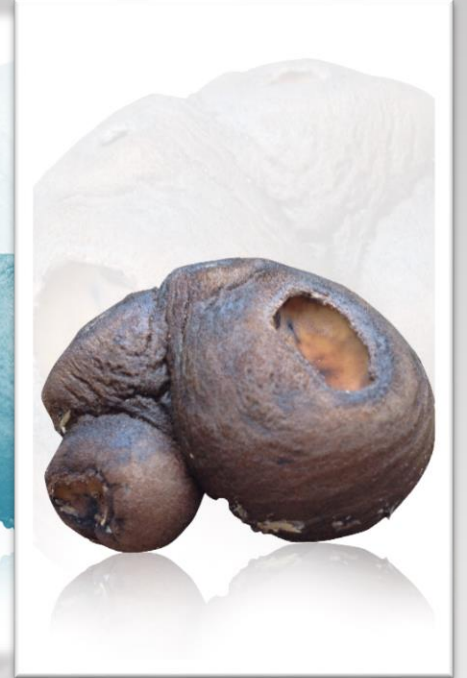
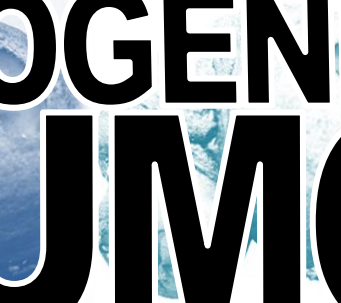


ISOLATION of
the **CYTOTOXIC**
ANGIOGENIC and
ANTITUMOR
COMPOUNDS
from **TRICHALEURINA**
CELEBICA



Project Description

- **The previous findings on this *T. celebica* commence more assays on the biological properties of this fungal species with the hope to find a suitable application as food supplement, on food fortification and/ or drug development.**
- **In this particular project, the isolation of the cytotoxic components and the determination of its angiogenic and antitumor properties were considered prelude to finding a novel drug.**

Objectives

- **Isolate the pure compounds from *T. celebica*, exhibiting cytotoxic property,**
- **Determine the anti-angiogenic property of the isolates,**
- **Determine the antitumor potential,**
- **Determine the bioactive metabolites which are characterized as cytotoxic, angiogenic and anti-tumor.**

FLOWCHART of the PROCEDURES

01 **Collection and Mycelial Propagation
of the Mushroom Sample**

**Mass production of secondary
metabolites**

02

03 **Solvent Fractionation**

Cytotoxicity Assay

04

05 **Gravity Column Chromatography**

**Chicken Chorioallantoic
Membrane Assay**

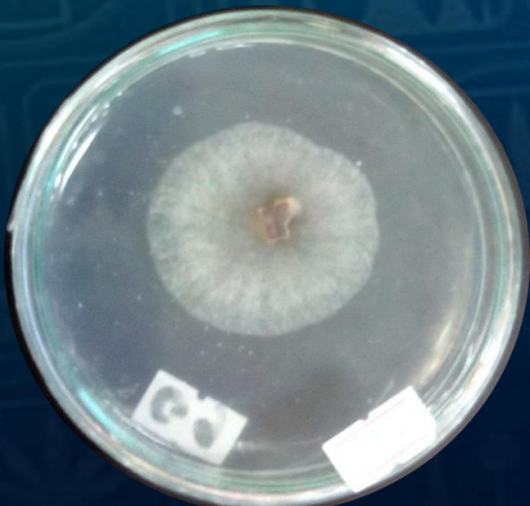
06

07 **Potato Disc Antitumor Assay**

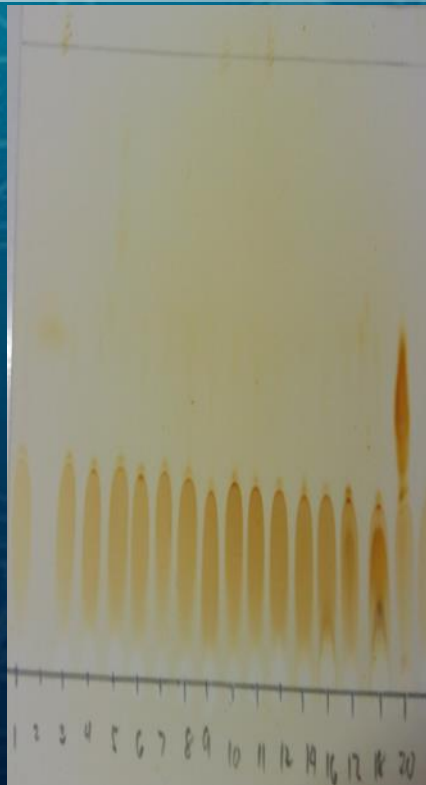
Snapshots of the procedures



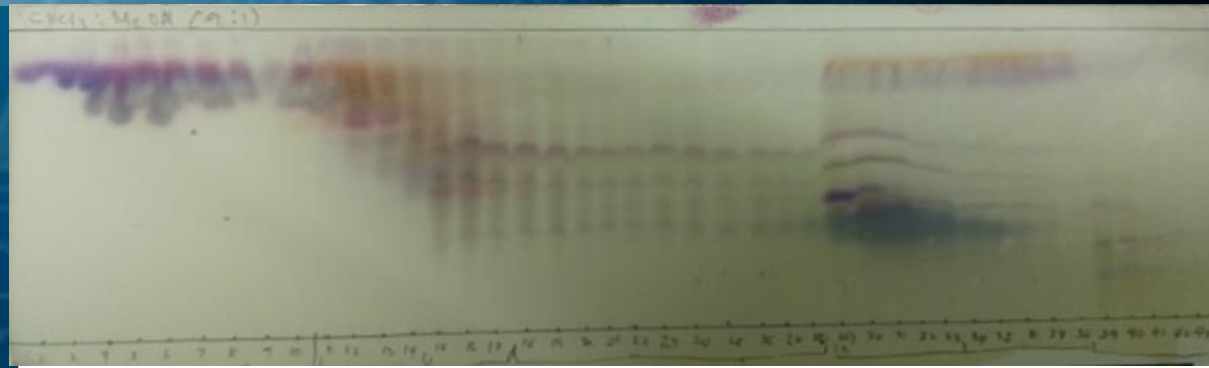
Collected *T. celebica* from the wild



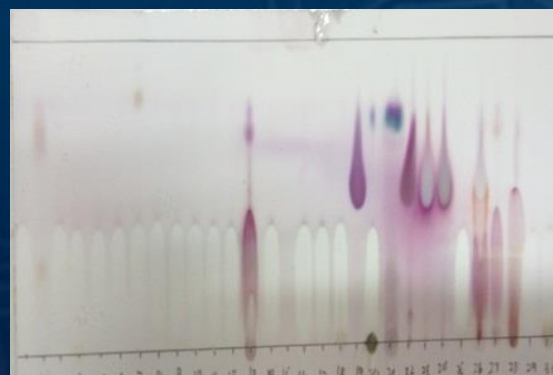
Tissue culture of *T. celebica*



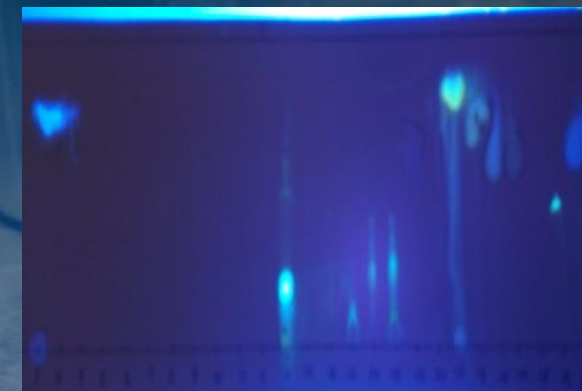
IDENTIFICATION OF FATTY ACIDS IN SEVERAL ISOLATES USING IODINE VAPORS.



Chromatogram of the solvent fractions in the gravity column chromatography



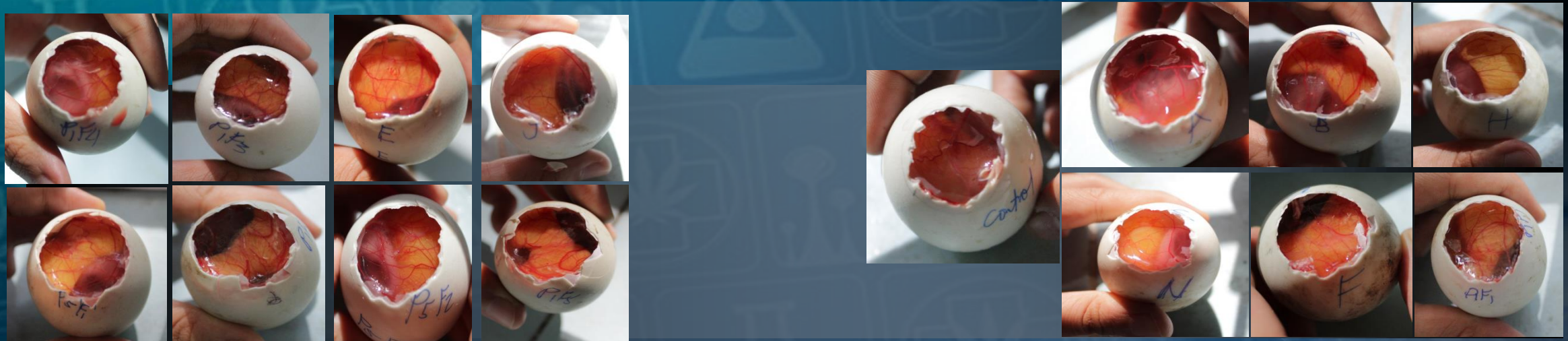
A CHROMATOGRAM IN VANILLIN – SULFURIC ACID AS VISUALISING AGENT SHOWING TRITERPENES IN PINK SPOTS, PHENOLS IN BROWN COLORATION, VIOLET FOR ESSENTIAL OILS



FLOURESCENT METABOLITES IN METHANOLIC-POTASSIUM HYDROXIDE. BLUE FLOURESCENCE INDICATING COUMARINS AND ORANGE WITH ATHRAQUINONES

Isolation of the Cytotoxic, Angiogenic and Antitumor Compounds from *Trichaleurina celebica*

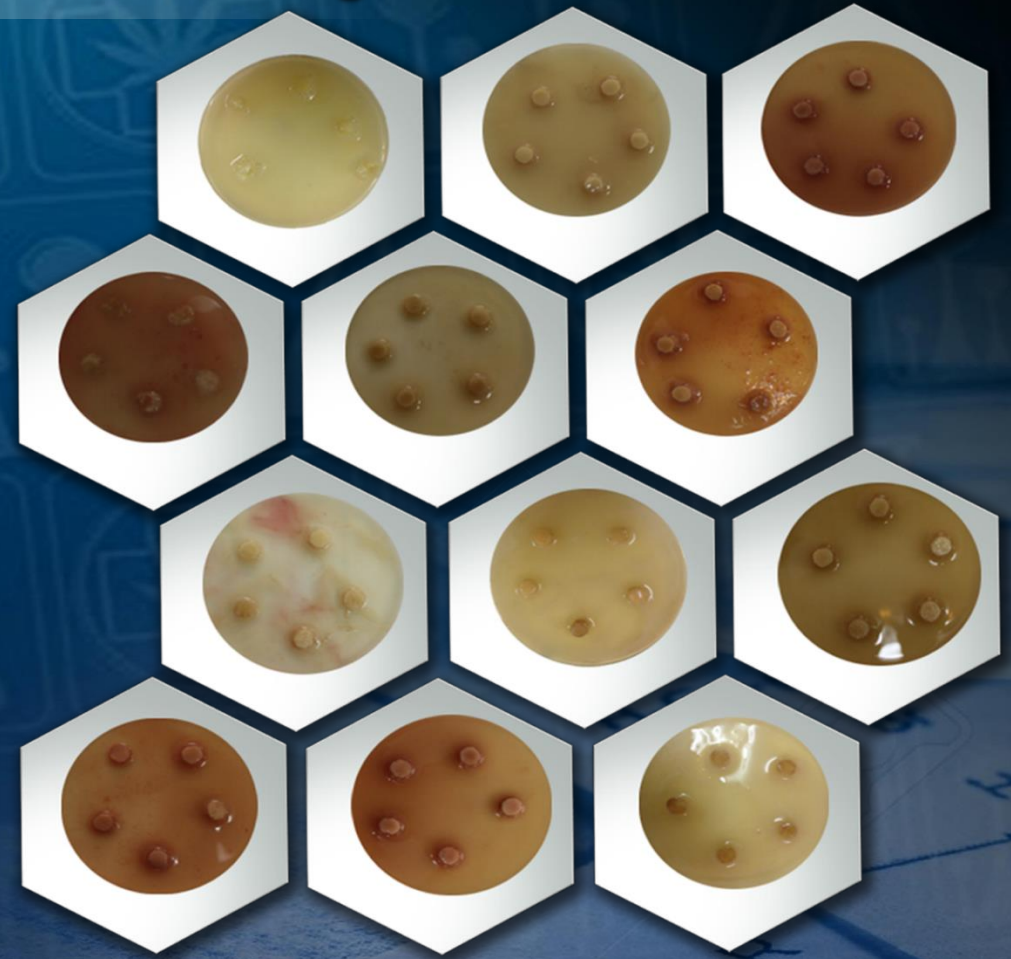
Chicken Chorioallantoic Membrane Assay



$$\% \text{ Vascularity} = \frac{\text{No. of branched points (treated)} - \text{No. of branched points (control)}}{\text{No. of branched points (control)}} \times 100$$

Isolation of the Cytotoxic, Angiogenic and Antitumor Compounds from *Trichaleurina celebica*

Potato Disc Antitumor Assay



$$\% \textit{inhibition} = 100 - \frac{\textit{Ave No. of tumors in sample}}{\textit{Ave No. of tumors in control}} \times 100$$

Isolation of the Cytotoxic, Angiogenic and Antitumor Compounds from *Trichaleurina celebica*

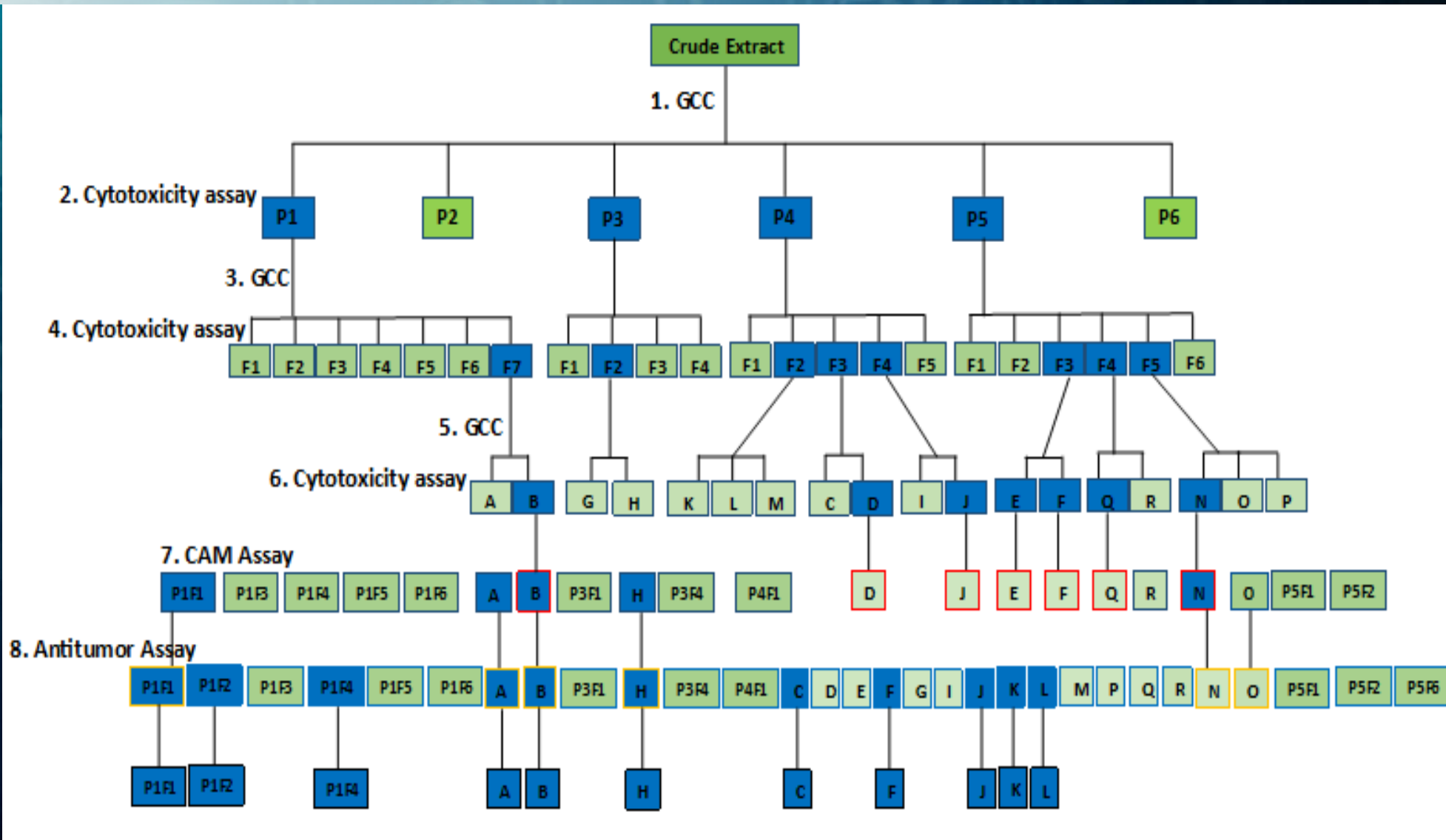
Result of the Study

No.	Isolates	BIOACTIVITY	BIOACTIVE METABOLITE
1	A	Anti-angiogenic, antitumor	Fatty acid
2	B	Cytotoxic, anti-angiogenic, antitumor	Coumarin
3	C	Antitumor	Fatty acid
4	D	Cytotoxic, pro-angiogenic	Fatty acid
5	E	Cytotoxic, pro-angiogenic	Fatty acid
6	F	Cytotoxic, antitumor	Fatty acid
7	G	None	Fatty acid
8	H	Anti-angiogenic, antitumor	Fatty acid
9	I	None	Fatty acid
10	J	Cytotoxic, pro-angiogenic, antitumor	Fatty acid
11	K	Antitumor	Fatty acid
12	L	Antitumor	Fatty acid
13	M	None	Coumarin
14	N	Cytotoxic, anti-angiogenic pro-angiogenic	Fatty acid
15	O	Pro-angiogenic	Fatty acid

No.	Isolates	BIOACTIVITY	BIOACTIVE METABOLITE
16	P	None	Fatty acid
17	Q	Pro-angiogenic	Fatty acid
18	R	Pro-angiogenic	Phenols
19	P ₁ F ₁	Anti-angiogenic, antitumor	Essential Oil
20	P ₁ F ₂	Antitumor	Fatty acid
21	P ₁ F ₃	Pro-angiogenic	Anthrone
22	P ₁ F ₄	Pro-angiogenic	Anthraquinone
23	P ₁ F ₅	Pro-angiogenic	Anthraquinone
24	P ₁ F ₆	Pro-angiogenic	Essential Oil
25	P ₃ F ₁	Pro-angiogenic	Fatty acid
26	P ₃ F ₁	Pro-angiogenic	Anthrone
27	P ₄ F ₁	Pro-angiogenic	Triterpenes
28	P ₅ F ₁	Pro-angiogenic	Coumarin
29	P ₅ F ₂	Pro-angiogenic	Fatty acid
30	P ₅ F ₆	none	Not identified

Isolation of the Cytotoxic, Angiogenic and Antitumor Compounds from *Trichaleurina celebica*

Summary of the isolation process



Isolation of the Cytotoxic, Angiogenic and Antitumor Compounds from *Trichaleurina celebica*

Conclusion and Recommendation

Several bioactive compounds isolated and were identified from *T. celebica* have potential as lead in the exploration of pharmaceutical products as anti-tumor, anti – angiogenic and pro – angiogenic agents.

It is therefore recommended that the isolates from *T. celebica* will be screened for its anti – cancer potential using human cancer cell lines and in the treatment of cardiovascular diseases due to its high pro-angiogenic potential in CAM assay.