Extraction of Parthenin from Parthenium Hysterophorus

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Abstract

*Parthenium Hysterophorus plant contains toxins called sesquiterpene lactones. The major component of toxin is **Parthenin** & other phenolic acids such as Caffeic acid, Vanillic acid, Anisic acid, Panisic acid, Chlorogenic acid & parahydroxy benzoic acid are found to be lethal to human beings and animals.The proposed work intend to obtain Parthenin from Parthenium Hysterophorus by solvent extraction and Soxhlet Extraction methods, using different solvents at different flow rates and to calculate Mass Transfer Coefficients (MTC) for each of the system, thereby exploring the possibility for the extraction of Parthenin from Parthenium Hysterophorus. The comparison of two applied methods shows that Soxhlet extraction is an efficient method by which solute of limited solubility can be extracted with higher efficiencies.Seemingly, the extraction of Parthenin from Parthenium Hysterophorus is a very complicated process. Our experiments indicate that the extracted amount of Parthenin is affected by several parameters like solvent properties, solvent volume, extraction time, temperature, particle size and the microstructure of the sample.*

Keyword: Parthenium hysterophorus, soxhlet extraction, solvent extraction

Introduction

Parthenium Hysterophorus is one of the ten worst weeds in the world, popularly known as Congress weed, Gajar Ghas, Carrot weed, Star weed, Fewerfew, White top, Chatak Chandani, Bitterwood, and Ramphool etc., believed to have entered India accidentally in mid fifties. **[1]** The Chemical Analysis has indicated that all the parts of Parthenium Hysterophorus plant contain toxins called sesquiterpene lactones. The major component of toxin is **Parthenin** & other phenolic acids such as Caffeic acid, Vanillic acid, Anisic acid, Panisic acid, Chlorogenic acid & parahydroxy benzoic acid are found to be lethal to human beings and animals**. [2, 3]** Although a Parthenium Hysterophorus is considered a toxic plant, sesquiterpene lactones extracted from the plants posses certain medicinal, germicidal and pesticidal properties. Figure 1 shows the Parthenium Hysterophorus Plant. It has been reported that the Parthenin – a sesquiterpene lactone present in Parthenium Hysterophorus root, stem, leaf & inflorescence extracts, inhibited seed germination and seedling growth (root and shoot length) of various crops like linseed, chickpea etc. **[4]**. Also the Pesticidal properties of Parthenin & sesquiterpene lactones were tested and found effective as an insecticide **[5]**.

Figure 1 Parthenium Hysterophorus Plant

It has been found that Parthenin and its derivatives are practically insoluble in water but are soluble in alcohol, chloroform, ether, and acetone and ethyl acetate **[6]**. Also it is found that boiling point of all the constituents of Parthenium Hysterophorus extract is in between 165 ºC – 220 ºC, while that of Methanol is 64.7 ºC and they do not form an azeotropic mixture and hence can be separated by simple distillation techniques **[7]**. It was proposed to use Chemical Engineering techniques (Soxhlet Extraction and Packed Bed Extraction) for separation of Sesquiterpene Lactones from Parthenium Hysterophorus which could be effectively used as Pesticides or Insecticides **[8, 9]** The proposed work intend to obtain Parthenin from Parthenium Hysterophorus by solvent extraction and Soxhlet Extraction methods, using different solvents at different flowrates and to calculate MTC for each of the system, thereby exploring the possibility for the extraction of Parthenin from Parthenium Hysterophorus.

MATERIAL AND METHOD

Parthenium Plant

Figure 2 Parthenium Aerial Parts

Common Name: Carrot Grass, Congress grass, Wild carrot weed

Hindi: Gajar Gavat, Chatak Chandani

Botanical name: Parthenium Hysterophorus

Family: *Asteraceae* (Sunflower family)

Carrot Grass is native to the subtropics of North and South America. It is a fast-maturing annual perennial with a deep tap root and an erect stem that becomes woody with age. It may eventually reach a height of 2 m. Its leaves are pale green, branched and covered with soft fine hairs. The small white flowers (4 mm across) have five distinct corners and grow on the stem tips. Each flower produces 4-5 black wedge shaped seeds that are 2 mm long with thin white scales. It is considered a highly invasive weed. Its large and persistent soil seed bank, fast germination rate and ability to undergo dormancy make it well adapted to semi-arid environments. It also releases chemicals that inhibit the germination and growth of pasture grasses and other plants **[11]**.

Soxhlet Extraction

Extraction was accomplished in Soxhlet extraction unit (Figure 3) at 65ºC and normal atmospheric pressure. Soxhlet extraction was performed using a Soxhlet apparatus (kimax) and a heating mantle/electric heater (electro thermal). 500 ml of solvent (methanol) was filled in a flask, which was placed on a heating mantle to change the solvent to gaseous phase.

The sample (small particles of Parthenium Hysterophorus) was filled in porus cellulose thimble and placed in mid portion – (Butt tube) of Soxhlet apparatus. The solvent was liquefied by cooling the solvent vapor by a water cooled condensor. The flow rate of cold water and temperature of heater was adjusted to liquefy the solvent at the rate of 20 drops per minute so that the liquefied solvent trickles in the extraction chamber containing the sample to perform the extraction.

Extraction cycle: the extraction chamber is designed so that when the solvent surrounding the thimble exceeds a certain level (200 ml) is overflows and trickles back down in to the boiling flask. This is considered as one cycle. It took approximately 20 min for each. The extraction process was carried over for 3.5 hours. After completion of the experiment the flask containing the extract was removed. Similar runs were taken using ethanol, acetic acid and chloroform as solvent.

Packed Bed Extraction

Extraction was performed using a packed bed column. 5 litres of methanol was filled in a tank. The sample (small particles of Parthenium Hysterophorus) was introduced in the column from the top to a height of 35 cm. The solvent was pumped from the tank to the top portion of column. The solvent (methanol) was trickled into the column through a glass sparger. The Flow rate of solvent was adjusted at 20 drops per min to perform as extraction. The extract was collected from the bottom. The extraction was carried for 3.5 hours. Figure 4 gives us the details of the experimental setup of packed bed column.

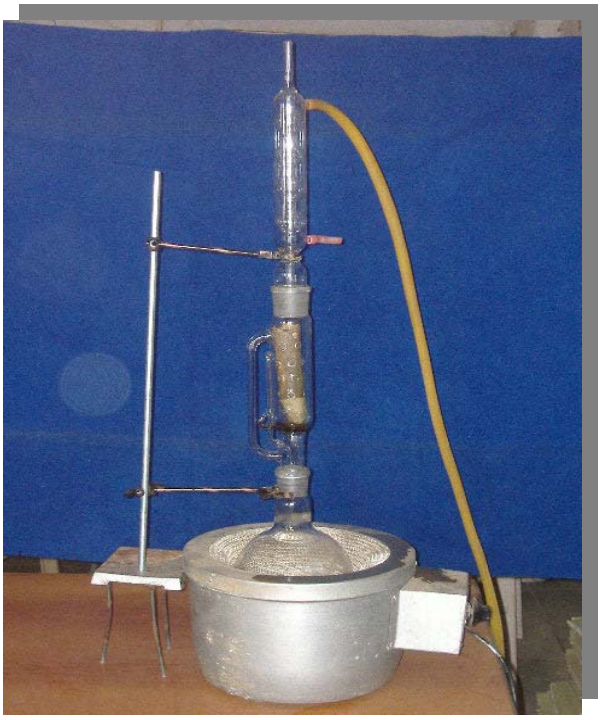

Figure 3 The Soxhlet Extractor Unit

Figure 4 The Packed Bed Reactor Unit

Experimental Methods

Figure 5 shows The Process Flow Diagram.

i) Parthenium from aerial parts (Figure 2) are manually separated from plant and sundried for the duration of 3 weeks.

ii) After drying, the aerial parts are shade dried for the period of seven days. The net weight of separated aerial part of the flowers is 150 grams.

iii) Parthenium aerial or flower parts are crushed in grinder after drying.

iv) Solvent Extraction: Apparatus use for extraction purpose is Soxhlet Extractor.

   a. First the raw material is divided into three parts of 50 gram each.

   b. The thimble is first covered with filter paper so that raw material cannot directly pass through the thimble and secondly to obtain filter solution.

   c. The Parthenium was extracted using the solvents methanol, ethanol and chloroform. The extraction time for each run was 12 hours.

v) After extraction process filtration is carried out.

vi) Extract is distilled to remove excess quantity of solvent and finally the sample of extract obtained is sent to High – performance liquid chromatography, HPLC, for detection of Parthenin content in the extract.[14]

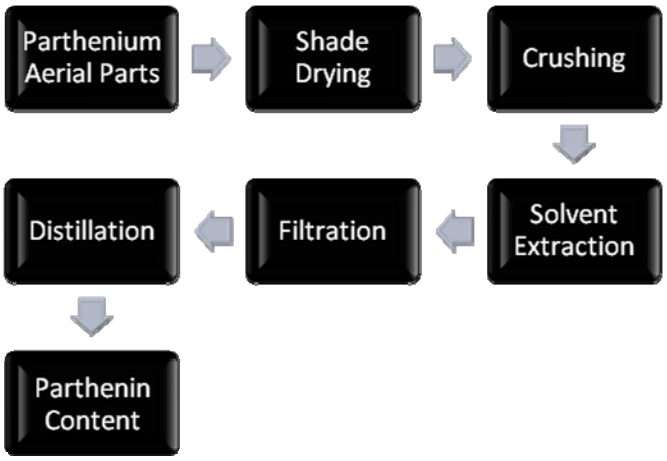

Figure 5 Process Flow Diagram

Diffusivity (D_{AB}) & Mass Transfer Coefficient (K_C) in Soxhlet Extractor and Packed Bed Extractor

The Wilke–Chang equation is a general-purpose equation for calculating diffusion coefficients. This equation is applicable to situations where the solute is dilute in the solvent. The Wilke – Chang equation is given and is used to calculate the diffusion coefficient of solute A in solvent B. **[12]**

Using packed bed, large amount of mass transfer area can be contained in a relatively small volume. The diffusion coefficient or diffusivity of liquids may be estimated by using Wilke-Chang correlation.

$$D_{AB} = \frac{(117.3\times10^{-18})\times(\varphi M_B)^{0.5}\times T}{\mu_B V_A^{0.6}} \qquad (1)$$

Where,

D_{AB} = Diffusivity of A in very dilute solution B, m^2/sec

M_B = molecular Weight of solvent, kg/k mole

φ = association factor for solvent

= 1.9 for methanol

μ_B = viscosity of solution , kg/m. sec

V_A = Solute molar volume at normal boiling point m^3/k mole

T = Absolute temperature, K

Procedure for calculating mass transfer coefficient: [12, 13]

1) Calculate the cross section area of the tower , $A = \frac{\pi}{4} \times D^2, m^2$

2) Calculate the velocity of the tower , $V = \frac{Q}{A}, \frac{m}{s}$

3) Calculate Schmidt number, $N_{Sc} = \frac{\mu}{\rho D_{AB}}$ (substitute D_{AB} from equation 1))

4) Calculate $\mathbf{J_d}$ i.e. mass transfer (dimension less group),

To calculate J_d we use

Wilson- Geankoplis correlation [13] –

$$N_{Re} = \frac{VD\rho}{\mu}$$

a) For N_{Re} = 0.0016 – 55 , and Nsc = 165 – 70600

$$J_d = \frac{1.09(Re)^{0.67}}{\epsilon}$$

OR

b) For N_{Re} = 55 – 1500 , Nsc = 165 – 10690

$$J_d = \frac{0.230(Re)^{-0.31}}{\epsilon}$$

Where $\epsilon = void\ fraction = \frac{void\ volume}{total\ volume}$ =0.3 to 0.5

5) Calculate Kc

$$J_d = \frac{Kc(Nsc)^{0.67}}{V}$$

Rearranging the above equation we get

$$Kc = \frac{J_d \times V}{(Nsc)^{0.67}}$$

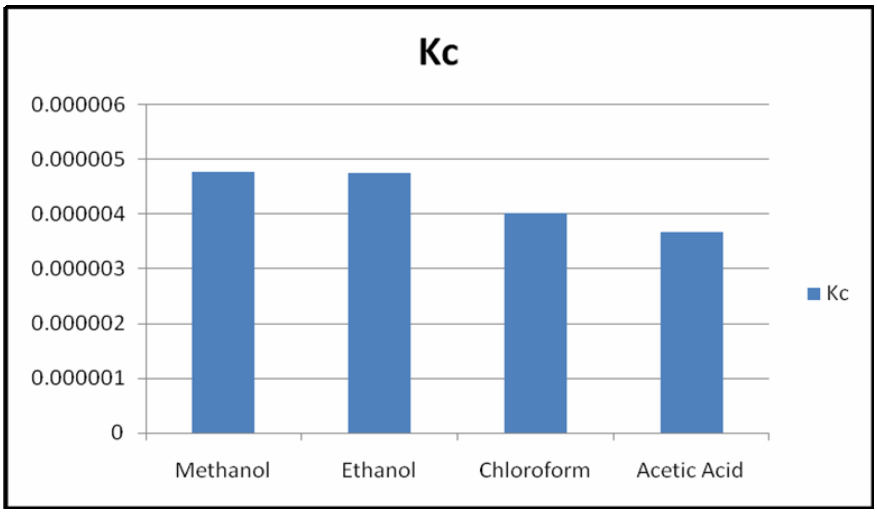

Figure 6: MTC for different solvents in Soxhlet Extractor

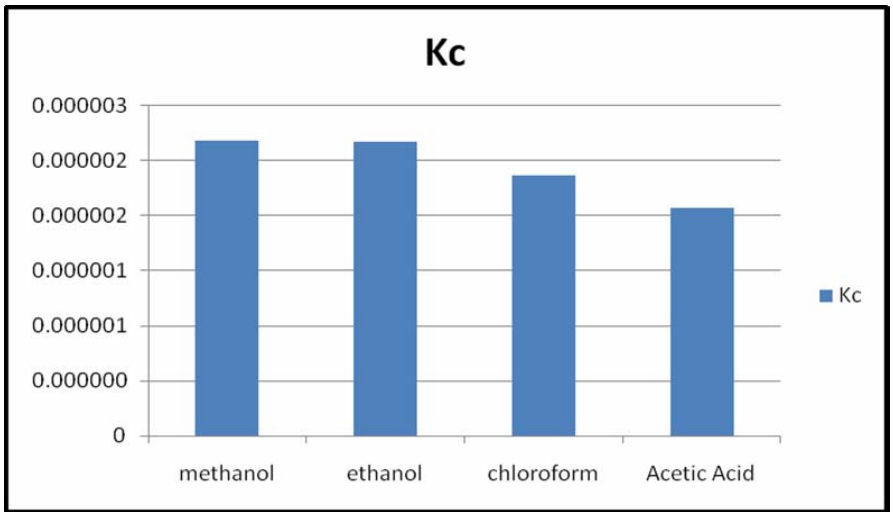

Figure 7: MTC for different solvents in Packed Bed Extractor at Flowrate of 10 ml/min

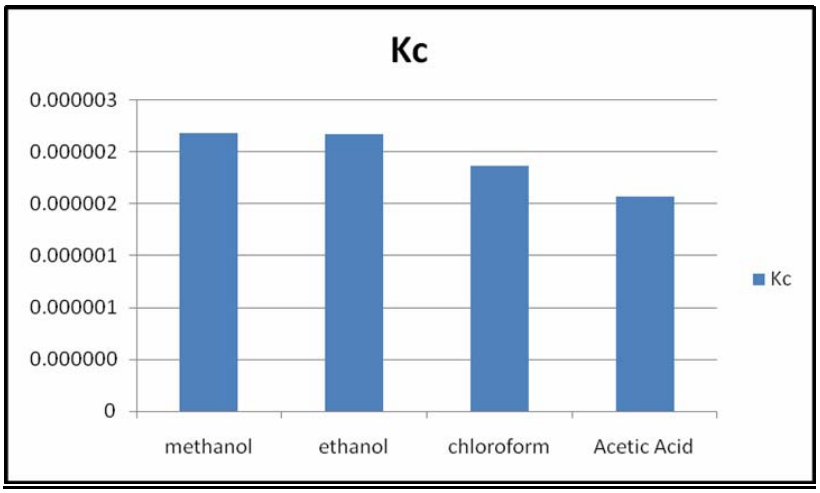

Figure 8: MTC for different solvents in Packed Bed Extractor at Flowrate of 20 ml/min

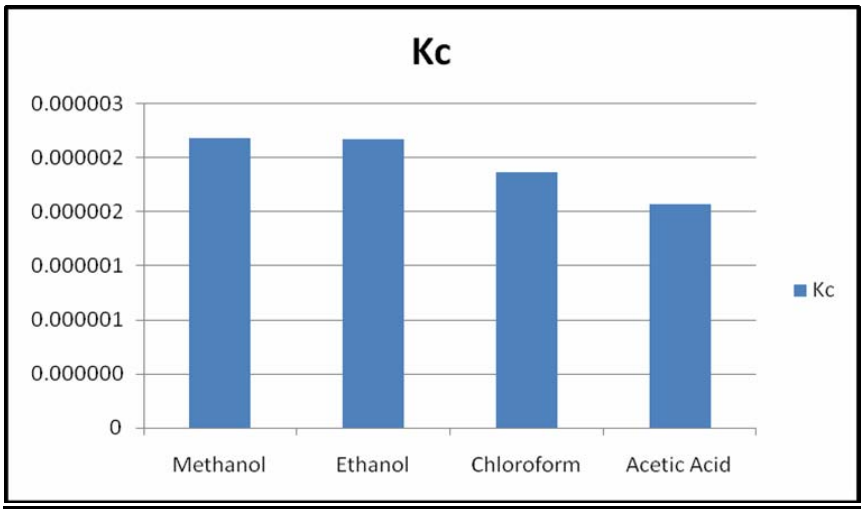

Figure 9: MTC for different solvents in Packed Bed Extractor at Flowrate of 30 ml/min

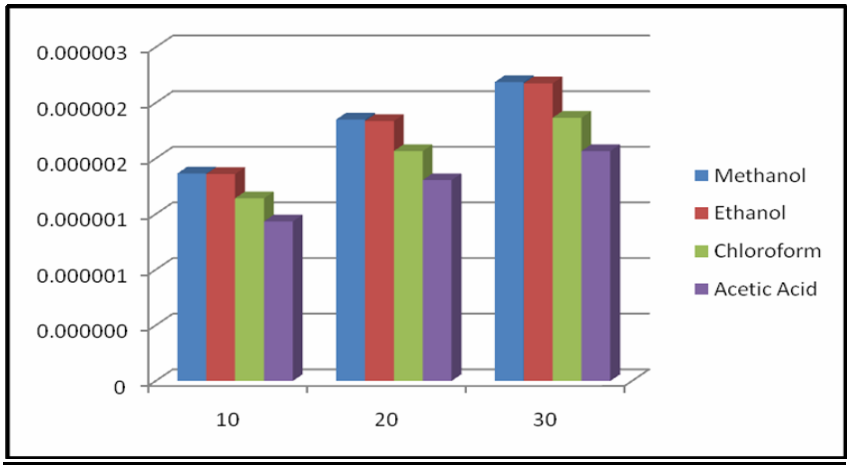

Figure 10: MTC of Packed Bed Extraction at different Flow rate of different solvents.

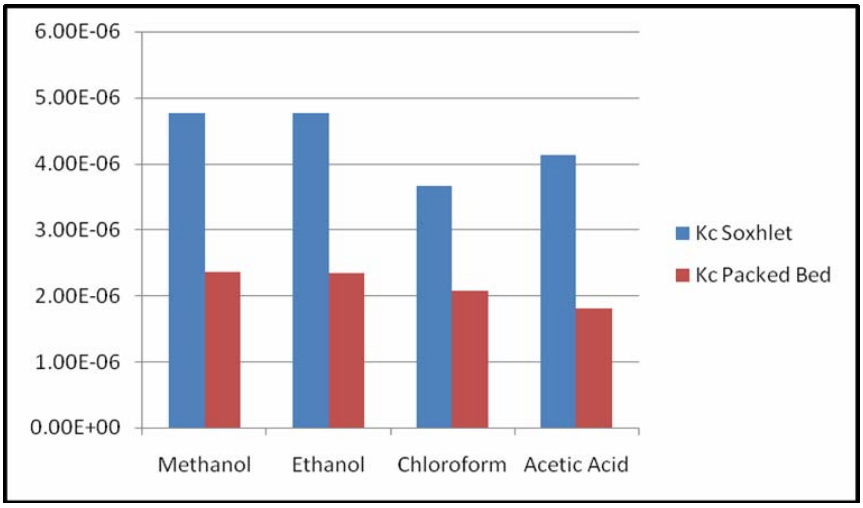

Figure 11: Comparison of MTC Of Soxhlet Extraction & Packed Bed Extraction.

Result and Discussions

MTC For Soxhlet Extraction

The four extraction experiments with different solvents (Methanol, Ethanol, Chloroform, Acetic Acid) were carried out in Soxhlet Extractor at constant solvent flow rate of 20 ml/min (Figure 6). MTC were calculated. MTC for Soxhlet extraction using Methanol as solvent found to be highest (4.77×10^{-6} m/s) where as MTC for Soxhlet Extraction using Acetic Acid as solvent found to be lowest (3.6678×10^{-6} m/s).

MTC For Packed Bed Extraction

Flowrate = 10 ml/min

For the solvent flow rate of 10 ml/min, MTC for Packed Bed Extraction was maximum at 1.8652×10^{-6} m/s for Methanol and was minimum at 1.4344×10^{-6} m/s for Acetic Acid (Figure 7). It should be noted that MTC for Ethanol as a solvent found to be nearly same as that of Ethanol (1.8600×10^{-6}).

Flowrate = 20 ml/min

For the flow rate of 20 ml/min, MTC in Packed Bed Extraction found to be increased than that of flow rate at 10 ml/min(Figure 8). Highest MTC found for Methanol at 4.77×10^{-6} m/s and lowest MTC found for Acetic Acid at 2.0677×10^{-6} m/s.

Flowrate = 30 ml/min

When the extraction was conducted at solvent flow rate of 30 ml/min, overall drop was observed in the values of MTC (Figure 9). However, the same trend is observed with maximum MTC for Methanol (2.6872×10^{-6} m/s) and minimum MTC for Acetic Acid (2.0668×10^{-6} m/s).

Comparison of MTC of Packed Bed Extraction at Different Flow Rates oOf Different Solvents.

Overall 12 experiments were conducted to study the effect of flow rate as well as solvents on MTC of Packed Bed Extraction (Figure 10). After conducting experiments it was found that –

i) Methanol could be considered as best solvent to extract Parthenin from Parthenium Hysterophorus.
ii) Optimum flow rate could be 30 ml/min.
iii) Maximum MTC was obtained for Methanol at flow rate of 30 ml/min (4.77×10^{-6} m/s)
iv) Lowest MTC was obtained for Acetic Acid at flow rate of 10 ml/min (2.066×10^{-6}).
v) Methanol and Ethanol exhibits nearly or approximately same MTC for all flow rates. Thus we could state that alcohols exhibits same behavior in case of extraction carried out in Packed Bed/ Fixed bed or Static Bed extractor.
vi) MTC values found to be minimum at 10 ml/min and maximum at 30 ml/min. MTC found to be increasing with increase in flowrates. This would be due to increase in minimum required time to

wet the surface area, to penetrate the micropores and to extract Parthenin from the plant of Parthenium Hysterophorus. [6]

Comparison of MTC of Soxhlet Extraction and Packed Bed Extraction.

For the constant flow rate of 20 ml/min, four experiments for four different solvents were performed on Soxhlet extractor and same four experiments, under identical conditions were performed on Packed Bed extractor (Figure 11). After performing eight experiments in total following observations were noted –

1) Highest MTC is obtained in Soxhlet Extractor. (4.77×10^{-6} m/s)
2) Lowest MTC is obtained in Packed Bed Extractor.(1.81×10^{-6} m/s)
3) Out of all four solvents , Methanol is found to be best since Parthenin dissolves in it preferably as compared to other solvents.
4) Every solvent shows better extraction efficiency in Soxhlet Extractor than that of Packed Bed Extractor.

Comparison Between Soxhlet and Packed Bed Extraction

According to study, extraction conditions and particle size and microstructure affected the process. Parthenin was extracted from a particle into the solvent in 5 major steps-

1. Entry of the solvent into the particle ,
2. Redistribution of solvent and expansion of the solid matrix,
3. Dissolution of Parthenin depending on solvent properties and temperature,
4. Diffusion of Parthenin to the exterior of the particle, and
5. Migration of the extracted Parthenin from the particle surface into the bulk solution of the methanol solvent.

The Soxhlet and static extraction (fixed bed extraction) have steps (1)-(3) above in common.

Soxhlet extraction increases the rate controlling diffusion and migration of Parthenin compared to static extraction. Distilled pure solvent continuously washes away Parthenin on the particle surface. Gradually the solvent inside the particles will contain less solute increasing its capacity for dissolution of Parthenin and other molecules. After certain time all extractable molecules will be found in the collecting flask.

On the other hand, during static extraction the concentration of Parthenin and other molecules will increase in the bulk solution. This will slow down diffusion and possibly also the dissolution of Parthenin and other molecules **[10]**. The concentration of Parthenin and other extractable substances will reach a constant pseudo-equilibrium level in the bulk solution. Thus, static or fixed bed extraction of Parthenin from Parthenium Hysterophorus is slow and complex process.

Advantages and Disadvantages of Soxhlet Extraction over Fixed Bed Extraction.

Advantages:

1) The displacement of transfer equilibrium by repeatedly bringing fresh solvent in contact with the solid matrix.
2) Maintaining a relatively high extraction temperature with heat from distillation flask, and
3) No filtration requirement after leaching.
4) Also, the Soxhlet method is very simple and cheap **[15]**.

Disadvantages:

1) Extraction time is long.
2) Large amount of solvent is used.
3) Agitation cannot be provided in the Soxhlet device to accelerate the extraction.
4) The large amount of solvent used requires an evaporation/ concentration procedure; and
5) The possibility of thermal decomposition of target compounds cannot be ignored as the extraction usually occurs at the boiling point of the solvent for the long time **[15]**.

Conclusions

The comparison of two applied methods shows that Soxhlet extraction is an efficient method by which solute of limited solubility can be extracted with higher efficiencies.

Seemingly, the extraction of Parthenin from Parthenium Hysterophorus is a very complicated process. Our experiments indicate that the extracted amount of Parthenin is affected by several parameters like solvent properties, solvent volume, extraction time, temperature, particle size and the microstructure of the sample.

Detailed systematic study is still required on the chemical transformations, to ascertain the physical, chemical and biological properties of Parthenin and its transformation products and their efficacy in multilocation fields. This may lead to commercial exploitation of Parthenin and its transformed products in pest control.

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