

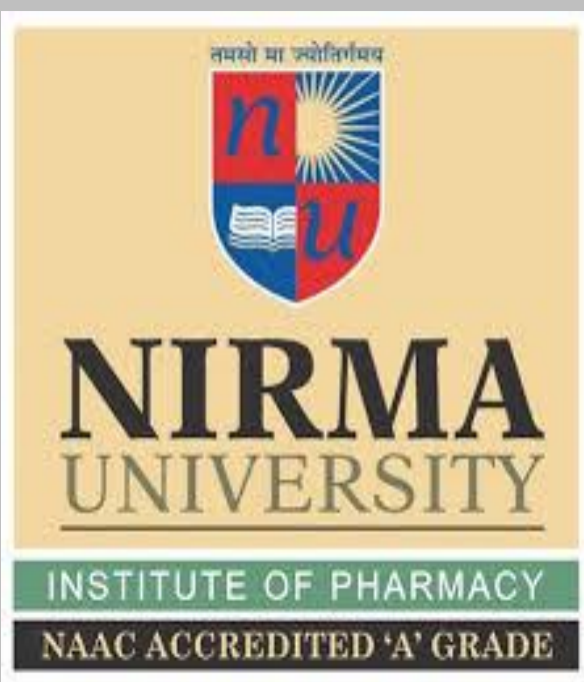
To screen the effects of Insulin Plant extract on rat liver CYP isoenzyme system

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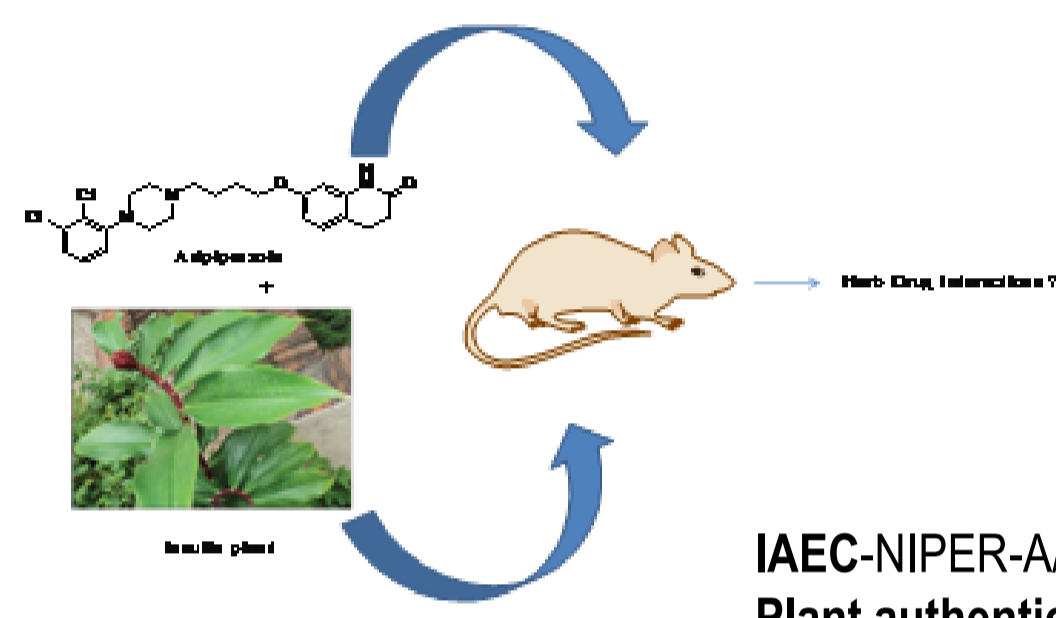
Objectives:

❖ Aripiprazole (Abilify) belongs to the atypical antipsychotic category, frequently prescribed for the cure of Psychosis or Schizophrenia.

❖ Cytochrome P2D6 (CYP2D6) is a prominent enzyme that plays key role in the metabolism of Aripiprazole and formation of an active metabolite Dehydroaripiprazole takes place.

❖ Patients under the treatment with this potent moiety have been reported to have high blood glucose level as a side effect. Additionally, literatures say that the leaves of Insulin plant is usually administered by people 2-3 times a day to control the sugar level.

❖ The present work was done to investigate the effect of Insulin plant extract (IPE) on CYP2D6, with the co-administration of Aripiprazole (to examine the changes in metabolite of Aripiprazole).



IAEC-NIPER-A/IAEC/2017/031/R
Plant authentication- NIPER-A/NP/0816/001

Material & Method:

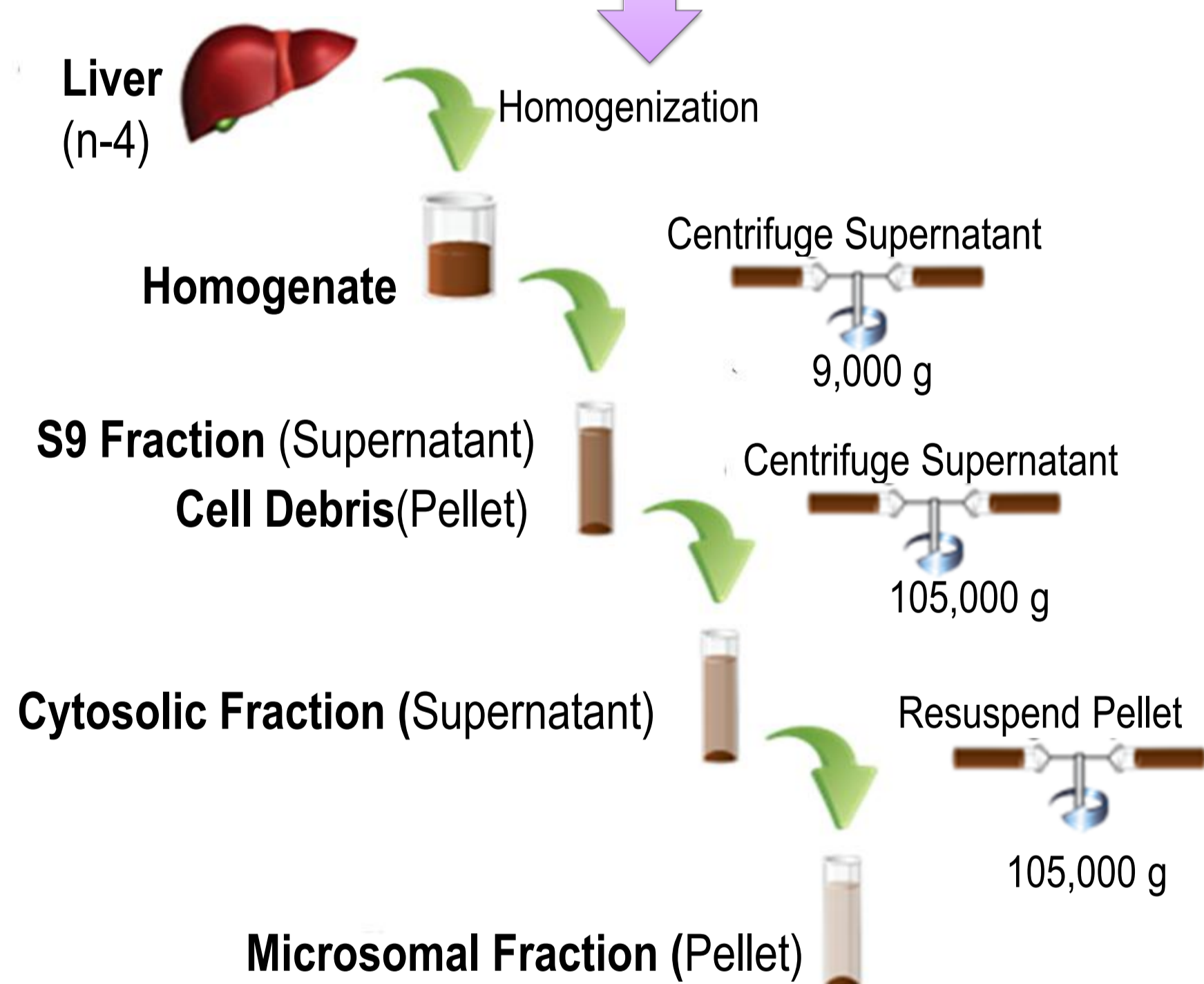
Preparation of IPE

Successive extraction method, Methanolic extract was found enriched with Quercetin.

Chemo profiling of IPE

HPLC RP_C-18; ACN: MQ (3pH) & UV

Isolation of Microsome



CYP2D6 assay

• Aripiprazole was used as a probe substrate and formation of Dehydroaripiprazole was used as a measure of CYP2D6 activity.

• In sample tubes, reaction mixture = phosphate buffer + test samples (extract 20-1000µg/ml) + Aripiprazole + RLM were added.

• The reaction was initiated with the addition of NADPH and incubated → centrifuged.

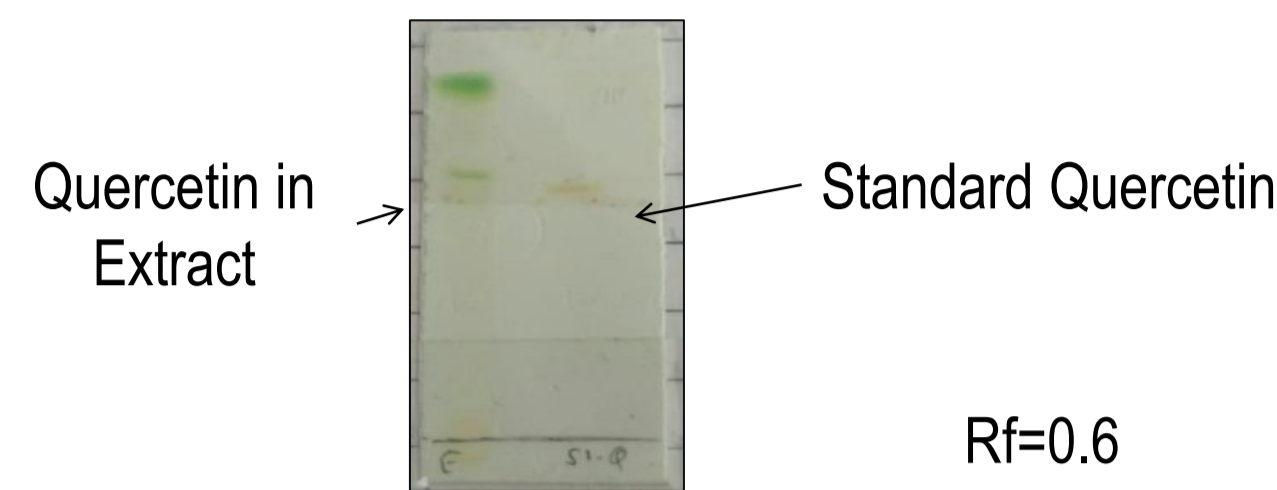
• The residue was used for the high performance liquid chromatography analysis.

• The assay was performed in triplicate.

• IC50 value was calculated.

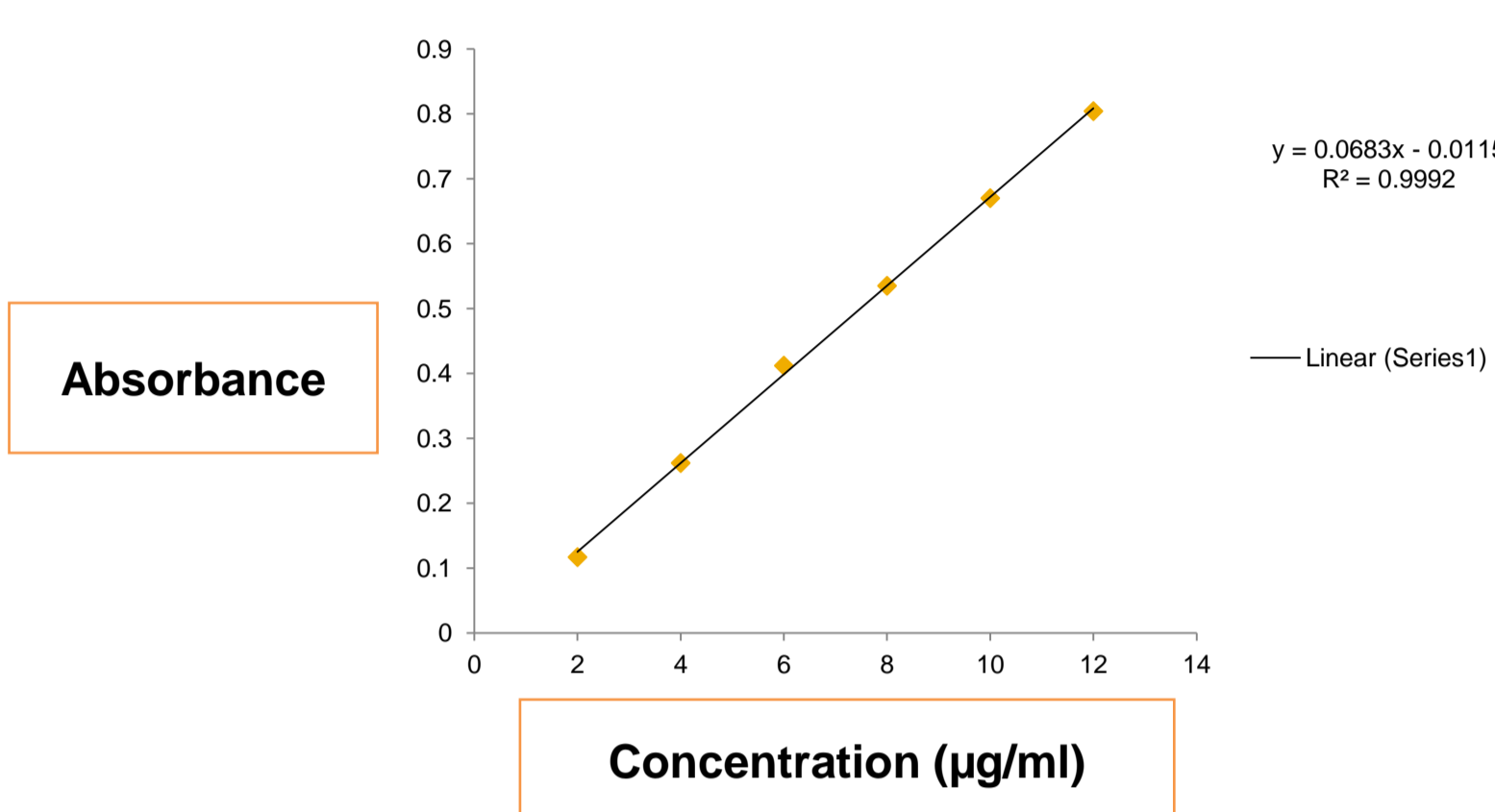
Results:

1. TLC of IPE (Methanolic):



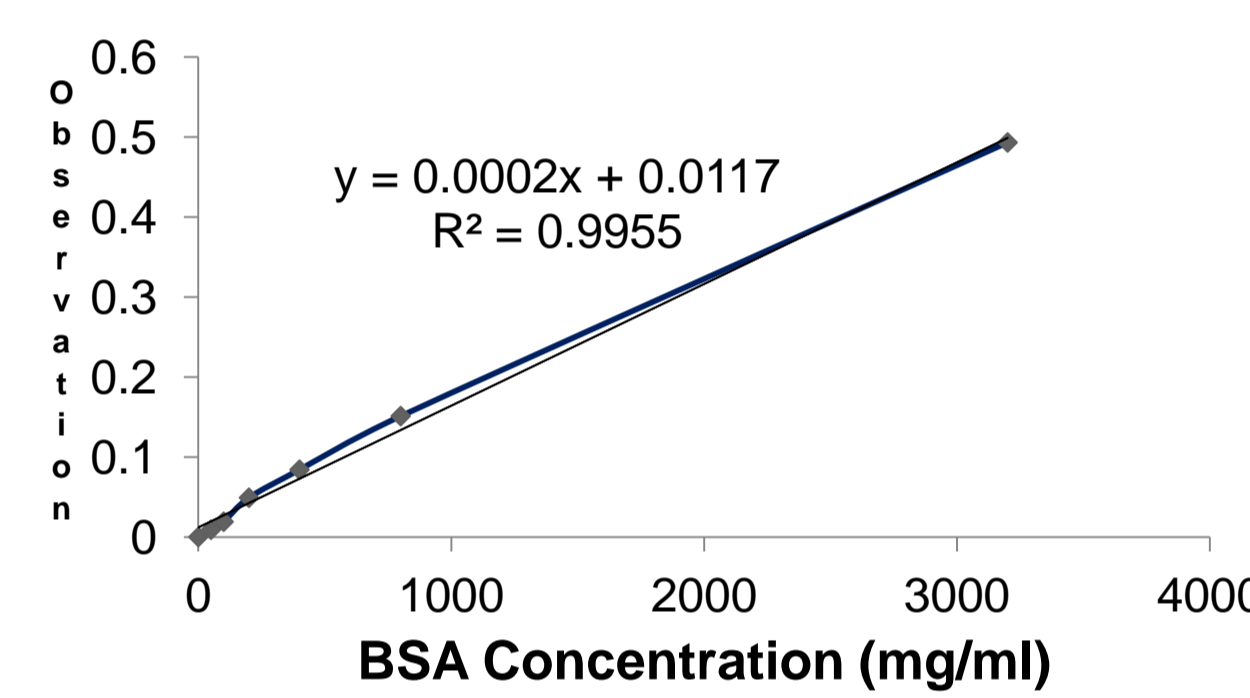
TLC of Methanolic extract with standard Quercetin

2. Quantification of Quercetin by UV spectroscopy:

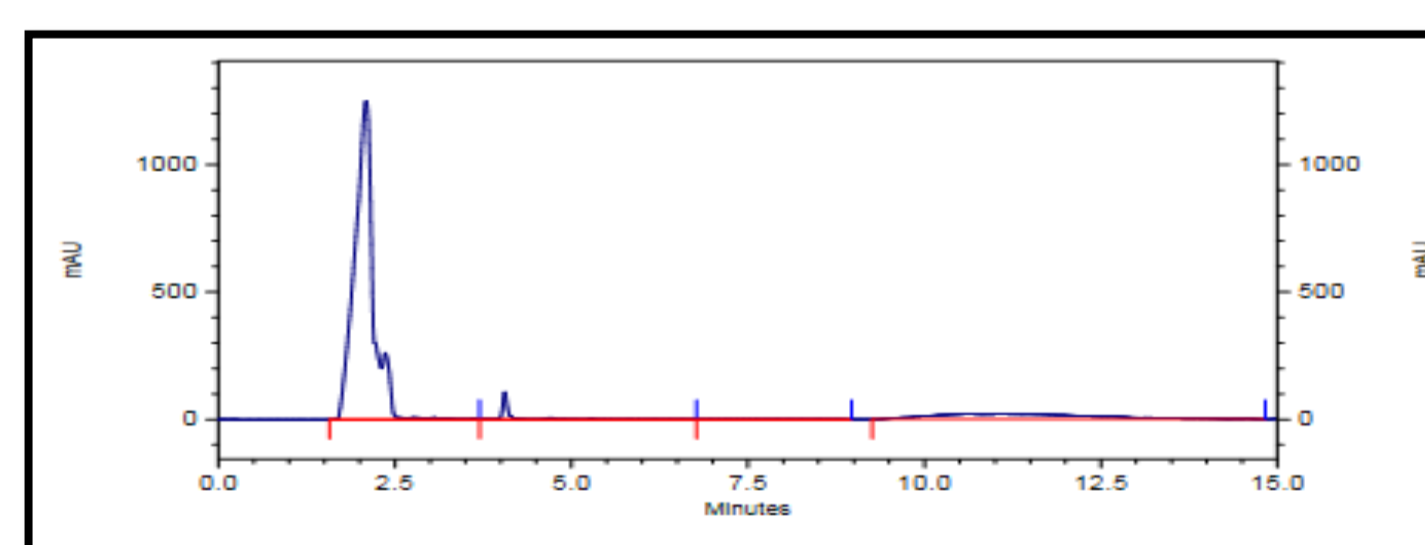


Quercetin in extract was found to be in 3.9604µg/ml.

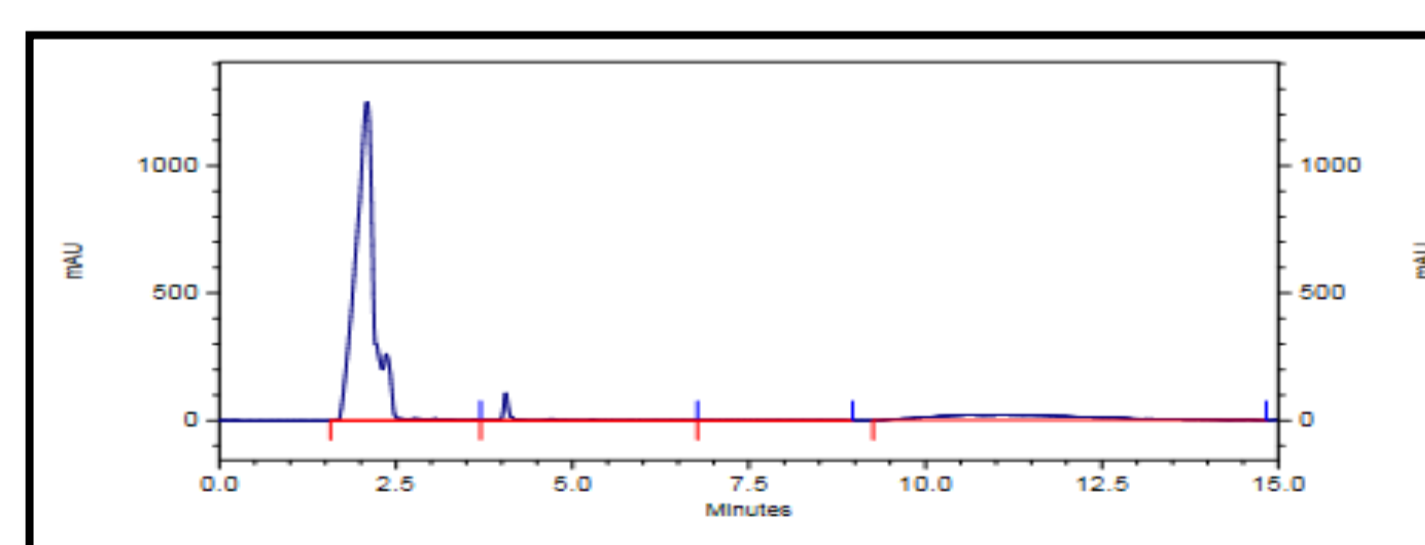
3. Microsome Protein content:



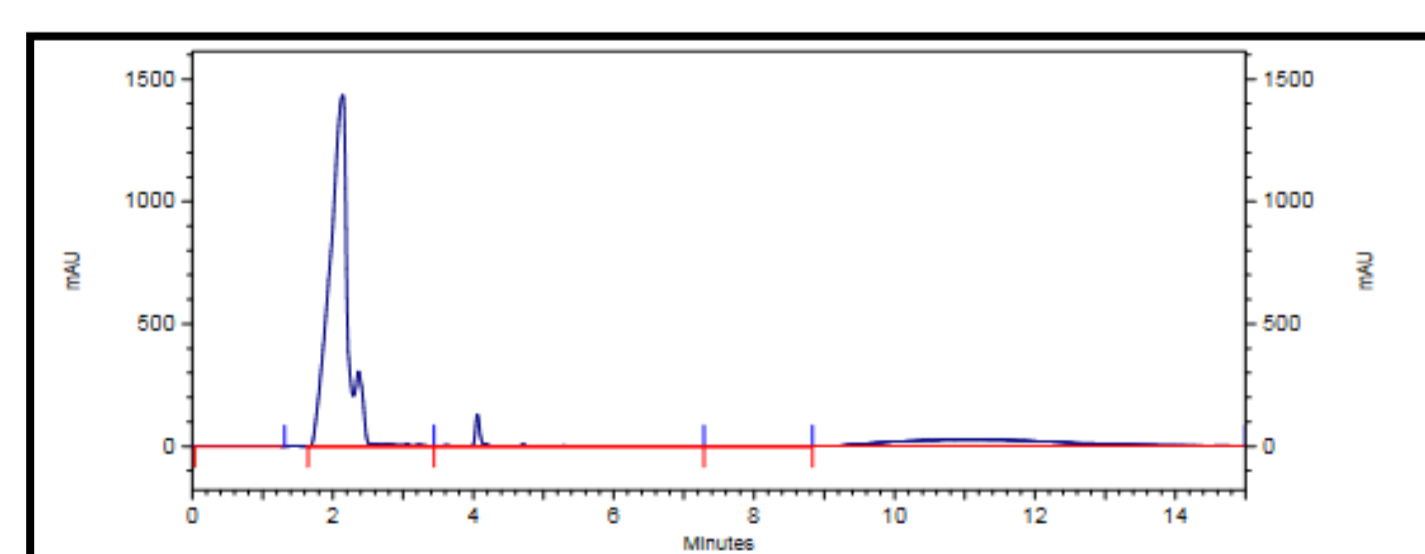
4. HPLC Chromatogram:



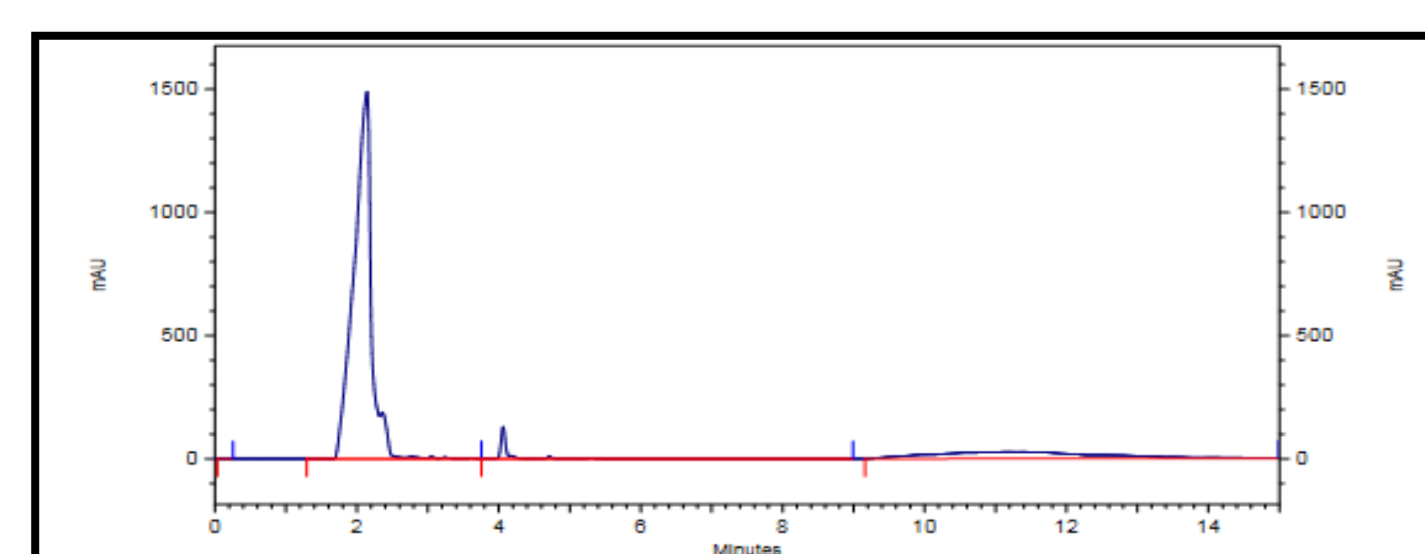
Mixture of Aripiprazole + rat liver microsome



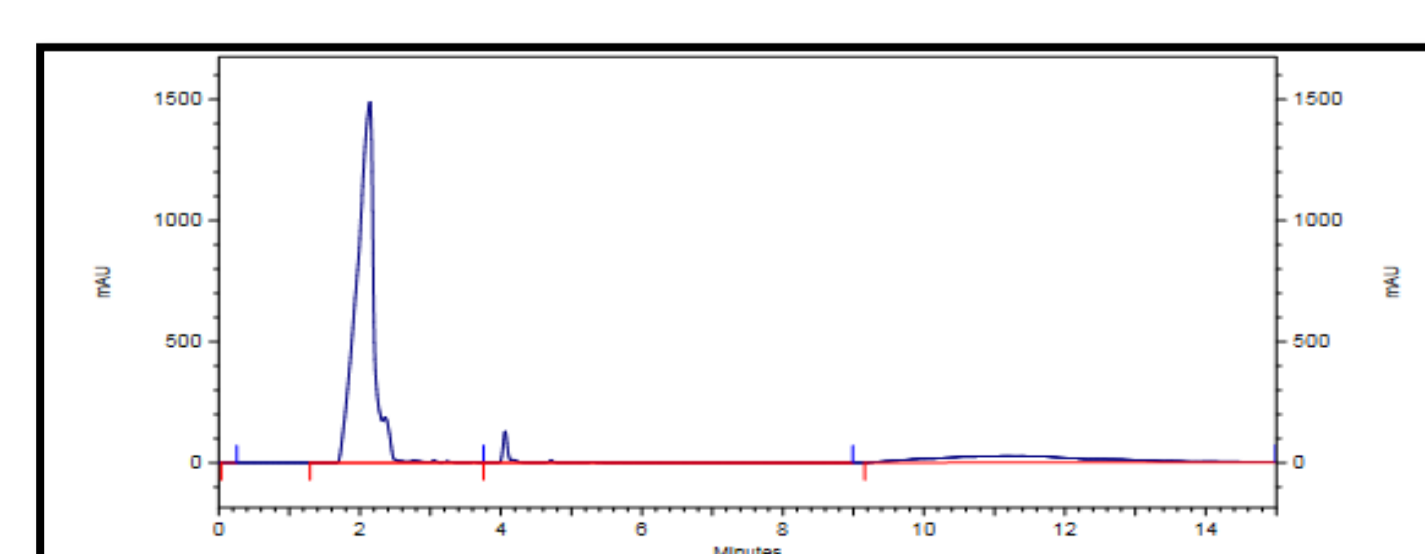
Aripiprazole + RLM + IPE 200µg/ml



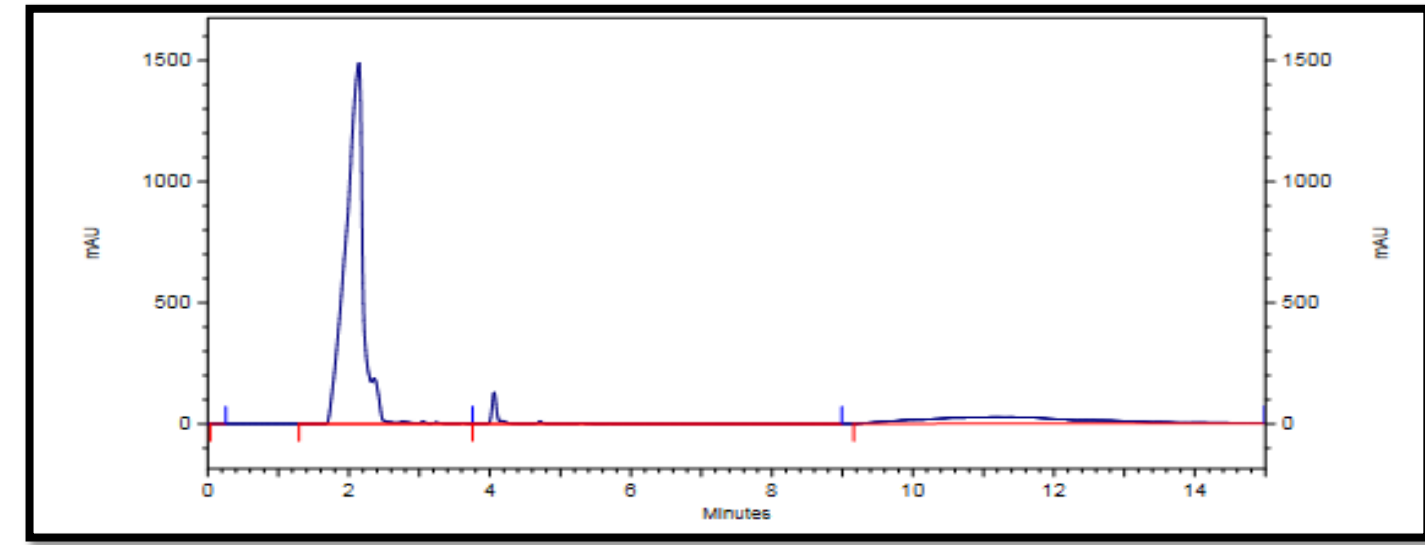
Aripiprazole + RLM + IPE 400µg/ml



Aripiprazole + RLM + IPE 600µg/ml



Aripiprazole + RLM + IPE 800µg/ml



Aripiprazole + RLM + IPE 1000µg/ml

Discussion:

5. Log inhibitory concentration: Data are represented as mean ± standard (n=3).

Log Dose (µg/ml)	Inhibition
4.301	0.107 0±0.0065
4.602	0.221 0±0.0076
4.778	0.268 8±0.0096
4.903	0.551 5±0.075
5	0.221 1± 0.0083

■ IPE was prepared by the successive extraction method.

■ Preliminary phytochemical screening was performed and Methanolic extract was found to be enriched with the analytical marker Quercetin, & Rf value was found to be 0.6.

■ Qualitative and quantitative determination was done using HPLC and UV where Quercetin was found to be 3.96 µg/ml.

■ HPLC study showed that, no significant changes were observed in the retention time and peak shift of Aripiprazole when incubated with the IPE and RLM.

■ This observation may conclude that extract is not inhibiting CYP2D6 which is responsible for metabolite of Aripiprazole.

Conclusion:

The IC₅₀ value (Graph Pad Prism 5) of IPE has been found to be (4.49 µg/ml) beyond 1000µg/ml and hence it has been concluded that extract is safe from inhibitory reaction point of view. As the dose of herbal extract which is used by Ayurvedic physician are much lower than that used here so that chances of occurring inhibitory interaction is very rare. But in order to confirm this, clinical study is required which can also focus on enzyme induction and interplay as well so that complete HDI profile can be studied.

Acknowledgement:

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