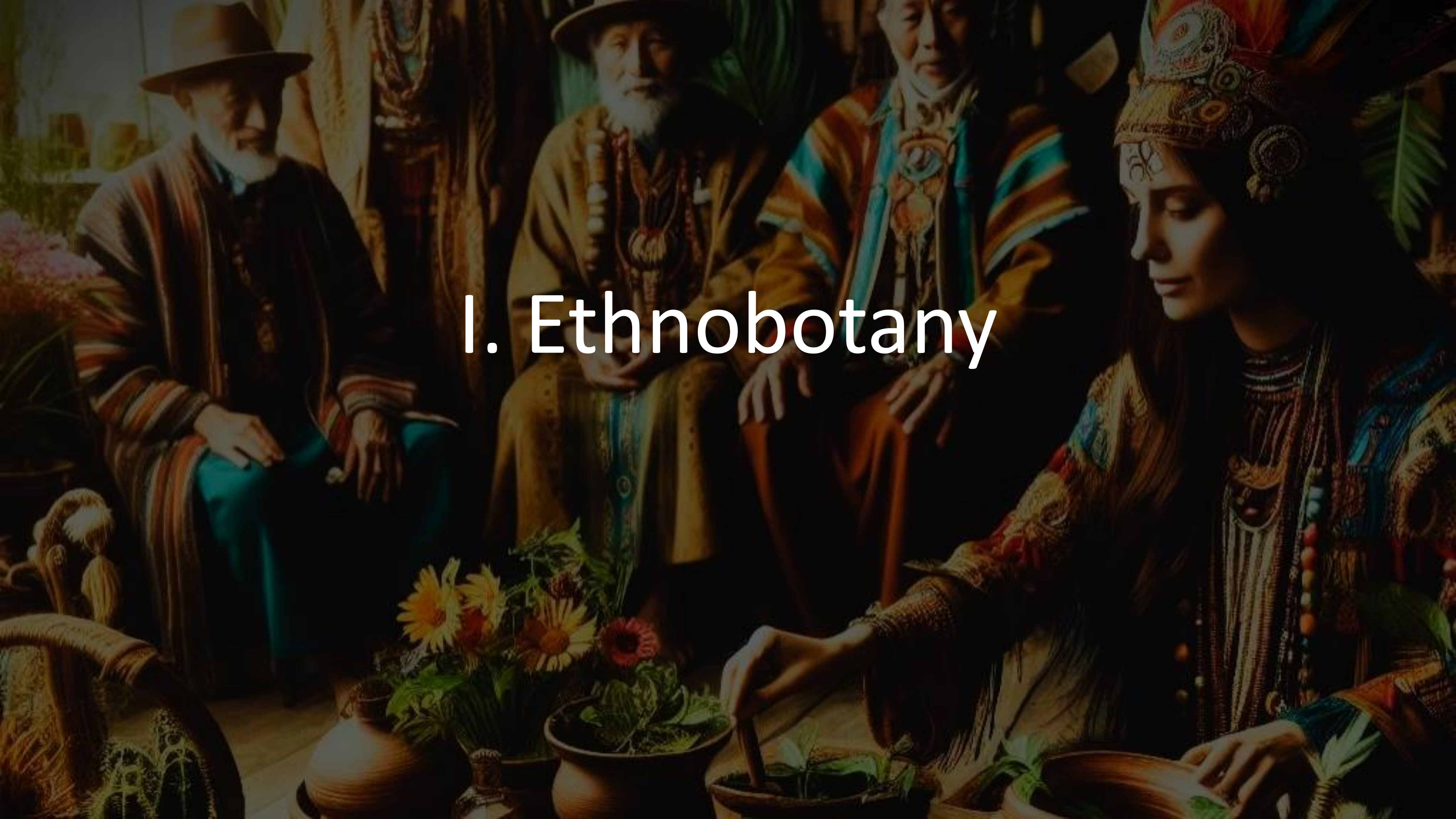
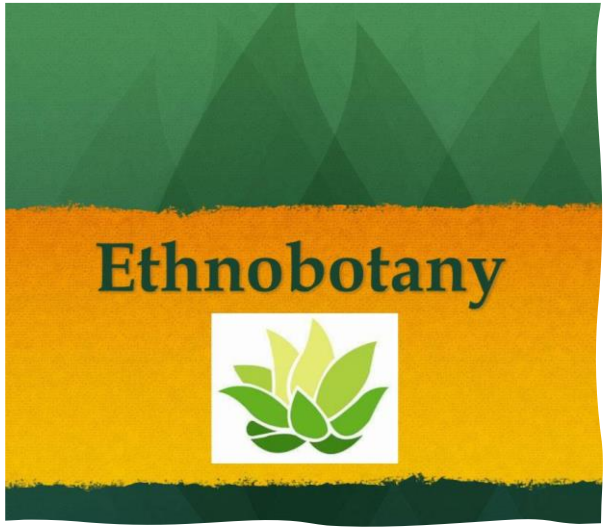
I. **Ethnobotany and Herbarium**
II. **Phytochemistry**
III. **Biological activity studies**

I. Ethnobotany






PLANT-HUMAN RELATIONSHIPS

RELATIONS PLANTES-HUMAINS

- The term Ethnobotany, which is formed by combining the Greek words ethnos (people) and botanae (plant). It is the branch of science that studies the plant-human relationship from the past to the present.
- L'ethnobotanique, terme formé à partir des mots grecs ethnos (peuple) et botanae (plante), est la branche de la science qui étudie les relations entre les plantes et l'homme, du passé à nos jours.
- It was first used by American botanist J.W. Harshberger in 1895. He wrote the first book about ethnobotany which is called “The Purposes of Ethnobotany” published in 1896.
- Elle a été utilisée pour la première fois par le botaniste américain J.W. Harshberger en 1895. Il a écrit le premier livre sur l'ethnobotanique intitulé « The Purposes of Ethnobotany », publié en 1896.

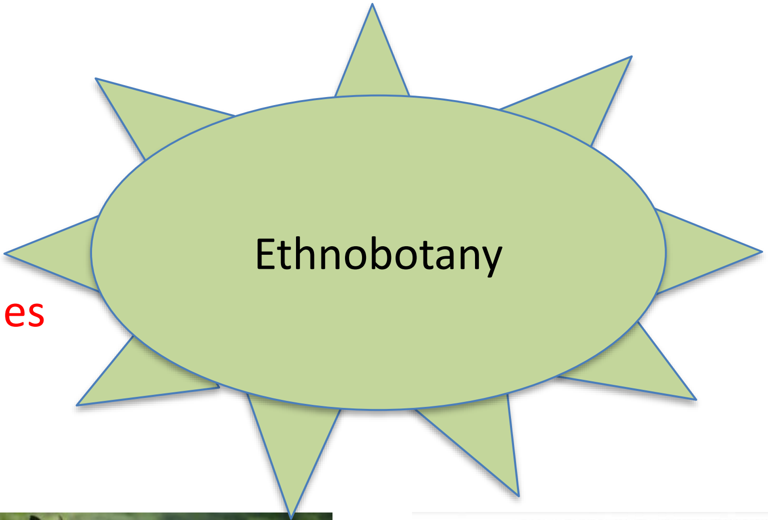
Ethnobotanical research areas - Domaines de recherche ethnobotanique

Veterinary medicinal plants
Plantes médicinales vétérinaires

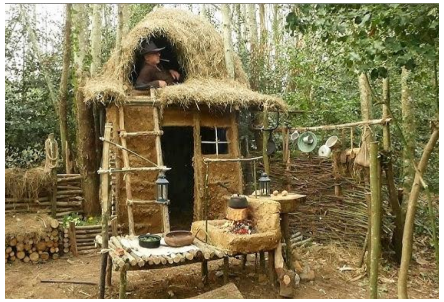
Medicinal plants-Plantes médicinales

Edible wild plants
Plantes sauvages comestibles

Ornamental plants
Plantes ornementales

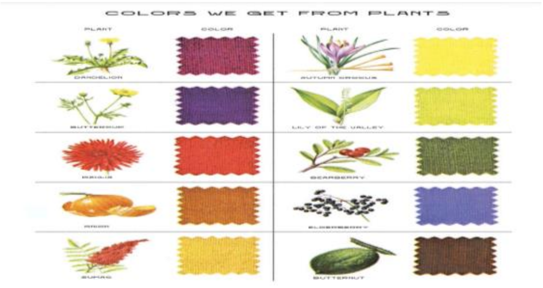


Art plants
Plantes artistiques



Plants to make tools, shelter
Plantes pour fabriquer des outils, des abris

Ceremony plants
Plantes de cérémonie

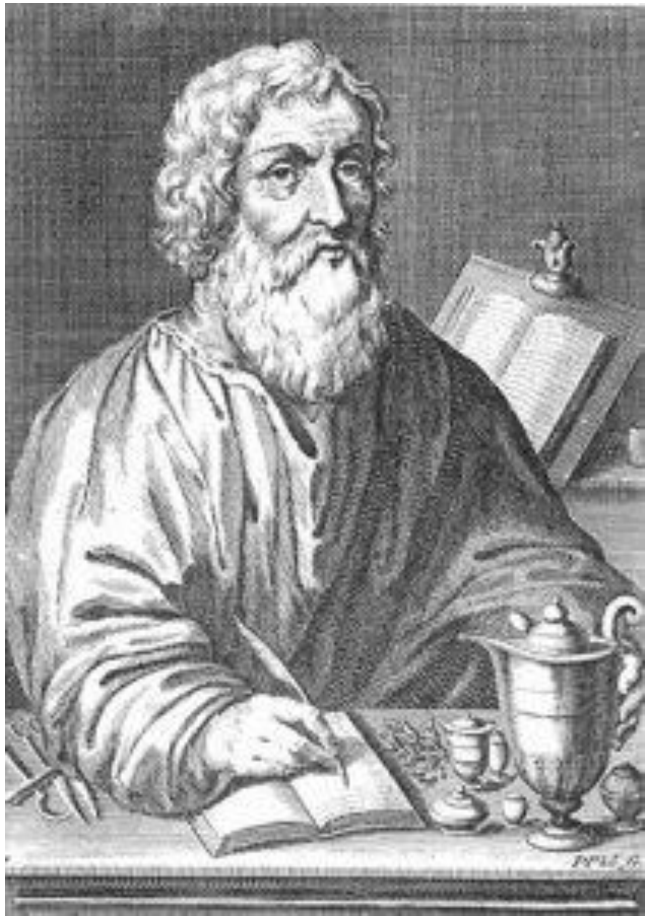


Dye Yielding Plants
Plantes tinctoriales

Cosmetic plants
Plantes cosmétiques

The transformation of ethnomedical knowledge into scientific medicine began with Hippocrates

La transformation des connaissances ethnomédicales en médecine scientifique a commencé avec Hippocrate



- Hippocrates (460-370 BC) is the father of medicine and he played an important role as an herbalist.

Hippocrate (460-370 av. J.-C.) est le père de la médecine et il a joué un rôle important en tant qu'herboriste.

- Hippocrates traveled for years throughout Greece to collect a wealth of information about medicinal plants for various diseases suffered by people.

Hippocrate a parcouru la Grèce pendant des années pour recueillir une mine d'informations sur les plantes médicinales utilisées pour soigner diverses maladies dont souffraient les gens.

Galen (around 129–216 CE) was one of the most famous physicians of Ancient Rome. Born in the ancient city of Pergamon (present-day Bergama, Turkiye).

Galien (vers 129-216 ap. J.-C.) fut l'un des médecins les plus célèbres de la Rome antique. Il naquit dans l'ancienne ville de Pergame (aujourd'hui Bergama, en Turkiye).

He organized and expanded the medical plants knowledge that already existed from Hippocrates and earlier Greek, Egyptian, and Mesopotamian traditions.

Il a organisé et développé les connaissances sur les plantes médicinales qui existaient déjà grâce à Hippocrate et aux traditions grecques, égyptiennes et mésopotamiennes antérieures.

Herbal mixtures (Galenic medicines): Instead of using a single plant, Galen combined several herbs to create complex mixtures. That’s why even today, the term *“galenic preparation”* is used to describe medicines made from natural substances.

Préparations galéniques: au lieu d’utiliser une seule plante, Galien combinait plusieurs herbes pour créer des mélanges complexes. C’est pourquoi, encore aujourd’hui, le terme « préparation galénique » est employé pour décrire les médicaments à base de substances naturelles.

Importance of Ethnobotany

Importance de l'ethnobotanique

1. Preserving knowledge about the traditional local uses of plants which is going to be lost due to migration from the village to the city

Préserver les connaissances relatives aux usages traditionnels des plantes au niveau local, qui risquent de disparaître en raison de l'exode rural.

2. Provides preliminary information for phytochemistry and bioactivity studies

Fournit des informations préliminaires pour les études de phytochimie et de bioactivité

3. Recording the traditional local names that could be forgotten over time

Consigner les noms locaux traditionnels qui pourraient tomber dans l'oubli avec le temps

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Medicinal plants are traditionaly has been used in Anatolia for centuries. There is a rich ethnobotanical research collection in Türkiye.

Les plantes médicinales sont utilisées traditionnellement en Anatolie depuis des siècles. La Turquie possède une riche collection de données ethnobotaniques.


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ELSEVIER

An ethnobotanical survey of medicinal plants in western part of central Taurus Mountains: Aladaglar (Nigde – Turkey)

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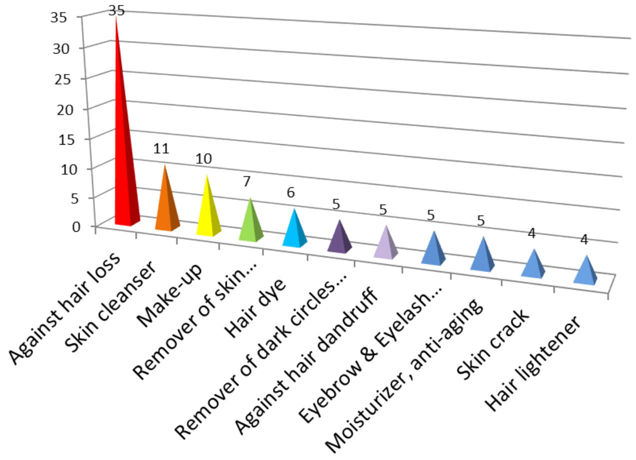
Methods

Méthodes



Ethnobotanical research has three main stages

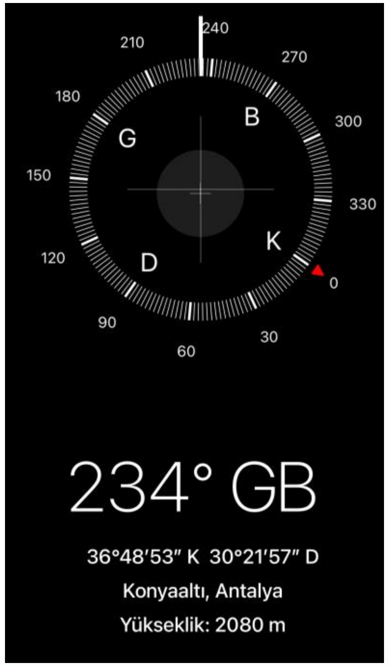
La recherche ethnobotanique comporte trois étapes principales

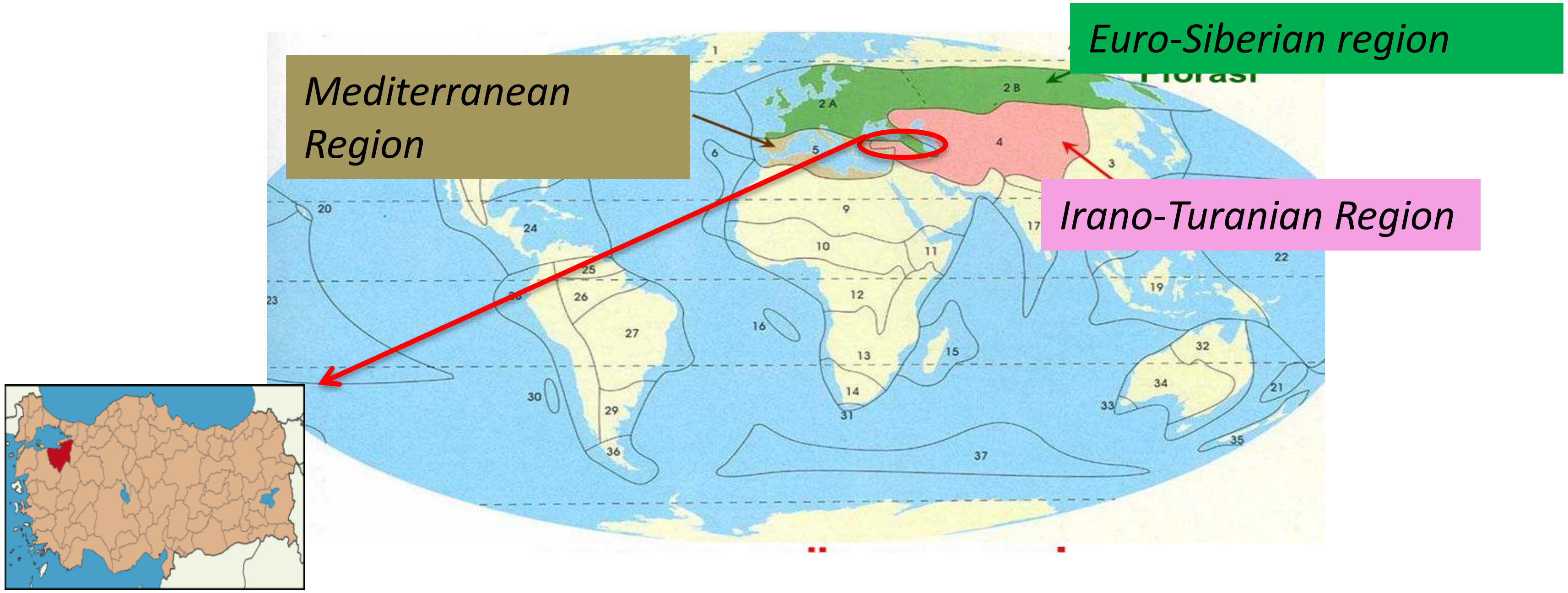
1. Field studies in the research area (Études de terrain dans la zone de recherche)
2. Herbarium studies (Études d'herbier)
3. Evaluation of findings, statistics (Évaluation des résultats, statistiques)

1. Field studies (études de terrain)

- Choose good representatives of the plant species (Choisissez de bons représentants de l'espèce végétale).
- Be careful that these plants must include root, stem, flower and fruit (Veillez à ce que ces plantes comprennent racine, tige, fleur et fruit).
- Take notes and record by taking photos in the field at the time of collection (Prenez des notes et documentez les données en prenant des photos sur le terrain au moment de la collecte).

Date, Collection Number, Location (GPS), Habitat, Special characteristics (Date, numéro de collecte, localisation (GPS), habitat, caractéristiques particulières)





Türkiye is one of the richest countries in the world in terms of plant diversity. To date, approximately **<u>12000 plant species</u>** have been identified and the endemism rate is **34 %.**

La Turquie est l'un des pays les plus riches au monde en termes de diversité végétale. À ce jour, environ 12 000 espèces végétales ont été recensées et le taux d'endémisme est de 34 %.

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An ethnobotanical survey of cosmetic plants used in Marmara region of Türkiye

Étude ethnobotanique des plantes cosmétiques utilisées dans la région de Marmara en Turquie

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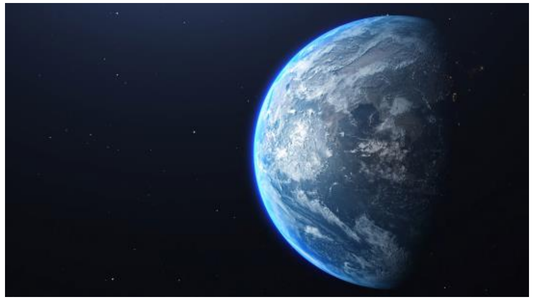

The Marmara Region connects Europe and Asia as a bridge in the northwest of Türkiye.

La région de Marmara relie l'Europe et l'Asie comme un pont au nord-ouest de la Turquie.

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Field studies in the research area

- **90 days** in total (90 jours au total)
- **126 villages** visited (126 villages visités)
- **348 people** were interviewed (348 personnes ont été interrogées)

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Ebru Özdemir

Etnhnobotanical research form for cosmetic plants

Formulaire de recherche ethnobotanique pour les plantes cosmétiques

Name, Surname: Nom, Prénom:

Phone number: Numéro de téléphone:

Research area: Domaine de recherche:

Age, Gender: Âge, sexe: Female/Male: Femme/Homme:

Education status: Niveau d'études:

Uneducated..........Primary school..........Secondary school........High school........University.........

Sans instruction.... École primaire.......... École secondaire.........Lycée.................. Université........

Interwiev date: Date de l'entretien:

Used part of the plant should be written for each categories

La partie utilisée de l'installation doit être indiquée pour chaque catégorie.

Preparation methods of herbal medicine should be written for each categories

Les méthodes de préparation des médicaments à base de plantes doivent être décrites pour chaque catégorie.

Cosmetic plants Plantes cosmétiques

Plants used for hair loss Plantes utilisées contre la chute des cheveux

Plants used for make-up Plantes utilisées pour le maquillage

Plants used for hair dandruff Plantes utilisées contre les pellicules

Plants used as hair lightener Plantes utilisées comme éclaircissant pour les cheveux

Plants used as hair dye Plantes utilisées comme teinture capillaire

Plants used for skin hyperpigmentation Plantes utilisées pour l'hyperpigmentation cutanée

Plants used for skin cleanser Plantes utilisées pour nettoyer la peau

Plants used for skin crack Plantes utilisées pour les gerçures cutanées

Plants used as moisturizer Plantes utilisées comme hydratant

Plants used for eyebrow & eyelash growth Plantes utilisées pour la croissance des sourcils et des cils

Plants used for dark circles under the eyes Plantes utilisées pour atténuer les cernes sous les yeux

Interviews were made at busy places like bazaars, tea houses, farms, gardens, fields, the houses of the local people.

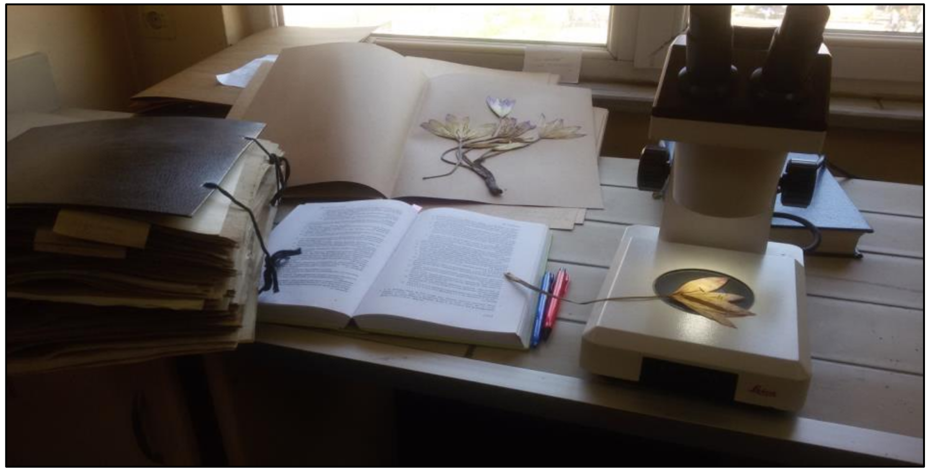
Les entretiens ont été menés dans des lieux fréquentés comme les bazars, les salons de thé, les fermes, les jardins, les champs et les maisons des habitants.

A reference plant collection was prepared by collecting and drying naturally growing plants in the study area.

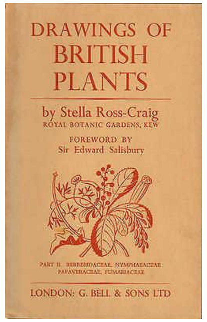
Une collection de plantes de référence a été constituée en collectant et en séchant des plantes poussant naturellement dans la zone d'étude.

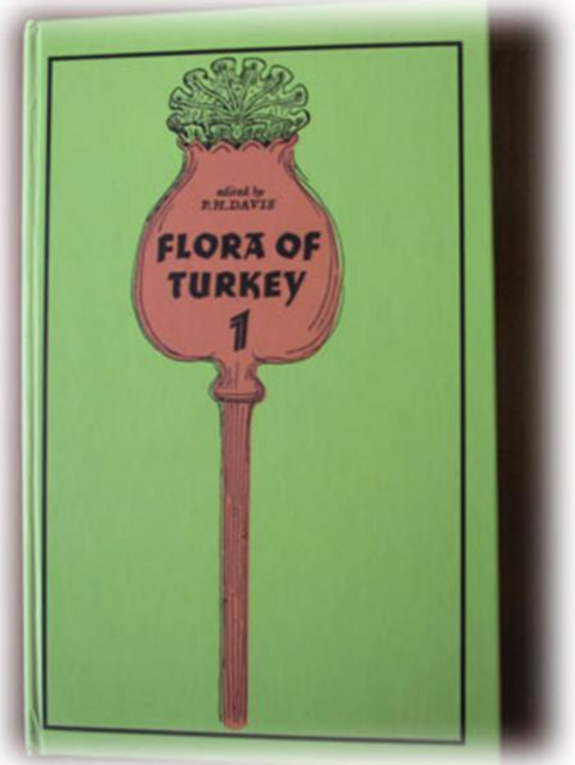
2. Herbarium studies

Études d'herbier



- The collected plant samples were pressed, dried, stored in a deep-freezer at -28 ° C for one week, then stucked to cartons.
- Les échantillons de plantes collectés ont été pressés, séchés, conservés dans un congélateur à -28 °C pendant une semaine, puis collés sur des cartons.





Flora of Turkey and the East Aegean Islands was written under the editorship P.H.Davis. It is in English and has 11 volumes. The first volume is dated 1965 and the last volume is 2000.

La Flore de Turquie et des îles de la mer Égée orientale a été rédigée sous la direction de P.H. Davis.Cet ouvrage en anglais compte 11 volumes.Le premier volume date de 1965 et le dernier de 2000.

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Benefits of Herbarium
Avantages de l'herbier

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Conservation:

- Preserves rare and endangered species for future generations
- Specimens can be used to provide samples of DNA to study relationships and evolutionary processes.
- Very valuable sources of information for ethno-botanical researches
- Herbariums preserve the national plant wealth
- Provide scientific information to the public

Conservation :

- Préserve les espèces rares et menacées pour les générations futures.
- Les spécimens peuvent être utilisés pour fournir des échantillons d'ADN permettant d'étudier les relations phylogénétiques et les processus évolutifs.
- Sources d'information très précieuses pour la recherche ethnobotanique.
- Les herbiers préservent le patrimoine végétal national.
- Diffusion d'informations scientifiques auprès du public.

Qian, L. S., Chen, J. H., Deng, T., & Sun, H. (2020). Plant diversity in Yunnan: Current status and future directions. *Plant diversity*, *42*(4), 281-291.

Benefits of Herbarium

Avantages de l'herbier

Research

- <u>Serve as a permanent record allowing anyone to go back and check the identification, re-sample or repeat research-with Index Herbarium Code and number!!!!</u>
- It is used to identify species. How the plants look like? environment, flowering time, habitat and ecological data, proper collection time
- Enables study of plant anatomy, morphology, palynology and systematics
- It is important to discover new species
- Floras and Monographs are prepared by studying the herbarium specimens

Educations: Teaches botany, ecology and environmental science

Recherche

- <u>L'herbier sert de registre permanent permettant à quiconque de vérifier l'identification, de prélever de nouveaux échantillons ou de répéter les recherches, grâce à son code et son numéro d'indexation.</u>
- Il est utilisé pour identifier les espèces. On y trouve des informations sur l'apparence des plantes, leur environnement, leur période de floraison, leur habitat, les données écologiques et la période de collecte optimale.
- Il permet l'étude de l'anatomie, de la morphologie, de la palynologie et de la systématique des plantes.
- Il est important pour la découverte de nouvelles espèces.
- Les flores et les monographies sont préparées à partir de l'étude des spécimens d'herbier.

Enseignement: L'herbier enseigne la botanique, l'écologie et les sciences de l'environnement.

Major Herbarium in Turkiye

Principal Herbier de Turkiye

Name	Code	Taxa number
Ankara University Science Faculty Herbarium	ANK	c. 200.000
Istanbul University Faculty of Pharmacy Herbarium	ISTE	c. 105.000
Ankara University Faculty of Pharmacy Herbarium	AEF	c. 25.000
İst. Üniversitesi Fen Fak. Herbaryumu	ISTF	c. 40.000
Ege Üniv. Ecz. Fak. Herbaryumu	IZEF	c. 20.000
Herbarium of Altınbaş University Faculty of Pharmacy	HERA	c. 2.000

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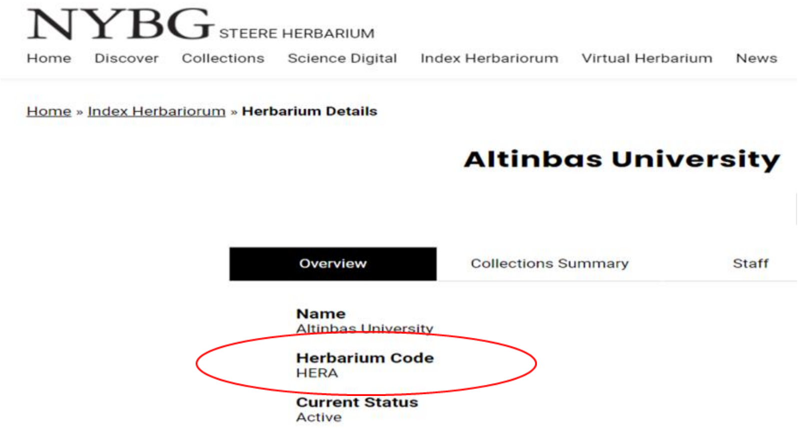
Pulicaria dysenterica

HERA registered with the international code in New York Botanical Garden (October 1, 2019). In HERA, there are approximately 2000 plant specimens, especially of medicinal and cosmetic plants.

HERA est enregistré sous le code international au Jardin botanique de New York (1er octobre 2019). HERA abrite environ 2 000 spécimens de plantes, principalement des plantes médicinales et cosmétiques.



HERA

HERBARIUM OF ALTINBAŞ UNIVERSITY
FACULTY OF PHARMACY - ISTANBUL / TURKEY

DÜAGEM

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DÜAGEM Herbarium

ALTINBAŞ ÜNİVERSİTESİ

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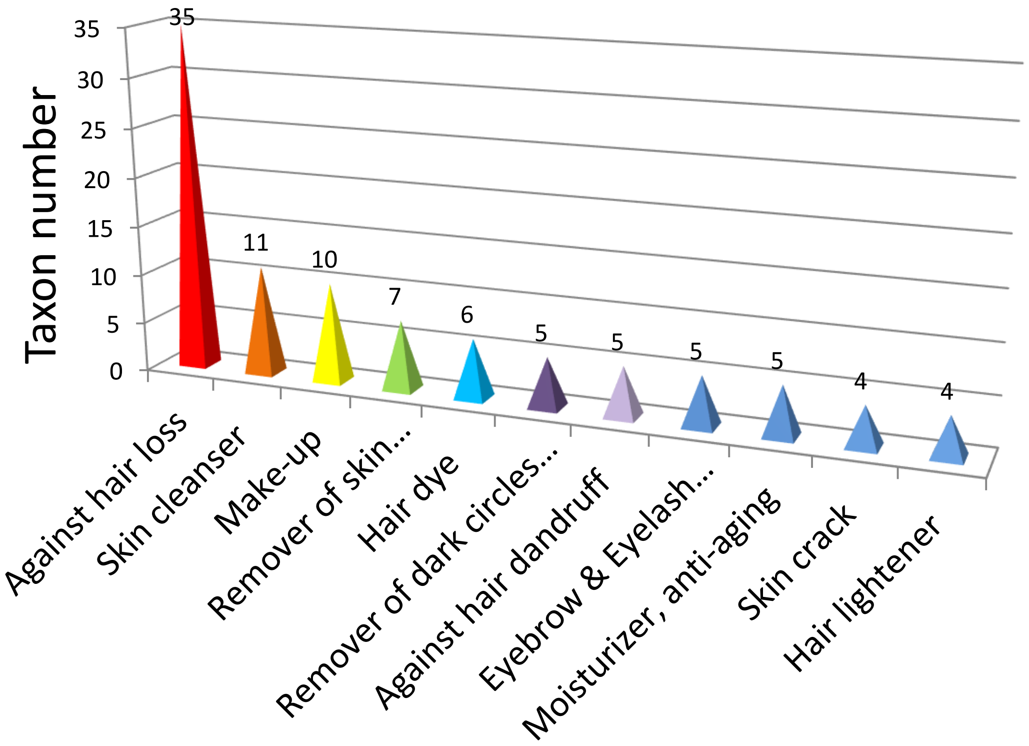
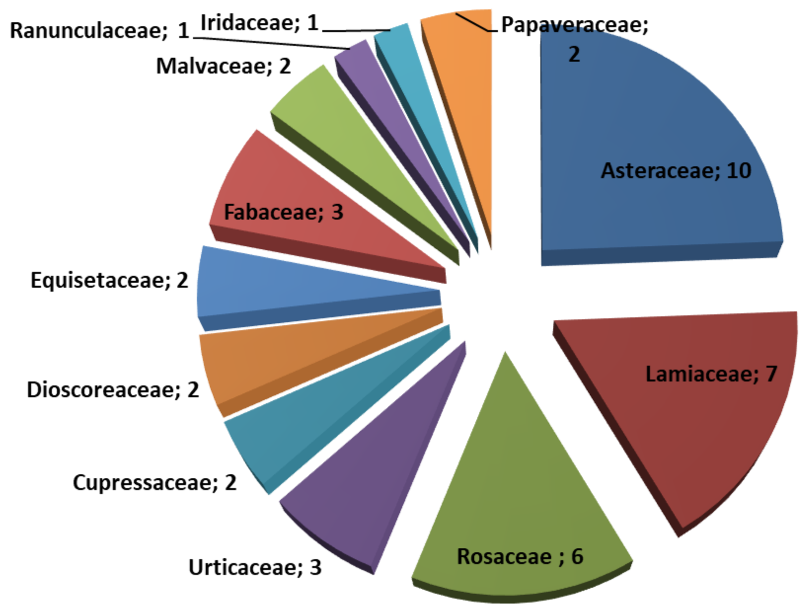
3. Evaluation of findings
Évaluation des résultats

During the Ethnobotanical study in Marmara region, 63 cosmetic plant taxa belonging to 37 families were determined. The 11 different usage categories are indicated in the chart.

Lors d'une étude ethnobotanique menée dans la région de Marmara, 63 taxons de plantes cosmétiques appartenant à 37 familles ont été recensés. Leurs 11 catégories d'utilisation sont indiquées dans le tableau.



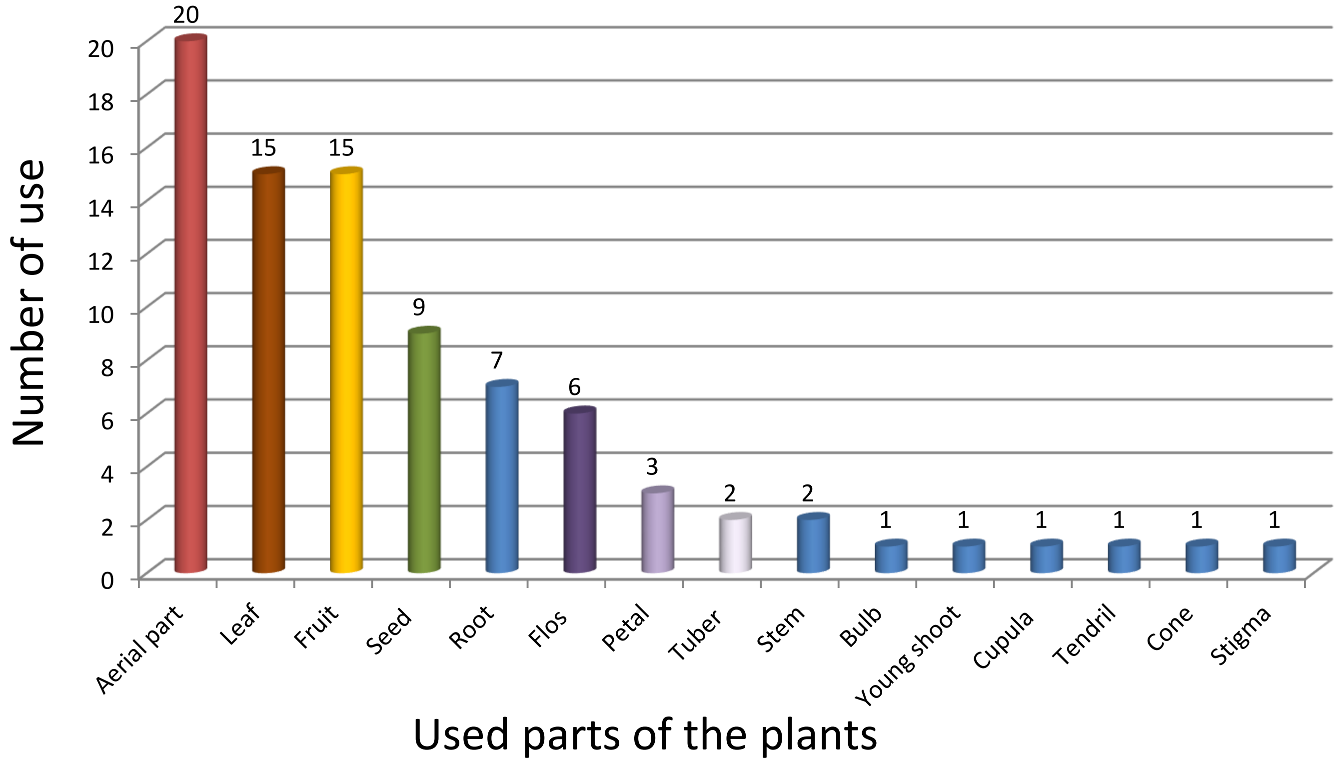


Used parts of the plants

Parties utilisées des plantes

The most commonly used parts of the plants were the aerial part (20) of use reports. Other parts were leaf (15), fruit (15), seed (9), root (7), flos (6), tuber (2), stem (2), bulb (1), young shoot (1), cupula (1), tendril (1), cone (1), stigma (1).

Les parties les plus fréquemment utilisées des plantes étaient la partie aérienne (20) des rapports d'utilisation. Les autres parties étaient la feuille (15), le fruit (15), la graine (9), la racine (7), la fleur (6), le tubercule (2), la tige (2), le bulbe (1), la jeune pousse (1), la cupule (1), la vrille (1), le cône (1), le stigmate (1).



Informant consensus factor (FIC)

Facteur de consensus des informateurs (FIC)

FIC=(Nur-Nt)/(Nur-1)
Nur= The number of citations in each category
(Le nombre de citations dans chaque catégorie)
Nt= The number of the species used in each category
Le nombre d'espèces utilisées dans chaque catégorie)

The reported uses were grouped into **11 categories.** Usages against **hyperpigmentation had the highest FIC value (0.82)**. High FIC value shows that the plants which had been used for **hyperpigmentation** are presumed to be the most effective.

Les usages recensés ont été regroupés en 11 catégories. Les usages contre l'hyperpigmentation présentaient la valeur FIC la plus élevée (0,82).Une valeur FIC élevée indique que les plantes utilisées contre l'hyperpigmentation sont présumées être les plus efficaces.

The use value a quantitative method that demonstrates the relative importance of species known locally.

La valeur d'usage est une méthode quantitative qui démontre l'importance relative des espèces connues localement.

UV=U/N
UV= Use value of a species (valeur d'usage d'une espèce)
U= The number of citations per species (Le nombre de citations par espèce)
N= The number of total informants (Le nombre total d'informateurs)

Most commonly used plants (Les plantes les plus couramment utilisées)

Hypericum perforatum 0.83
Helichrysum plicatum 0.80
Juglans regia 0.77
Thymus sipyleus 0.75
Pulicaria dysenterica 0.72
Sideritis montana 0.70
Glycrrhiza glabra 0.69
Arbutus unedo 0.68

Rosa canina 0.68
Laurus nobilis 0.65
Malva sylvestris 0.64
Achillea arabica 0.55
Plantago lanceolata 0.53
Rhus coria 0.52
Trachystemon orientalis 0.45
Urtica dioica 0.51

Crocus sativus L. (Stigma)

Saffron stigmas were crushed and used as eyeshadow
(Les stigmates de safran étaient broyés et utilisés comme fard à paupières)

Two main natural carotenoids of saffron, crocin and crocetin, are responsible for its color
(Deux caroténoïdes naturels principaux du safran, la crocine et la crocétine, sont responsables de sa couleur)

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Plants used as hair dye

Plantes utilisées comme teinture capillaire

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1. *Alcea setosa* Alef. (Aarial part)
2. *Cistus creticus* L. (Seed)
3. *Cornus mas* L. (Leaf)
4. *Juglans regia* L. (Fruit pericarpium)
5. *Mespilus germanica* L. (Fruit)
6. *Quercus cerris* L. (Fruit, cupula)

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Plantes tinctoriales naturelles

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IUFS Journal of Biology
IUFS J Biol 2015, 74(2): 25-32
Research Article

Natural dye plants in Kepsut (Balıkesir, Turkey)

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Abstract

Natural dyes have traditionally been used in Anatolia for several years. An ethnobotanical study was conducted between 2012 and 2015 in order to determine wild plants used in Kepsut, which is a district of Balıkesir province in the Marmara region of Turkey. The purpose of this research was to get acquainted to know plants used as dyes in Kepsut, Balıkesir. The regional name of the plants, the parts used for dye production, and obtained colors have been reported. According to the results of the identification, 21 plant species belongs to 14 families were used as dye source in Kepsut. Used parts of species were aerial part, cortex, flower, fruit, gall, leaf, and root. Also 5 mixture receipts for dyeing were reported in Kepsut.

Key words: Ethnobotany, natural dye, Kepsut, Balıkesir, Turkey.
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Original Article

Natural dye plants in Savaştepe (Balıkesir, Turkey)

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Abstract: Natural dyes are recently becoming object of consumer interests because of the harmful effects of the synthetic dyes. The dyeing with natural colourants was one of the oldest techniques practiced by the ancient civilization people. An ethnobotanical study was conducted between 2012 and 2015 in order to determine wild plants used in Savaştepe. Savaştepe is a town and district of Balıkesir Province in the Marmara region of Turkey. With this study, we aimed to document traditional uses of dye plants in Savaştepe (Balıkesir). According to the results of the identification, 15 species belonging to 12 families were used as dye source. Used parts of plants were flowers, fruits, leaves, roots, seeds and galls.

Key words: Ethnobotany, natural dye, Savaştepe, Balıkesir, Turkey.

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II. Phytochemistry &ICOA studies

Phytochimie & Études de l'ICOA

LE STUDIUM Conference Plant rediscovery with advanced tools for well-being applications, 09.10.2023, Orleans, France

An ethnobotanical survey of cosmetic plants used in Marmara region of Türkiye

LE STUDIUM
Loire Valley
Institute for Advanced Studies

DÜAGEM
DOĞAL ÜRÜNLER AR-GE MERKEZİ

SMART LOIRE VALLEY (SLV) Programme

LE STUDIUM VISITING RESEARCHER Award
Call for Applications 2024

Part B – Scientific proposal

Applicant: Assoc. Prof. Dr. Ebru Özdemir Nath

Host Research institute/Laboratory: University of Orleans/ The Institute of Organic and Analytical Chemistry (ICOA)

Host Scientist: Prof. Dr. Emilie Destandau

Title of the project: Chemical analysis of some plants used for cosmetic purposes in Turkish ethnobotanical studies and their in vitro physiological effects on human origin cell lines.

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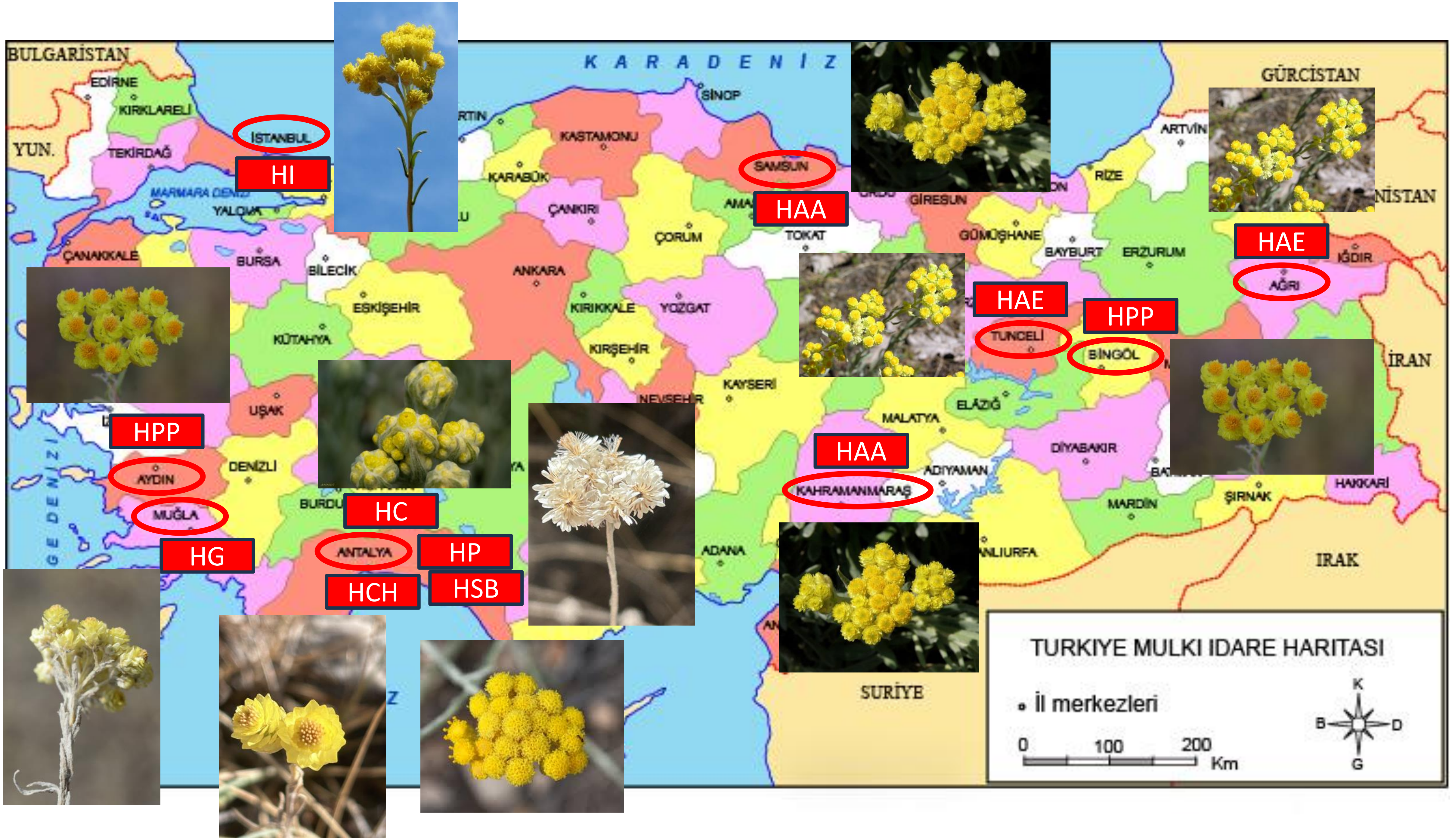
Assoc. Prof. Dr. Ebru Özdemir Nath ebru.ozdemir@altinbas.edu.tr



BULGARİSTAN
EDİRNE
KIRKLARELİ
YUN.
TEKİRDAĞ
İSTANBUL
HI
MARMARA DENİZİ
YALOVA
ÇANAKKALE
BURSA
BİLECİK
ESKİŞEHİR
KÜTAHYA
UŞAK
HPP
AYDIN
DENİZLİ
MUĞLA
HG
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K A R A D E N İ Z
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ÇANKIRI
ÇORUM
ANKARA
KIRIKKALE
YOZGAT
KIRŞEHİR
NEVŞEHİR
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SAMSUN
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BAYBURT
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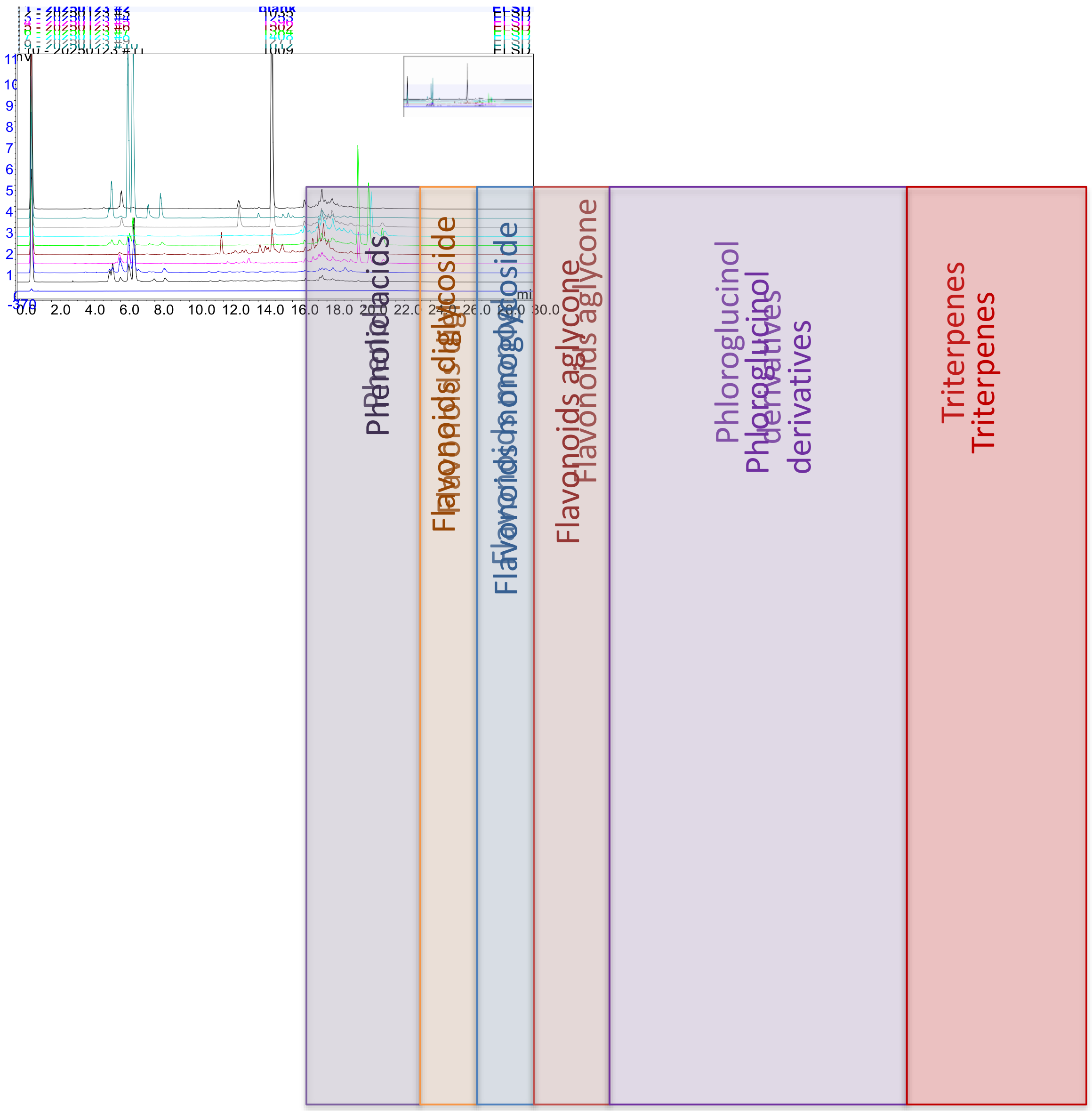
HERA CODE	Plant species	Endemism
October 2023, 1/10 EtOH extracts		
HERA 1035	*Helichrysum arenarium* subsp. *aucheri* (Boiss.) P.H.Davis & Kupicha (HAA)	**Endemic**
HERA 1038	*Helichrysum arenarium* subsp. *erzincanicum* P.H.Davis & Kupicha (HAE)	**Endemic**
HERA 1009	*Helichrysum plicatum* subsp. *plicatum* DC. (HPP)	
HERA 1012	Helichrysum graveolens (M.Bieb.) Sweet (HG)	
January 2025, 1/10 EtOH extracts		
HERA 1384	*Helichrysum chionophilum* Boiss. & Balansa (HCH)	**Endemic**
HERA 1396	*Helichrysum compactum* Boiss.(HC)	**Endemic**
HERA 1502	*Helichrysum italicum* (Roth) G.Don (HI)	
HERA 1408	*Helichrysum pamphylicum* P.H.Davis & Kupicha (HP)	**Endemic**
HERA 1271	*Helichrysum stoechas* subsp. *barrelieri* (Ten.) Nyman (HSB)	
HERA 1035	*Helichrysum arenarium* subsp. *aucheri* (Boiss.) P.H.Davis & Kupicha (HAA)	**Endemic**
HERA 1253	*Helichrysum arenarium* subsp. *erzincanicum* P.H.Davis & Kupicha (HAE)	**Endemic**
HERA 1009	*Helichrysum plicatum* subsp. *plicatum* DC. (HPP)	
HERA 1012	*Helichrysum graveolens* (M.Bieb.) Sweet (HG)	
January 2025, raw plants		
HERA 1376	*Helichrysum plicatum* subsp. *plicatum* DC. (HPP)	
HERA 1503	*Helichrysum arenarium* subsp. *aucheri* (Boiss.) P.H.Davis & Kupicha (HAA)	**Endemic**
HERA 1504	*Helichrysum arenarium* subsp. *erzincanicum* P.H.Davis & Kupicha (HAE)	**Endemic**
HERA 1506	*Helichrysum graveolens* (M.Bieb.) Sweet (HG)	

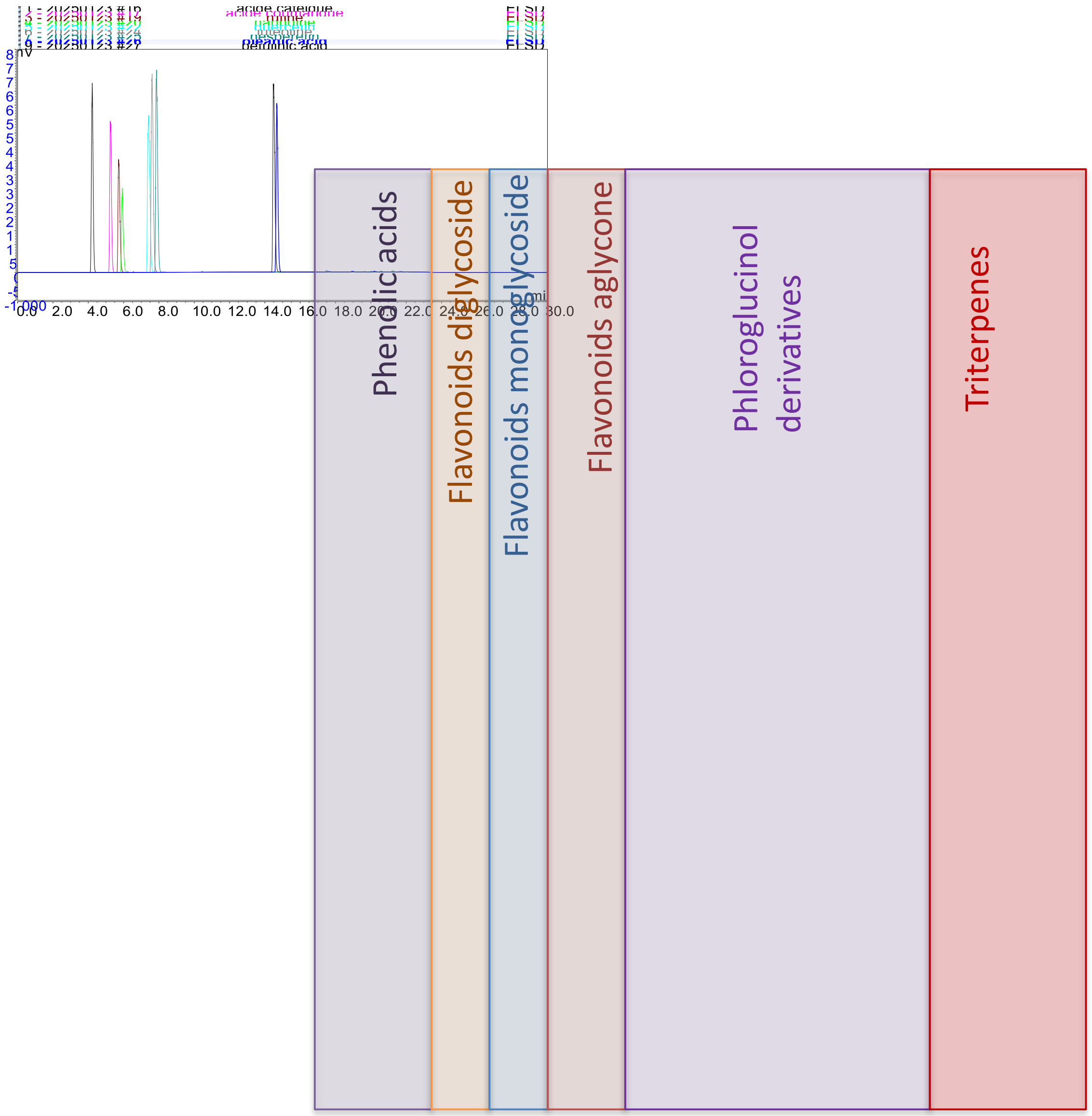
- *Helichrysum* Mill. is a member of the Asteraceae family (L'Helichrysum Mill. est un membre de la famille des Astéracées).
- The genus *Helichrysum* is represented by 656 taxa in the world (Le genre Helichrysum est représenté par 656 taxons dans le monde).
- 34 taxa grow naturally in the flora of Türkiye and 17 taxa are endemic (34 taxons poussent naturellement dans la flore de Turquie et 17 taxons sont endémiques).

Ethanol extracts of *Helichrysum* species (Extraits éthanoliques d'espèces d'Helichrysum)



ELSD

Standards (Normes)



ELSD

HPTLC

NMR
1H and 1H-13C HSQC

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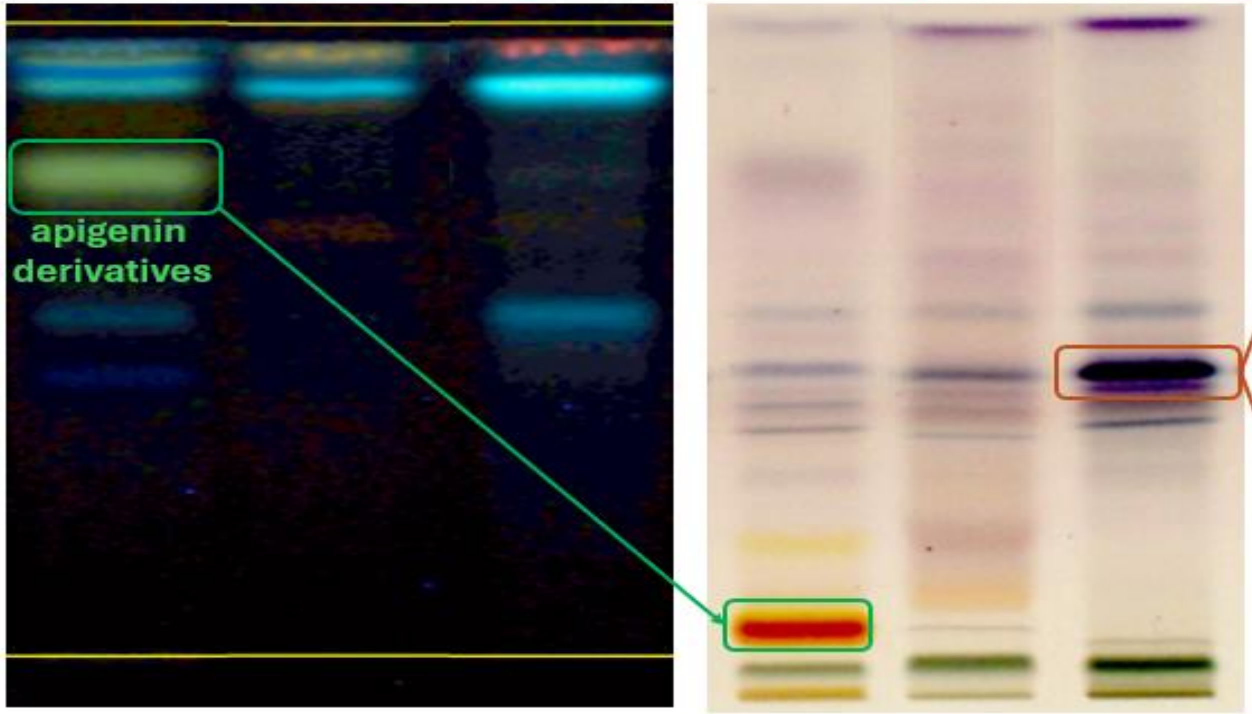
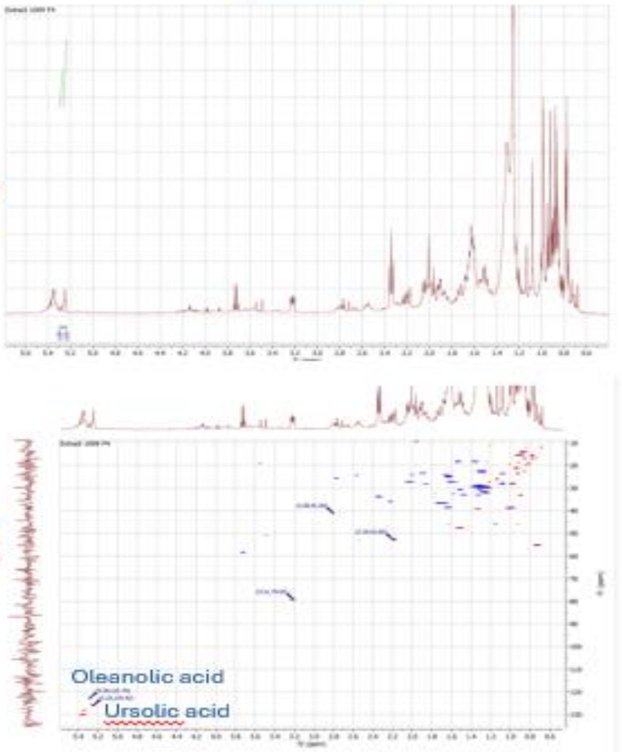
H.graveolens *H.italicum* *H. plicatum* *H.graveolens* *H.italicum* *H. plicatum*

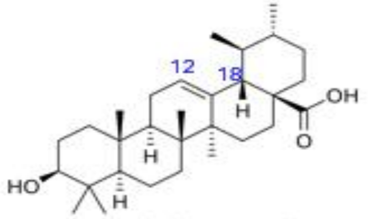


Merck, TLC Silica gel 60 F_{254}
Eluent :
AcOEt/HCOOH/CH3COOH/H2O
(100/11/11/27)
Revelation : Neu/PEG 366 nm

Merck, TLC Silica gel 60 F_{254}
Eluent :
Revelation : Sulfuric anisaldehyde

H. plicatum





Characteristic chemical shifts

n°	Oleanolic acid δC, δH	Ursolic acid δC, δH
12	122.7, 5.28	125.9, 5.25
18	41.0, 2.80	52.6, 2.20

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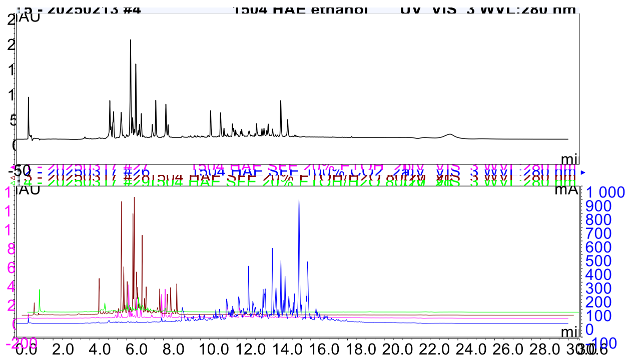
Supercritical fluid extraction (SFE)

UV-280nm

Helichrysum arenarium subsp. *erzincanicum*



III.Biological activity studies
Études sur l'activité biologique

- Anti-aging enzyme activities

(Activités enzymatiques anti-âge)

- Cox 1-2 inhibition activities

(Activités d'inhibition de la Cox 1-2)

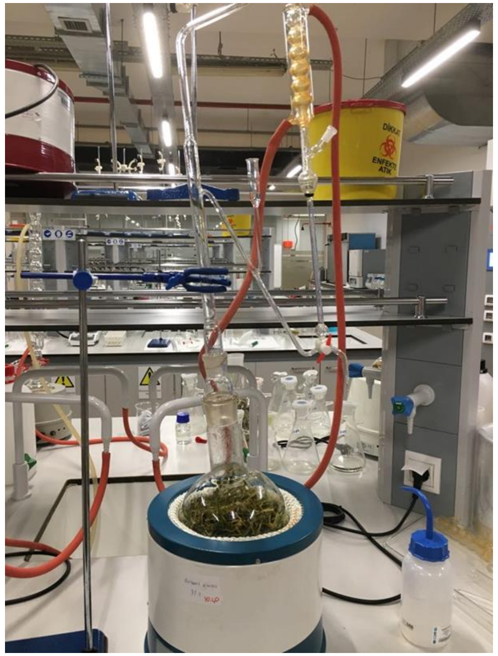
- Cell culture studies

(études de culture cellulaire)

We conduct in vitro anti-aging enzyme activity studies and cell culture studies on traditional cosmetic plants.

Nous menons des études in vitro sur l'activité enzymatique anti-âge et des études de culture cellulaire sur des plantes cosmétiques traditionnelles.

DÜAGEM-313-Natural Products Biological Activity R&D Laboratory



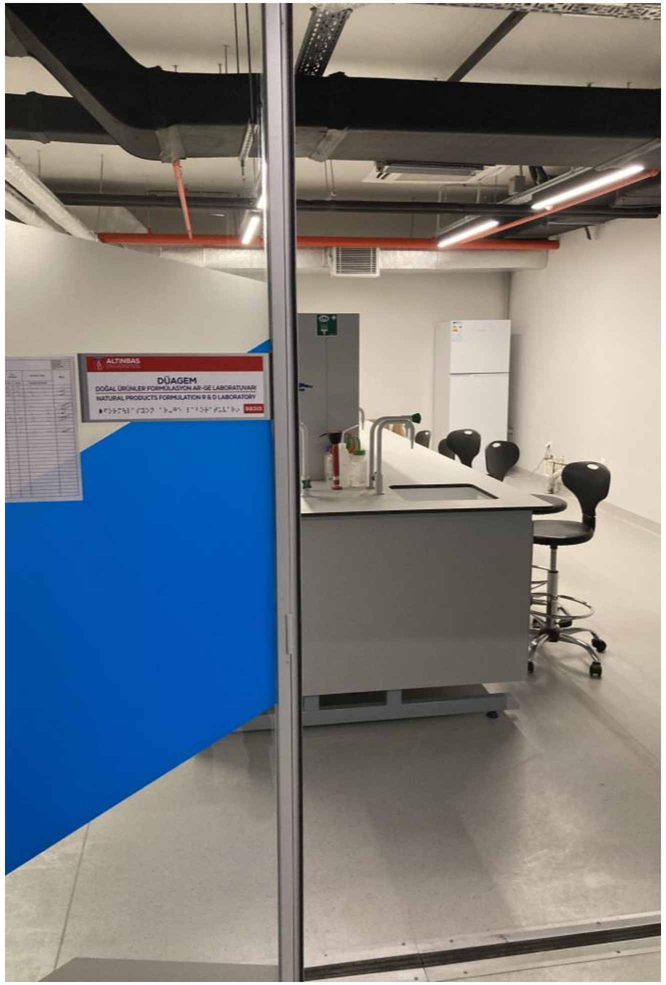
DÜAGEM-312-Natural Products Pharmacological Activity R&D Laboratory

Assoc. Prof. Dr. Ebru Özdemir Nath ebru.ozdemir@altinbas.edu.tr

DÜAGEM 316, Dry-air drying laboratory of natural products plants

Laboratoire de séchage à l'air sec de produits végétaux naturels

Assoc. Prof. Dr. Ebru Özdemir Nath ebru.ozdemir@altinbas.edu.tr



DÜAGEM 315, Natural Products Formulation R&D Laboratory

Laboratoire de R&D en formulation de produits naturels

Assoc. Prof. Dr. Ebru Özdemir Nath ebru.ozdemir@altinbas.edu.tr

Anti-aging enzyme activities (Activités enzymatiques anti-âge)

Hyaluronidase

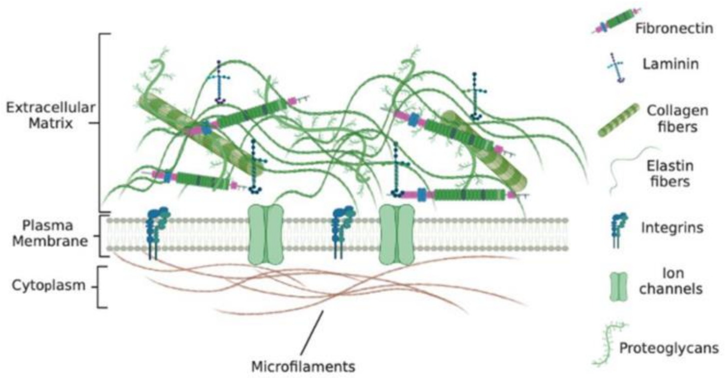
Tyrosinase

The current anti-aging strategy focuses on the inhibition of these enzymes.
The plant extracts may the potential to be a natural inhibitor.

La stratégie anti-âge actuelle se concentre sur l'inhibition de ces enzymes.Les extraits de plantes pourraient potentiellement constituer un inhibiteur naturel.

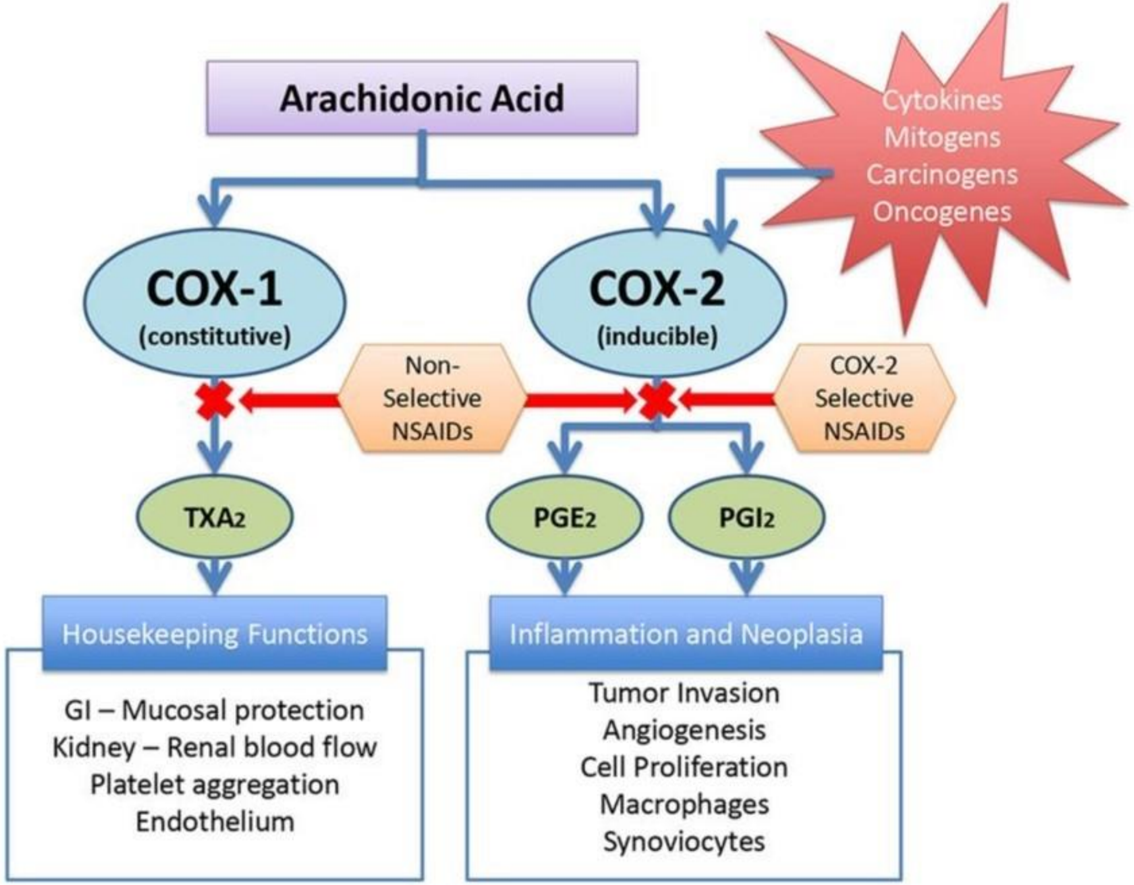
Elastase

Collagenase



Cyclooxygenase (COX) enzyme studies

- Cyclooxygenase (COX) enzymes convert arachidonic acid into prostaglandins. Overproduction of COX enzymes can result in excessive prostaglandin synthesis, producing persistent tissue inflammation and hastening the aging process.
- Les enzymes cyclooxygénases (COX) transforment l'acide arachidonique en prostaglandines. Une surproduction de ces enzymes peut entraîner une synthèse excessive de prostaglandines, provoquant une inflammation tissulaire persistante et accélérant le processus de vieillissement.
- Specially COX-2 inhibitors could conceivably intervene and delay the aging process.
- Plus particulièrement, les inhibiteurs de la COX-2 pourraient potentiellement intervenir et retarder le processus de vieillissement.



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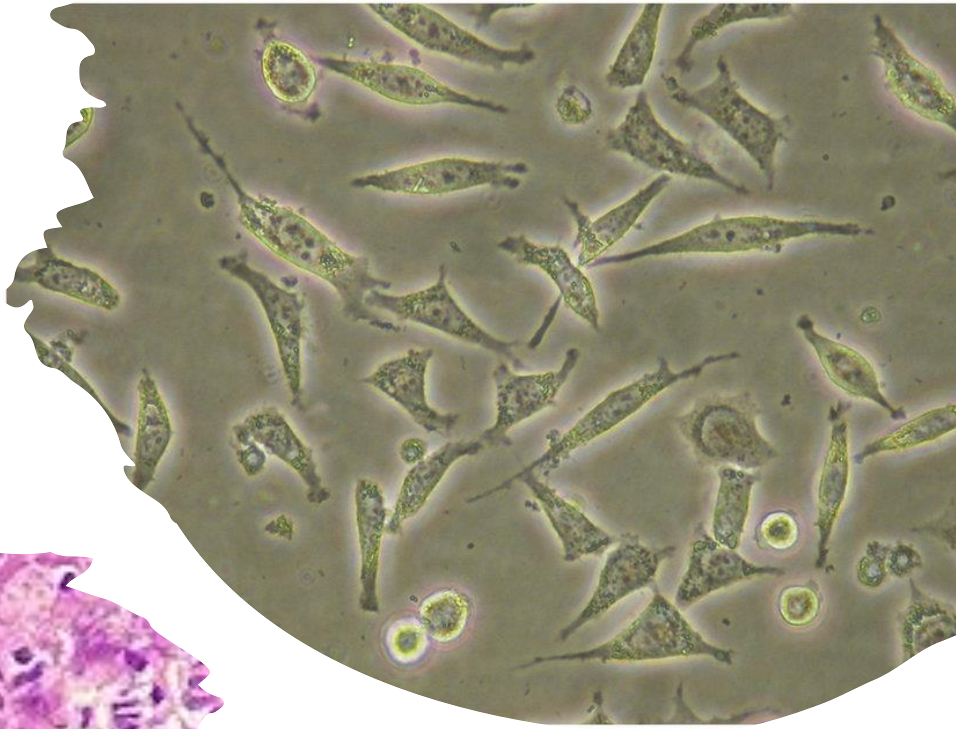
Cell culture studies
études de culture cellulaire

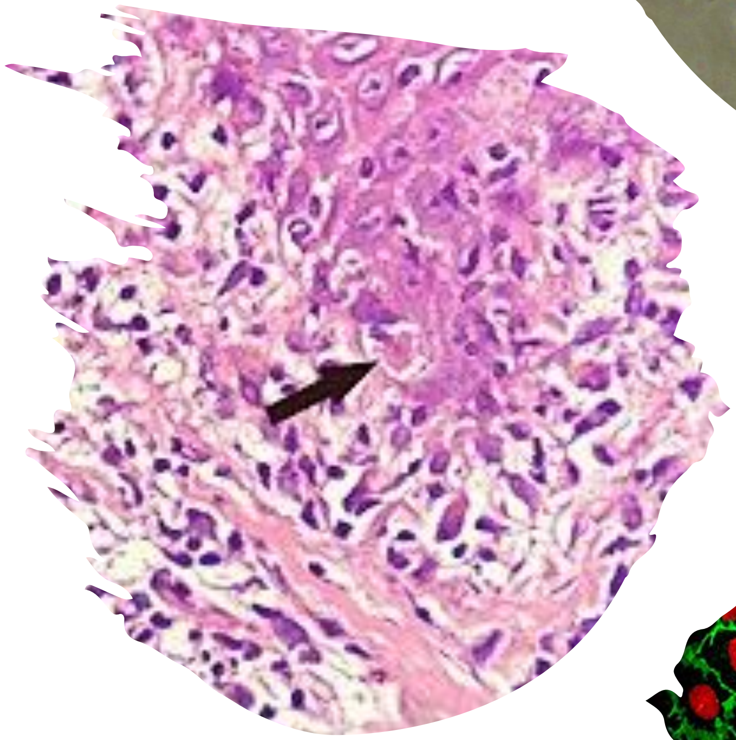


- **HaCat Human Keratinocyte cells:** Skin renewal, inflammatory response, signs of aging, wrinkles, wound repair processes

Renouvellement cutané, réaction inflammatoire, signes du vieillissement, rides, processus de cicatrisation

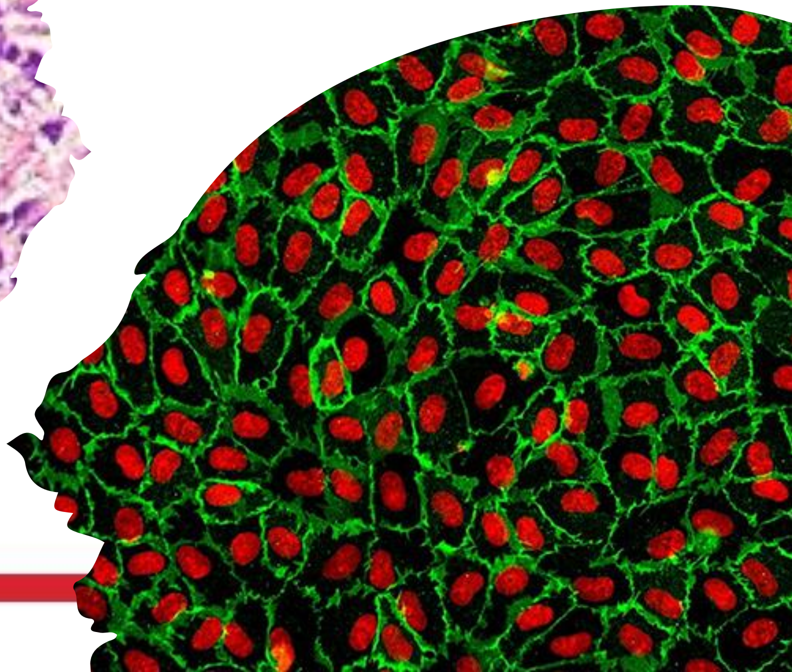
- **HuVeC Human Endothelial cells:** Improves Blood Circulation, Antioxidant, Anti-inflammatory, Promotes Cell Regeneration.

Améliore la circulation sanguine, antioxydant, anti-inflammatoire, favorise la régénération cellulaire.



- **L929 fibroblast cells:** Toxicity is measured by colorimetric methods using the MTT test in the evaluation of active substance exposure to L929 fibroblast cells, which is accepted within the scope of the ISO 10993-5 standard.

La toxicité est mesurée par des méthodes colorimétriques utilisant le test MTT dans l'évaluation de l'exposition des cellules fibroblastes L929 à la substance active, ce qui est accepté dans le cadre de la norme ISO 10993-5.



Pulicaria dysenterica cream

Crème pour Pulicaria dysenterica

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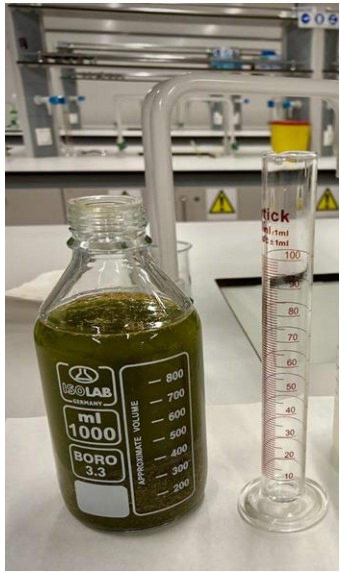

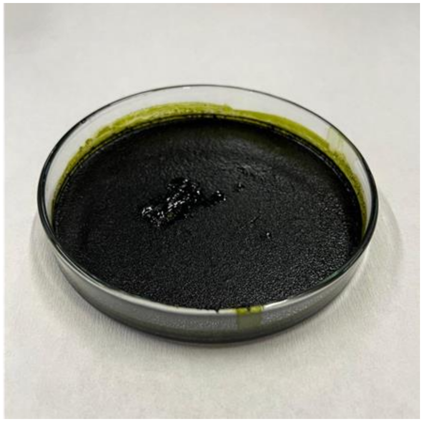

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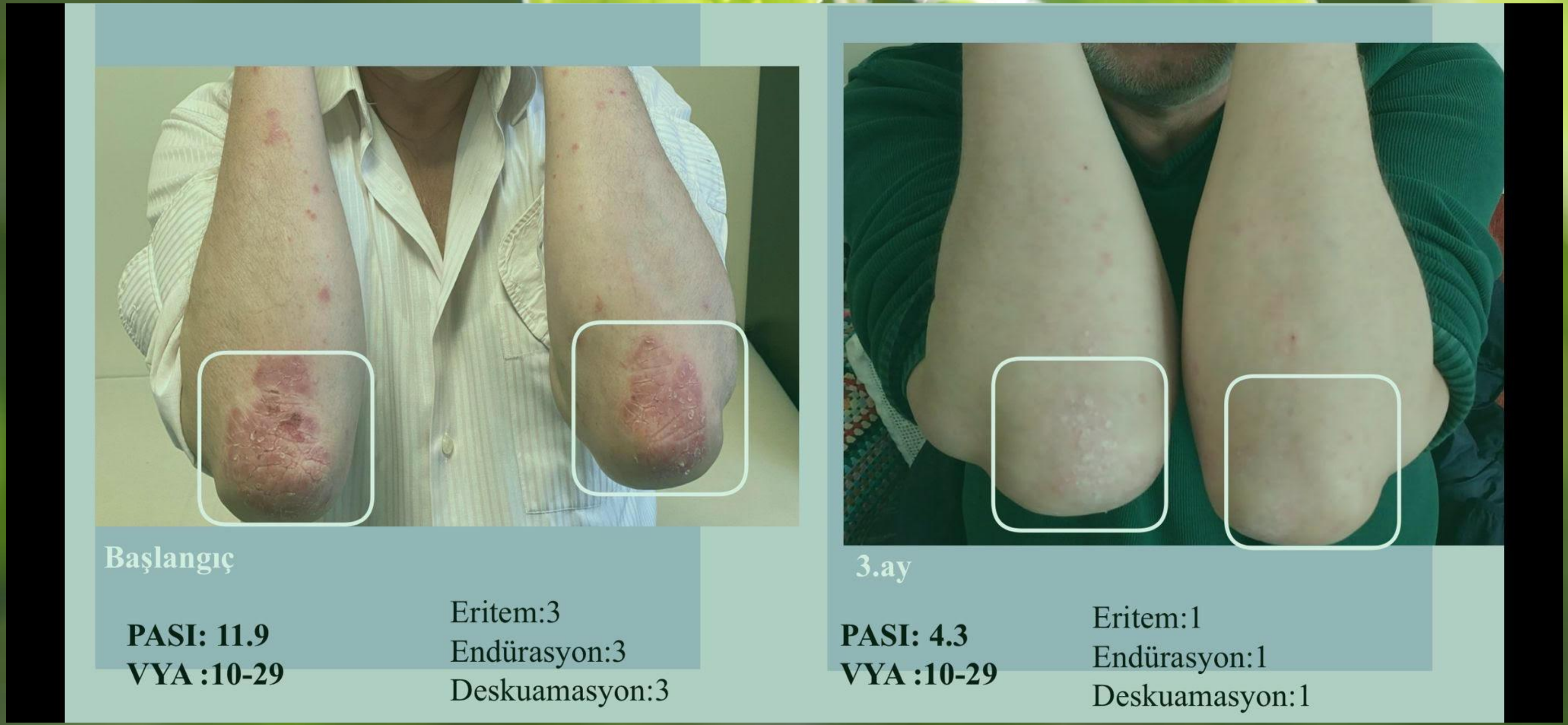
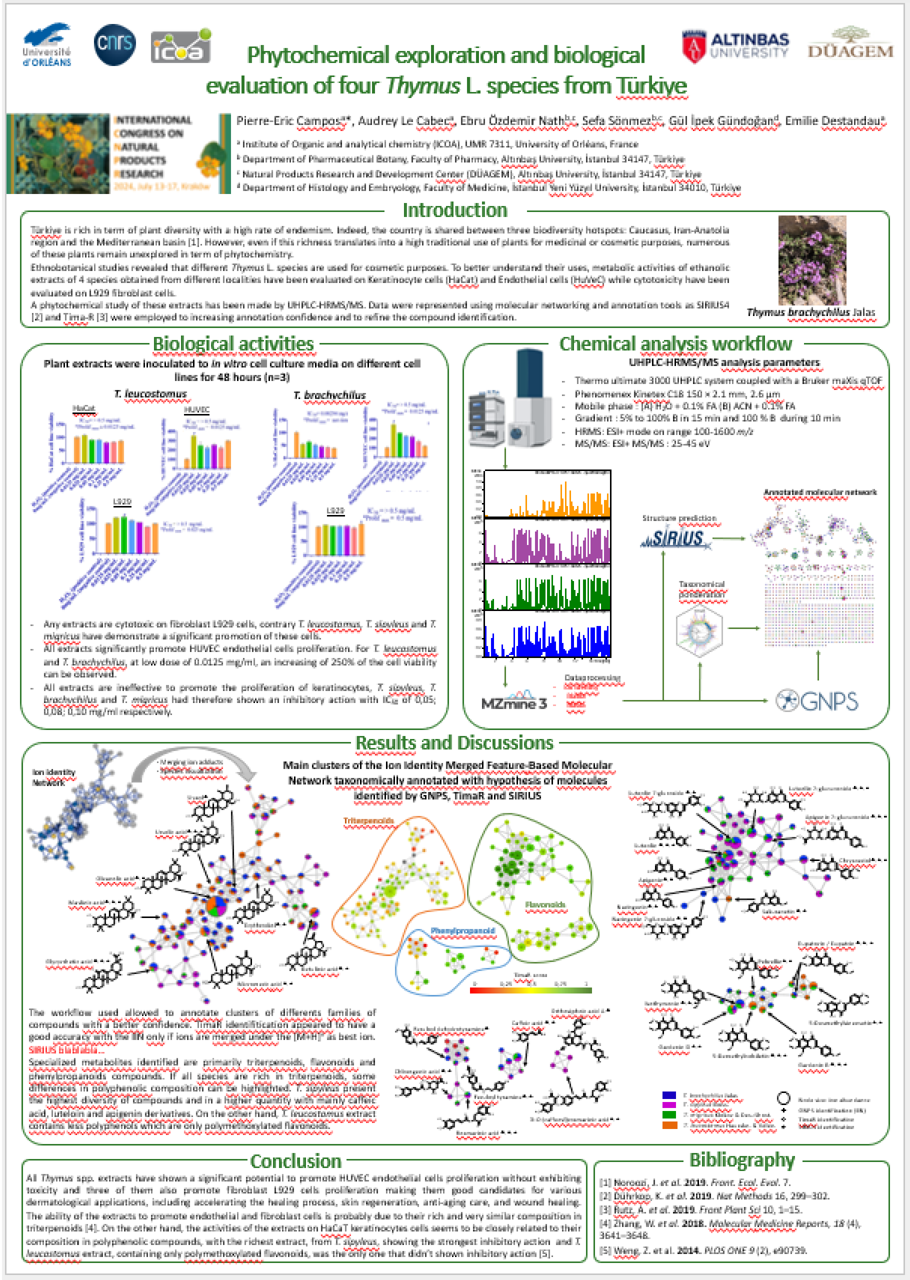
Psoriasis treatment (Traitement du psoriasis)

Viscum album



Thymus leucostomus (endemic)

Thymus migricus



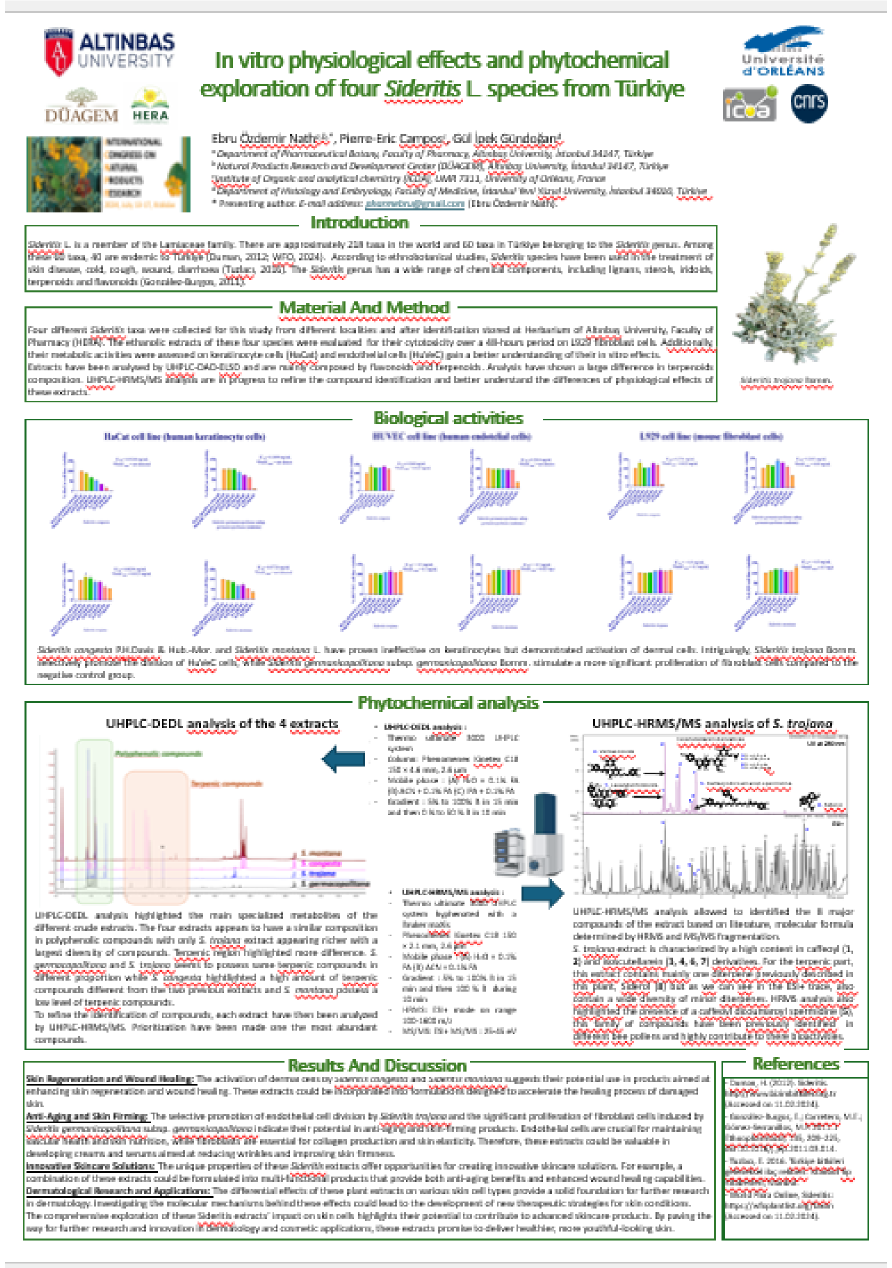
46 species in Türkiye,
19 species are endemic

Thymus sipyleus

Thymus brachychilus (endemic)

Sideritis congesta (endemic)

Sideritis montana



61 taxa in Türkiye

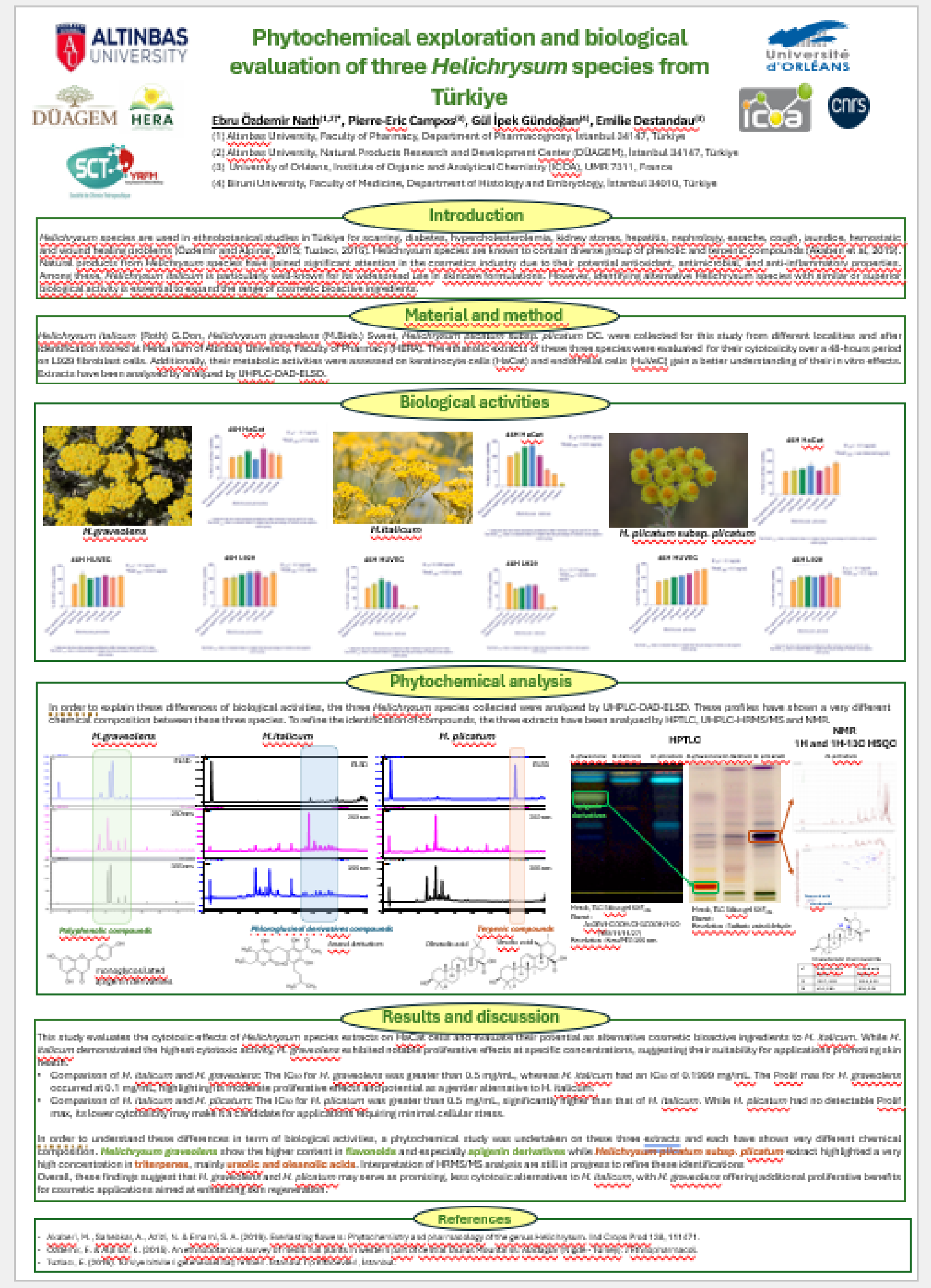
41 of them are endemic

Sideritis trojana (endemic)

Sideritis germanicopolitana (endemic)

Helichrysum graveolens

Helichrysum italicum



PHARMACIE Santé Université Paris Cité

Société de Chimie Thérapeutique YRFM

Université Paris Cité

32nd Young Research Fellows Meeting

24-26 February 2025

Helichrysum plicatum

University of Orleans, ICOA
Seminar 9.3.2025 and Herbarium workshop 18.3.2025

HERBARIUM OF ORLEANS UNIVERSITY (HERO)

Nom. :

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Loc. :

Dat. : **Alt.** :

Leg. : **Det.** :

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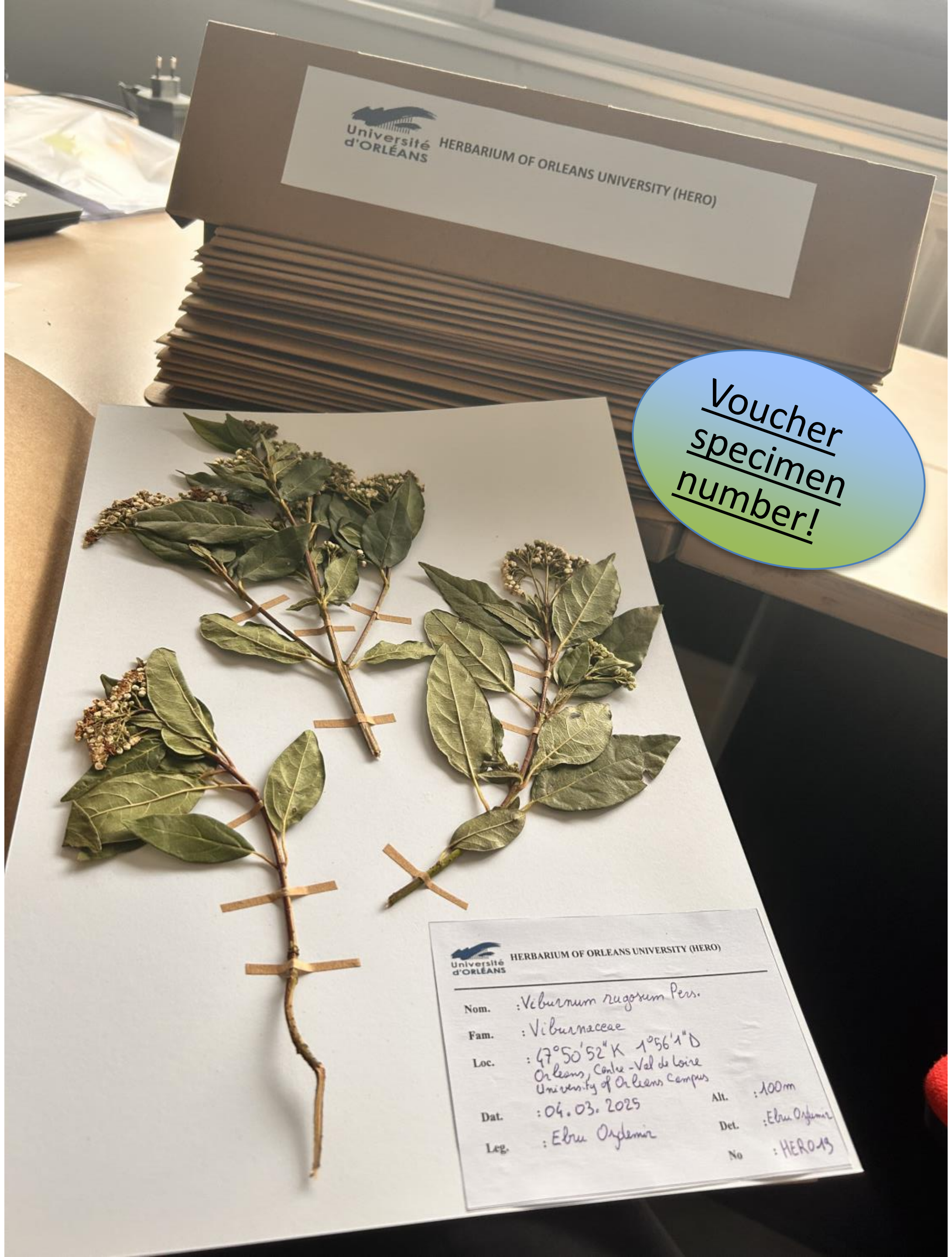

Integration of ethnobotany, phytochemistry and biological activity studiesin the design of phytocosmetic formulations

Phytochemical Investigation of *Helichrysum arenarium* subsp. *aucheri* (Boiss.) with Promising Wound Healing Potential and Targeted Extraction of Bioactive Compounds by Sequential Selective Supercritical Fluid Extraction (S^3FE)

Association Francophone pour l'Enseignement et la Recherche en Pharmacognosie

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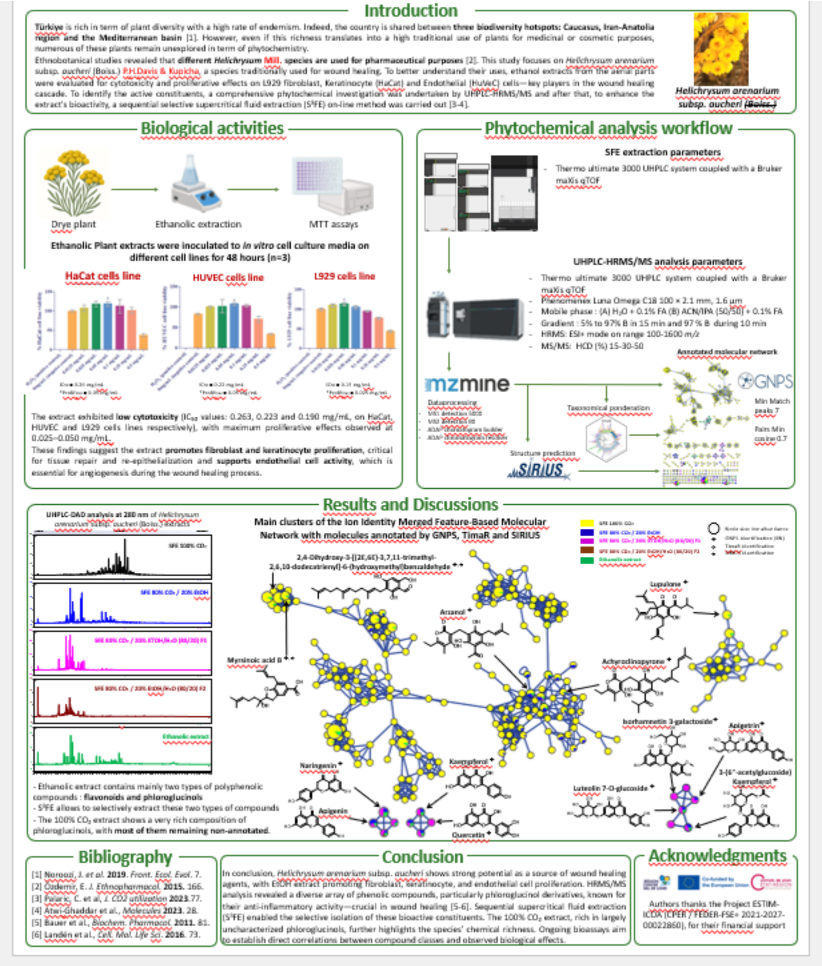
4ème Symposium International AFERP-STOLON
7-9 juil. 2025 Bordeaux (France)



Introduction

Türkiye is rich in term of plant diversity with a high rate of endemism. Indeed, the country is shared between **three biodiversity hotspots: Caucasus, Iran-Anatolia region and the Mediterranean basin** [1]. However, even if this richness translates into a high traditional use of plants for medicinal or cosmetic purposes, numerous of these plants remain unexplored in term of phytochemistry.

Ethnobotanical studies revealed that **different *Helichrysum* Mill. species are used for pharmaceutical purposes** [2]. This study focuses on *Helichrysum arenarium* subsp. *aucheri* (Boiss.) P.H.Davis & Kupicha, a species traditionally used for wound healing. To better understand their uses, ethanol extracts from the aerial parts were evaluated for cytotoxicity and proliferative effects on L929 fibroblast, Keratinocyte (HaCat) and Endothelial (HuVeC) cells—key players in the wound healing cascade. To identify the active constituents, a comprehensive phytochemical investigation was undertaken by UHPLC-HRMS/MS and after that, to enhance the extract's bioactivity, a sequential selective supercritical fluid extraction (S^3FE) on-line method was carried out [3-4].

Helichrysum arenarium subsp. *aucheri* (Boiss.)

Biological activities

Drye plant → Ethanolic extraction → MTT assays

Ethanolic Plant extracts were inoculated to *in vitro* cell culture media on different cell lines for 48 hours (n=3)

HaCat cells line · HUVEC cells line · L929 cells line

The extract exhibited **low cytotoxicity** (IC_{50} values: 0.263, 0.223 and 0.190 mg/mL, on HaCat, HUVEC and L929 cells lines respectively), with maximum proliferative effects observed at 0.025–0.050 mg/mL.

These findings suggest the extract **promotes fibroblast and keratinocyte proliferation**, critical for tissue repair and re-epithelialization and **supports endothelial cell activity**, which is essential for angiogenesis during the wound healing process.

Phytochemical analysis workflow

SFE extraction parameters

- Thermo ultimate 3000 UHPLC system coupled with a Bruker maXis qTOF

UHPLC-HRMS/MS analysis parameters

- Thermo ultimate 3000 UHPLC system coupled with a Bruker maXis qTOF
- Phenomenex Luna Omega C18 100 × 2.1 mm, 1.6 µm
- Mobile phase : (A) H_2O + 0.1% FA (B) ACN/IPA (50/50) + 0.1% FA
- Gradient : 5% to 97% B in 15 min and 97 % B during 10 min
- HRMS: ESI+ mode on range 100-1600 *m/z*
- MS/MS: HCD (%) 15-30-50

mzmine · Dataprocessing · Taxonomical ponderation · Structure prediction · SIRIUS · Annotated molecular network · GNPS · Min Match peaks 7 · Pairs Min cosine 0.7

Results and Discussions

UHPLC-DAD analysis at 280 nm of *Helichrysum arenarium* subsp. *aucheri* (Boiss.) extracts

Main clusters of the Ion Identity Merged Feature-Based Molecular Network with molecules annotated by GNPS, TimaR and SIRIUS

- Ethanolic extract contains mainly two types of polyphenolic compounds : **flavonoids and phloroglucinols**
- S^3FE allows to selectively extract these two types of compounds
- The 100% CO_2 extract shows a very rich composition of phloroglucinols, with **most of them remaining non-annotated**.

Bibliography

[1] Noroozi, J. et al. **2019**. *Front. Ecol. Evol.* 7.
[2] Özdemir, E. *J. Ethnopharmacol.* **2015.** 166.
[3] Palaric, C. et al, *J. CO2 utilization* **2023**.77.
[4] Atwi-Ghaddar et al., *Molecules* **2023**. 28.
[5] Bauer et al., *Biochem. Pharmacol.* **2011.** 81.
[6] Landén et al., *Cell. Mol. Life Sci.* **2016**. 73.

Conclusion

In conclusion, *Helichrysum arenarium* subsp. *aucheri* shows strong potential as a source of wound healing agents, with EtOH extract promoting fibroblast, keratinocyte, and endothelial cell proliferation. HRMS/MS analysis revealed a diverse array of phenolic compounds, particularly phloroglucinol derivatives, known for their anti-inflammatory activity—crucial in wound healing [5-6]. Sequential supercritical fluid extraction (S^3FE) enabled the selective isolation of these bioactive constituents. The 100% CO_2 extract, rich in largely uncharacterized phloroglucinols, further highlights the species' chemical richness. Ongoing bioassays aim to establish direct correlations between compound classes and observed biological effects.

Acknowledgments

Authors thanks the Project ESTIM-ICOA (CPER / FEDER-FSE+ 2021-2027-00022860), for their financial support

PSE MEETING 2025

PHYTOCHEMICALS AS DRUGS, FOODS AND BIOCOMMUNICATORS

Funchal • Madeira Island • PORTUGAL | 22-24 October 2025

ALTINBAS UNIVERSITY

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HERA

Phytochemical screening and investigation of physiological effects induced by *Helichrysum arenarium* subsp. *erzincanicum* in endothelial, keratinocyte, and fibroblast cell lines

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Introduction

Helichrysum Mill. is a member of the Asteraceae family and comprises approximately 656 taxa worldwide. While 34 taxa grow naturally in Türkiye, 17 taxa are endemic. In ethnobotanical research conducted in Türkiye, *Helichrysum* species are used for wound healing, hypercholesterolemia, diabetes, kidney stones, hepatitis, cough, and jaundice [1,2]. *Helichrysum* species are known to contain a diverse group of phenolic and terpenic compounds [3]. Our literature review has revealed that in vitro bioactivity studies demonstrating the anti-aging effects of the endemic species *Helichrysum arenarium* subsp. *erzincanicum* P. H. Davis & Kupicha, the subject of this research, have not been conducted before. This research topic was chosen to explore the potential of *H. arenarium* subsp. *erzincanicum* in the preparation of various formulations in the cosmetics industry, similar to *H. italicum*, a species used by major cosmetics companies. The aim of this study was to conduct chemical analyses of plant extracts prepared using various methods, determine their biological effects on L929 mouse fibroblasts, HuVeC human endothelial cells, and HaCat human keratinocyte cells, and determine IC_{50} values. Because the primary objective of the study was to develop a cosmetic product, it was planned to use HaCat human keratinocyte cells derived from human skin cells, and HuVeC human endothelial cells due to the potential penetration of the substance into the dermis. Toxicity is measured by assessing active substance exposure to L929 fibroblast cells. Endothelial cells are the cells that line the inner surface of blood vessels and play an important role in the vascular system. Keratinocyte cells play an important role in skin renewal, signs of aging and wrinkles.

Material and method

To determine the biological effects of *Helichrysum arenarium* subsp. *erzincanicum* was collected from different localities, after identification, stored at the Herbarium of Altınbaş University, Faculty of Pharmacy (HERA1504). It was extracted with ethanol, at the same time selective extractions were performed with NADES and sequential SFE. Extracts have been analyzed by UHPLC-DAD-ELSD. To perform biological activity experiments involving the addition of active ingredients, sufficient numbers of cell types were seeded into 96-well plates at 0.01 x 106 cells per well. Once the cells reached at least 95% density, the active ingredients were added to form experimental groups. All microscopic examinations and analyses were performed using the Carl-Zeiss/Axio observer 3 in our laboratory. Experimental groups were planned to have at least 3 replications.

Results and discussion

The extract of *H. arenarium* subsp. *erzincanicum* had IC_{50} values above 0.5 mg/mL, suggesting lower cytotoxicity. Notably, it exhibited significant proliferative effects at low concentrations on all cell lines, particularly in HaCaT cells (Prolif max at 0.0125 mg/mL, $P < 0.001$). These results indicate that this taxa may possess a promising proliferative profile with lower cytotoxic potential. To have a better understanding of these activities, a phytochemical study of these extracts was undertaken. Analysis by HPLTC and UHPLC-DAD-ELSD has shown high diversity of phenolic compounds particularly phloroglucinol derivatives, known for their anti-inflammatory activity—crucial in wound healing [5-6]. Ultrasounds assisted NADES extraction and Sequential supercritical fluid extraction (SFE) enabled the selective isolation of these bioactive constituents. The non-polar NADES extract and the 100% CO_2 extract, rich in largely uncharacterized phloroglucinols, further highlights the species' chemical richness. UHPLC-HRMS/MS analysis is in progress to refine compound identification and ongoing bioassays aim to establish direct correlations between compound classes and observed biological effects. As a result of this study, it was concluded that SFE extraction is a more practical and rapid method for the analysis of nonpolar substances in plant extracts and biological activity studies, but non-polar nades extraction can be used for large-scale extraction and product formulation.

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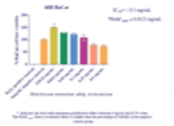
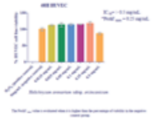
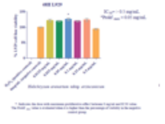
Biological activities

H. arenarium subsp. *erzincanicum*







The higher sensitivity observed in HaCaT and HUVEC cells compared to L929 suggests that dermal and endothelial cell responses should be considered in dose optimization for cosmetic or therapeutic formulations. Importantly, concentrations ≤0.1 µg/mL preserved ≥90% cell viability in all cell types, indicating a promising safety window. These findings support the biocompatibility and functional potential of *H. arenarium* subsp. *erzincanicum* for skin-targeted antioxidant and anti-aging applications.

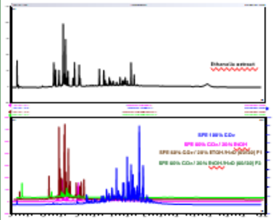
Phytochemical analysis

SFE extraction parameters

- Nexera UC system from Shimadzu configuration SFE-FRC
- Mobile phase : (A) CO_2 (B) EtOH or EtOH/H_2O
- Sequential selective extraction : 40 °C, 150 bar , 3mL/min
  - Step 1 : 100% of CO_2
  - Step 2 : 20% of EtOH
  - Step 3 : 20% EtOH/H_2O (80/20)

NADES extraction

- Polar Nades extraction: Betaine, lactic acid, water (1:2:1).
- Non-polar Nades extraction: Menthol, lactic acid (2:1).



UHPLC-HRMS/MS analysis parameters

-Thermo vanquish UHPLC system with a Bruker Ascend Tribrid

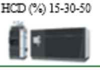
-Phenomenex Luna Omega C18 100 × 2.1 mm, 1.6 µm

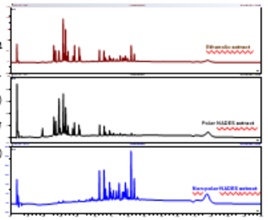
-Mobile phase : (A) H_2O + 0.1% FA (B) ACN/IPA (50/50) + 0.1% FA

-Gradient : 5% to 97% B in 15 min and 97 % B during 10 min

-HRMS: ESI+ mode on range 100-1600 m/z

-MS/MS: HCD (%) 15-30-50





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LE STUDIUM

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