

One Step Synthesis of Polythiophenes from the Partially Purified Crude Extract of the Roots of *Tagetes Erecta*

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Received: July 5, 2016 Accepted: July 20, 2016 Online Published: August 4, 2016

doi:10.5539/ijc.v8n4p1

URL: <http://dx.doi.org/10.5539/ijc.v8n4p1>

Abstract

Polythiophene and its derivatives are valuable conjugated polymers due to their physical properties and applications in various fields. Precursors of polythiophenes were conventionally obtained from petrochemical byproducts, which are non-renewable. Thiophene and its derivatives had been found naturally in *Tagetes* species such as *Tagetes erecta*, *Tagetes tenuifolia*, etc. *Tagetes* species produce 2, 2':5', 2''-terthienyl (Alpha-T), 5-(3-buten-1-ynyl)-2, 20-bithienyl (BBT), 5-(4-hydroxy-1-butynyl)-2, 20-bithienyl (BBTOH), and 5-(4-acetoxy-1-butynyl)-2, 20-bithienyl (BBTOAc), which are accumulated in different parts, specifically in the roots of the plants. These thiophene derivatives were extracted and identified by GC-MS, Uv-vis, FTIR and TLC analysis. The partially purified plant extract is polymerized through one step oxidative free radical polymerization to obtain polythiophenes. Product characterization has revealed that the selective polymerization of thiophene derivatives results in the phase-separated end product from the rest of the materials in the plant-extract.

Keywords: conjugated polymer, free radical oxidative polymerization, polythiophene, thiophene derivative

1. Introduction

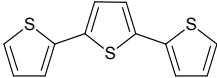
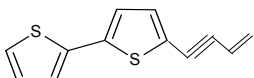
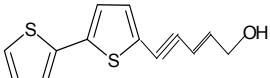
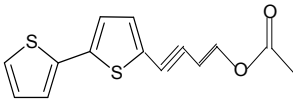
Thiophene and its derivatives have versatile applications in pharmaceuticals, agrochemicals, dye manufacturing, bio diagnostics, organic electronics and optoelectronic devices.(Barbarella & Sotgiu, 2005),(Abdou, 2013),(Mabkhot et al, 2015) Polythiophene has become a very valuable material for altering the photostabilization, brittleness, hardness, and other properties of plastic, and to alter the coloring of dye.(Abdou, 2013),(Balakit, Ahmed, El-Hiti, Smith, & Yousif, 2015). In the pharmaceutical industry, thiophene and its derivatives are used to impart pharmacological properties such as the anti-inflammatory, the antimicrobial, and the antianxiety.(Souza et al, 2012) Oligo and polythiophene are utilized as an indicator to recognize DNA or protein.(Barbarella & Sotgiu, 2005) Azo thiophene is used in the dye manufacturing industry, where its heterocyclic nature allows excellent sublimation rapidity on the dyed fiber.(Abdou, 2013) Polymer based solar devices (also known as organic solar cells) have been recently introduced as a competitive system for inorganic Si based solar cells.(Tang, 2010) Coatings of such materials have the ability to convert many physical objects into active light harvesting solar cell devices. Poly 3-hexylthiophene and poly(3,4-ethylenedioxythiophene) are commercially available conducting polymers, which are famous for designing organic solar devices, field effect transistors and thermoelectric devices.(Yao, Wen, Xu, Zhang, & Duan, 2013)

Precursors of polythiophenes are mainly obtained from non – renewable petrochemical byproducts, and they have competitive pricing. The non-renewability of petroleum resources and their high consumption rates will cause the depletion of petroleum in the near future as predicted by the Hubbert peak theory.(Hubbert, 1956) Therefore, identifying natural sources for green chemicals and developing green manufacturing processes have become the trend as an alternative for petrochemicals.

Precursors of polythiophenes are also available naturally in some plants of the Asteraceae family, such as, *Tagetes tenuifolia*, *Tagetes erecta*, *Tagetes patula*, *Porophyllum ruderale* and *Bidens radiata species*. (Takahashi et al, 2011),(Silva et al, 2011) Out of such plants *Tagetes species* are commonly available in tropical countries as an ornamental flowering plant, which are commonly known as Marigold. Thiophenes and thiophene derivatives in *Tagetes* plants are biologically active compounds in the defense system of the different parts of the plant.(Marotti et al, 2010) The concentration of thiophene in plant tissues increases as the plants get older, and reaches the maximum during the reproductive stages.

Several studies have been carried out to identify the different thiophene derivatives that exist in *Tagetes* plants. Natural thiophene derivatives identified in *Tagetes* species consist of two or three thiophene rings connected at α - position of the ring. These compounds include 2, 2':5', 2''-terthienyl (Alpha-T, α T), 5-(3-buten-1-ynyl)-2, 20-bithienyl (BBT), 5-(4-hydroxy-1-butynyl)-2, 20-bithienyl (BBTOH), and 5-(4-acetoxy-1-butynyl)-2, 20-bithienyl (BBTOAc). (Munhoz, 2014) Alpha-T consists of three connected thiophene rings, whereas BBT, BBTOH and BBTOAc consist only two thiophene rings with one alkyl side chain attached to the α - position of the ring. Therefore, alpha T can be considered a natural trimer that can be used to synthesize polythiophene. The structural characterization of the above thiophene derivatives were studied by many research groups and are summarized in Table 1. (Marotti et al, 2010), (Takahashi et al, 2011), (Silva et al, 2011), (Saha, Walia, Kundu, Kumar, & Joshi, 2012), (Vijay, Laxman, Balasaheb, Yuvraj & Janardhan, 2013), (Deineka, Tret'yakov, Lapshova, & Deineka, 2014)

Table 1. Different type of thiophene derivatives identified in roots of *Tagetes* species

IUPAC Name of the compound	Chemical structure	Abbreviation
2,2',5',2''-terthiophene		α - T
5-(3-buten-1-ynyl)-2,2'-bithienyl		BBT
5-(4-hydroxy-1-butynyl)-2,2'-bithienyl		BBTOH
5-(4-acetoxy-1-butynyl)-2,2'-bithienyl		BBTOAc

The aim of this study is to extract thiophene derivatives from *Tagetes erecta* and partially purify the thiophene derivatives from the crude extract. The potential of synthesizing polythiophenes from the partially purified crude extracts were studied and the resulting polymers were compared with polythiophenes made from commercially available thiophene monomers. One of the major objectives of this study was to establish a single step synthesis of polythiophene derivatives from the plant extract, and establish the purification of thiophenes during the polymerization.

2. Materials and Methodology

The roots of *Tagetes erecta* plant were harvested after their full flowering stage (about three months from planting) from the Gampaha botanical garden (Longitudes = 79.9857°, Latitudes = 7.0993°), Sri Lanka. Analytical grade solvents and chemicals were used throughout the research.

2.1 Extraction and Partially Purification of Thiophenes

Lateral (hairy) and primary roots were separated from collected *Tagetes erecta* plants. Roots were washed several times with distilled water to remove soil particles and other water-soluble impurities. Thereafter, roots were dried under a shade at room temperature until obtaining a constant weight. Dried roots were ground into a fine powder using a lab scale mechanical grinder. Defatting of root samples was done using petroleum ether to remove fat components present in the roots. GC-MS analysis was carried out to check whether thiophene components were also removed during defatting.

Fine powders of air-dried roots were refluxed with methanol: water (3:1) at 78 °C. The extract was filtered through cotton wool to remove insoluble impurities. The filtered extract was concentrated *in vacuo* (Laborota 4000 Efficient) at 50 °C. The concentrated extract was sequentially partitioned into hexane: chloroform (2:1) solvent system. The extract was treated with anhydrous sodium sulfate (1 g for 400 ml of the extract) to remove water. Then the partitioned fraction in the hexane: chloroform system was subjected to rotary evaporation at 35 °C under reduced pressure to further dry the extract to get a dark brown residue. The dried product was kept in a desiccator for 24 hours to achieve a completely dried solid crude extract.

Separation of compounds for the partial purification was carried out for the crude extract by using hexane and acetone.

A 16.60 g of the dried crude extract was successively extracted three times with 30 ml of hexane at each extraction. The hexane fractions were collected to a 100 ml round-bottomed flask and desolvatization was carried out using the rotary evaporation under reduced pressure at 45 °C to near dryness and completely dried by purging gas until it obtained a dark yellow solid residue. This crude residue was further dried for 24 hours using a desiccator. The weight of the partially purified hexane soluble fraction (HSF) was obtained and the mass yield was calculated. The hexane insoluble solid residue was extracted into 30 ml of acetone. Acetone fraction was dried by the rotary evaporation and further dried by keeping it in a desiccator for 24 hours. The weight of the acetone soluble fraction (ASF) was recorded and the mass yield was calculated. (Saha, Walia, Kundu, Kumar, & Joshi, 2012)

2.2 Analysis of the Partially Purified Product

HSF and ASF were analyzed using GC-MS, UV-Visible, FTIR and TLC in order to confirm the availability of thiophene derivatives in each solvent fraction.

2.2.1 Gas Chromatography-mass Spectroscopy (GC-MS)

The GC-MS analysis of plant extracts was carried out on a Agilent 5975C chromatograph coupled with a quadrupole mass detector and a HP-5MS 5% Phenylmethylsiloxane capillary column (30.00 m × 250 μm, film thickness 0.25 μm). A small aliquot (2 μl) of the sample was injected into the column. High pure Helium gas was used as the inert carrier gas at a flow rate of 1 mL min⁻¹. The initial temperature of the column was 70 °C which was maintained for 3 minutes and then the temperature was increased at a constant rate of 10 °C min⁻¹ for 21 minutes. The final temperature of the column was 280 °C and it was maintained for 6 minutes. The total retention time was 30 minutes. The chromatograms were analyzed using the NIST08 mass spectral database.

2.2.2 Ultraviolet-visible Absorption Spectroscopy (UV-Vis)

A small amount of the partially purified product (0.050 g) was dissolved in hexane (10 ml) and it was diluted to 100 times to get a colorless solution. The absorption spectra of these diluted samples were obtained using UV-Visible spectrophotometer (Perkin-Elmer, Lambda 35 spectrophotometer) within the spectral range of 200-400 nm. Hexane was used as a blank.

2.2.3 Fourier Transform Infrared Spectroscopy (FTIR)

A small amount of the extract (0.01 g) was dissolved in hexane (5 ml). One drop of that solution was taken to the analysis. The FTIR spectra were recorded in the range 600–4000 cm⁻¹ with a 4 cm⁻¹ resolution from ATR on a FT-IR system (Thermo Scientific Nicolet IS10). FT-IR bands were analyzed using OMNIC software (version Windows XP professional).

2.2.4 Thin Layer Chromatography (TLC)

A small quantity of (10 mg) of the partially purified extract having thiophene derivatives was dissolved in hexane (5 ml) and a drop of the solution was spotted on commercially available TLC plates (pre-coated TLC- sheets ALUGRAM® Xtra SIL G/UV254) using capillary tubes and solvent systems were used as the mobile phase as shown in Table 2. The best solvent system for separation of components in the partially purified extract was selected by observing the maximum separation of spots.

Table 2. TLC solvent systems used to separate the plant extract

System no.	Solvent system	Composition
1	Hexane : Acetone	10 : 0.1
2	Hexane : Acetone	9 : 1
3	Hexane : Acetone	8 : 2
4	Hexane : Acetone	7 : 3
5	Hexane : Acetone	6 : 4

Spots were visualized either by putting plates in an iodine bath or using UV lamp having wavelengths of 254 and 366 nm. The distribution of spots was recorded and R_f values were calculated with respect to the distance traveled by the solvent front.

The best solvent system was used to separate alpha T from the extract having thiophene derivatives. The extract having thiophene derivatives was purified to separate alpha T by using preparative TLC (silica gel 60 F254, layer thickness 0.25 mm, 10×20 cm). Each separated band was scratched out from TLC plates, dissolved into the relevant solvent, filtered and subjected to GC-MS analysis to confirm the availability of thiophene derivatives.

2.3 Synthesis of Polymers from Thiophene Monomers

2.3.1 Polymerization of Pure Thiophene

Thiophene (0.48 ml) was mixed with 70 ml of chloroform in a round-bottomed flask. Anhydrous FeCl_3 (1.0000 g) was dissolved in 180 ml of chloroform and added into the monomer solution in drop wise manner while stirring. The weight ratios of pure thiophene monomer to the oxidant were changed as 1:2, 1:5 and 1:7. The polymerization was carried out for 4 hours at room temperature. The dark-brown precipitate was collected by filtration and washed with anhydrous CHCl_3 to remove excess FeCl_3 . The precipitated products were further washed with anhydrous methanol to remove the residual oxidant. The powder products were dried in a vacuum drier at 55°C for 24 hours. (Gok, Omastova, & Yavuz, 2007)

2.3.2 Polymerization of the Plant Based Thiophene Monomers

The composition of the plant extract was determined based on the area under the curve of the calibrated Gas Chromatogram. The plant extract (0.9823 g) was mixed with 70 ml of chloroform in a round-bottomed flask containing a magnetic stir bar. Anhydrous FeCl_3 (4.9115 g) was dissolved in 180 ml of chloroform and it was added drop wise into the monomer solution while stirring. The weight ratios of plant extract and the oxidant were varied as 1:2, 1:5 and 1:7. Polymerization was carried out for 4 hours at room temperature. The dark-brown precipitate of the product was collected by filtration and washed with anhydrous CHCl_3 to remove excess FeCl_3 .

The product was further washed with anhydrous methanol to remove the residual oxidant. During this procedure, the colour changed from black to dark brown. The powder product was dried in a vacuum drier at 55°C for 24 hours.

2.4 Characterization of the Polymerized Products

2.4.1 Fourier Transform Infrared Spectroscopy (FTIR)

About 2 mg samples of the synthesized polymers were taken to the analysis. The FTIR spectra were taken using Thermo Scientific Nicolet IS10 within the wavenumber ranging from 600 to 4000 cm^{-1} . Instrument was used at the ATR mode with resolution of 4 cm^{-1} . FTIR spectral bands were analyzed using OMNIC software. The resulted spectra were compared with the reference FTIR spectra in literature which were obtained for polythiophene from pure monomers. (Marani, & Entezami, 1993), (Camurlu, Cirpan, & Toppare, 2005), (Gnanakan, Rajasekhar, & Subramania, 2009), (Karim, 2012)

2.4.2 X-Ray Diffraction (XRD)

Wide angle X-ray diffraction patterns of powder form of polymerized products were recorded using "Rigaku-Ultima IV" X-ray diffractometer with $\text{Cu} - \text{K}_\alpha$ radiation ($\lambda = 1.54 \text{ \AA}$). Samples were scanned at the rate of 2° min^{-1} in reflection mode over the 2θ range of $3^\circ - 45^\circ$. The resulted XRD spectra were compared with the literature available data. (Sakthivel, & Boopathi, 2014) Peak deconvolution was carried out using the Peak Fit 4.12 software. Degree of crystallinity of polymer was calculated using peak area measurements, which is shown in equation

$$\text{Degree of crystallinity} = (\text{Peak area of crystalline peaks} / \text{Total area under the curve}) \quad (1)$$

2.4.3 Ultraviolet-Visible Absorption Spectroscopy (UV-Vis)

A small amount of powder of the product (0.050 g) was dissolved in mono chlorobenzene (5 ml). Monochlorobenzene was selected as the blank. UV – Visible spectra of polymer solutions were recorded with respect to the blank using UV-Visible spectrophotometer (Perkin-Elmer, Lambda 35 spectrophotometer) within the spectral range of 200-400 nm.

3. Results and Discussions

3.1 Extraction Process

Few extraction methods are mainly reported in literatures such as steam distillation, soxhlet extraction, room temperature solvent extraction (maceration) and reflux extraction. In high thermal treatment thiophene derivatives are susceptible to degrade. Therefore, the steam distillation method was not used for the extraction. The soxhlet extraction method is suitable for a single solvent and is not possible to use for a solvent mixture. Therefore, the soxhlet technique was not used for this extraction. Moreover, room temperature solvent extraction may not be the most effective method because of its low temperature conditions. Furthermore, this extraction process consumes time because the plant material (roots) should be dipped for several days to obtain a considerable amount of the extract. Compared with other natural products present in the extract thiophene content is very low. Accordingly, the maximum attainable amount of thiophene derivatives should be extracted into the solvent. Therefore, the reflux extraction method was selected at 78°C as the most appropriate extraction method.

GC-MS analysis of defatted samples had revealed that some thiophene derivatives (especially $\alpha - \text{T}$) was extracted with the fat components. Hence, it caused to reduce the yield of thiophene derivatives. Therefore, the extraction process

was optimized without defatting the roots. Methanol-water solvent system was directly used to extract thiophene derivatives from the roots of the plant. The methanol – water solvent system is a polar solvent system, which can break down both cell walls and cell membranes thereby thiophene and its derivatives can easily be extracted into the solvent system.

Thiophene derivatives can be dissolved well in non-polar organic solvents. Hence, hexane was used to partially purify the crude plant extract. The hexane soluble fraction (HSF) was yellow in color. The remaining hexane insoluble residue was soluble in acetone and it has a dark red color. The mass yield of the dried HSF was obtained with respect to the mass of dried roots using equation 2 and the results are tabulated in Table 3.

$$\text{Percentage mass yield of HSF} = (\text{Weight of HSF (g)} / \text{Weight of the dried roots (g)}) \times 100 \quad (2)$$

Table 3. Percentage yield of HSF and ASF with respect to the weight of the dried roots

Total weight of dried roots/g	Percentage yield of HSF	Percentage yield of ASF
2500	0.288	0.376

3.2 GC-MS Analysis of the Partially Purified Extract

Zoomed GC-MS spectra obtained for both the hexane and the acetone soluble fractions are shown in Figure 1(a) and (b) respectively. According to the NIST08 mass spectral data base, the peak at the retention time of 22.46 minutes in Figure 1(a) corresponds to the elution band of alpha – T and 22.23 minutes to the BBTOH.

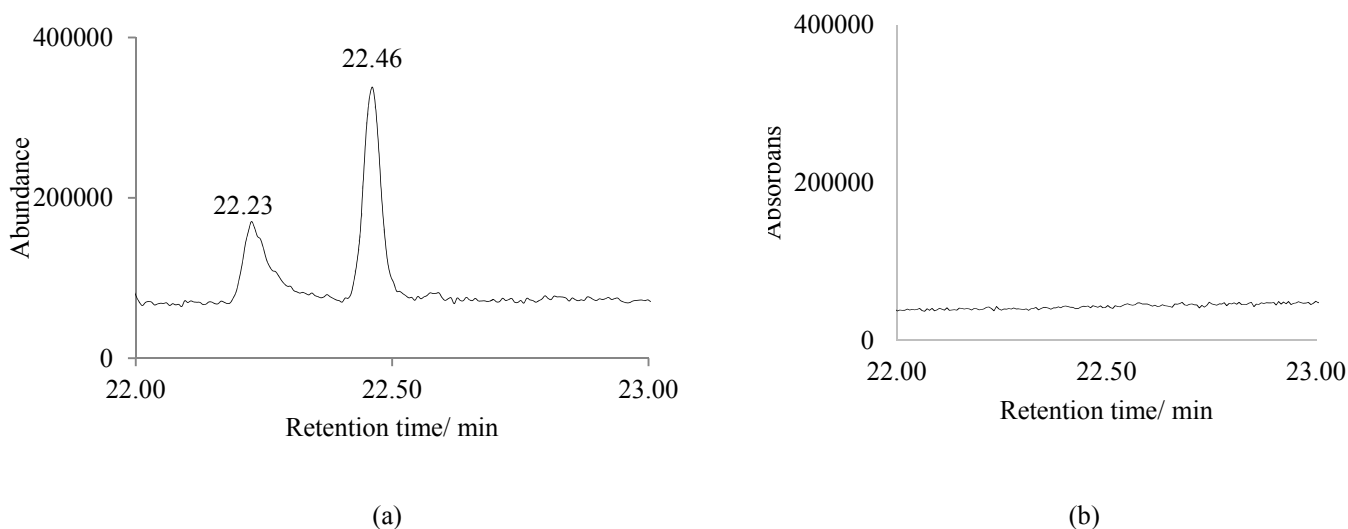


Figure 1. Gas chromatograms (GCs) of (a) HSF, (b) ASF of the plant extract of *Tagetes erecta*

Table 4. Major compounds observed from the Gas Chromatogram of HSF

Retention time (R _t)/min	Chemical Name	Chemical Structure
16.536	Benzeneacetic acid, alpha, 3,4 tris[(trimethylsilyl)oxy]-, trimethylsilyl ester	
19.336	Pentadecanoic acid, 14-methyl, methyl ester	
20.994	Octadecanoic acid, methyl ester	
22.231	5-(4-hydroxy-1-butenyl) 2, 2-bithienyl, 5-(4-acetoxy-1-butenyl)	
22.400	2, 2':5', 2''-Terthiophene	
24.909	1,2- Benzenedicarboxylic acid, mono(2-ethylhexyl) ester	

Mass spectra of the GC – MS for both alpha –T and BBTOH in the HSF are shown in Figure 2 (a) and (b). These MS spectra for alpha –T and BBTOH were further confirmed by comparing molar mass of molecule ions and other major fragments. The summary of possible molecular fragments for highly abundance peaks are summarized in Table 5 and 6.

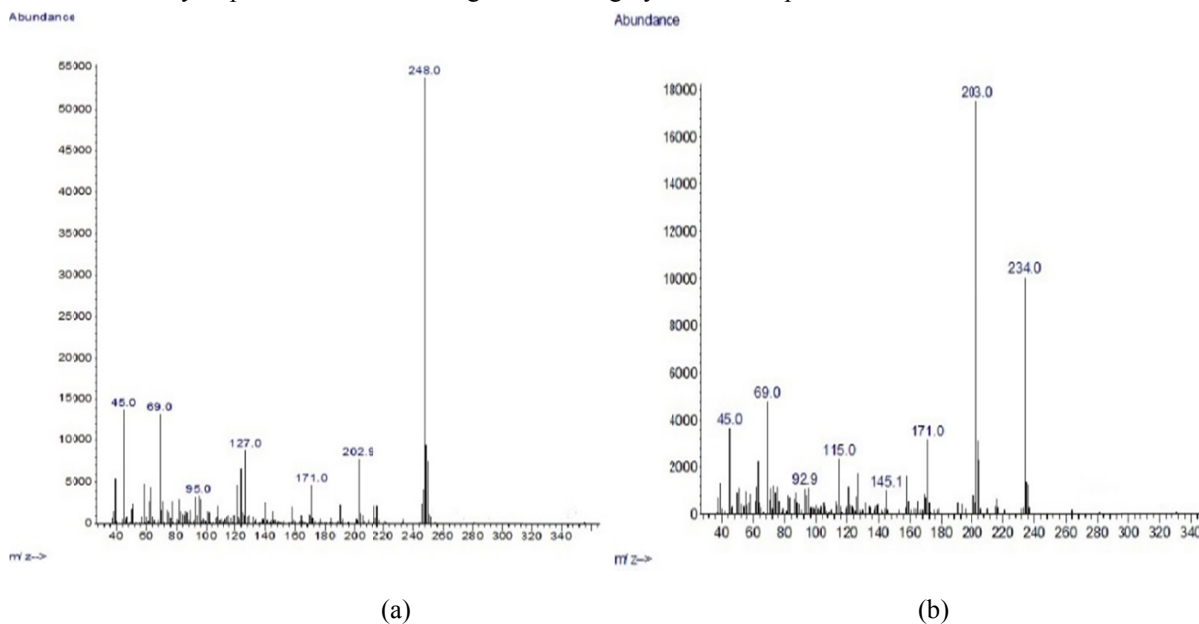


Figure 2. Mass spectra of GC – MS peaks of (a) alpha – T and (b) BBTOH

Table 5. Possible molecular ion fragments of Alpha – T

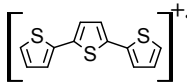
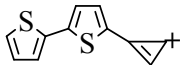
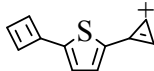
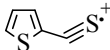

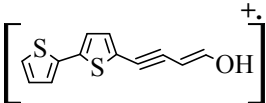
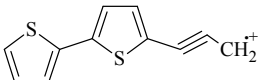
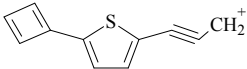
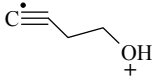
m/z value	Possible molecular ion fragments
248	
203	
171	
127	
69	
45	$\text{HC}^+=\text{S}$

Table 6. Possible molecular ion fragments of BBTOH

m/z value	Possible molecular fragments
234	
203	
171	
69	
45	$\text{H}_2\text{C}^+-\text{OH}$

3.3 UV-Vis Analysis of the HSF

UV-Vis spectra of pure thiophene monomers and HSF are shown in Figure 3 (a) and (b) respectively.

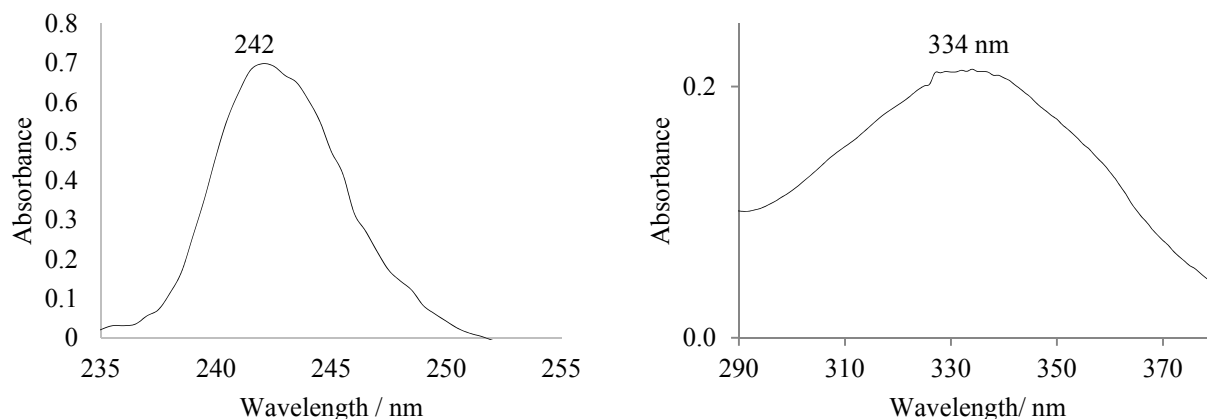


Figure 3. Uv-vis absorption spectrum of (a) thiophene monomer and (b) HSF of the extract of *Tagetes erecta*

The absorption maximum (λ_{\max}) of pure thiophene monomers was observed at 242 nm. However, the λ_{\max} of HSF was observed around 330 nm. This difference in λ_{\max} values might be due to the presence of other components in the HSF and due to the conjugation of thiophene monomers. Due to the conjugation, thiophene derivatives in HSF show a bathochromic shift with respect to pure thiophene.

3.4 FTIR Analysis of the HSF

FTIR spectroscopic analysis has been carried out to identify the possible functional groups available in HSF of the extract. Relevant FTIR spectrum of HSF is shown in Figure 4.

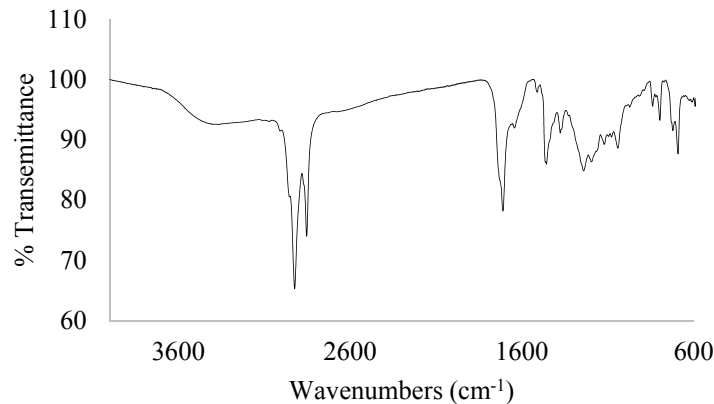


Figure 4. FT-IR spectrum of HSF of the plant extract

A detailed summary of the FTIR peak analysis is shown in Table 7. The tabulated results confirmed the presence of functional groups available in both alpha – T and BBTOH in the HSF of the plant extract. These results are summarized in Table 7.

Table 7. Summary of the band analysis of FTIR spectrum of the hexane soluble fraction

Wavenumber/cm ⁻¹	Bond	Possible origin of functional groups
3417	O-H stretch, H-bonded	Alcohols
2954	v _{as} -CH ₃ stretching	Alkanes
2921-2853	aliphatic -CH ₂ stretching	Alkanes
1710	C=O stretch	α, β- unsaturated esters
1642	C-C stretch (in- ring)	Aromatics
1457	The ring vibration of 2,5-disubstituted thiophene	Terthiophene and BBTOH
1377	C-C ring stretch	Aromatics
1241	The ring vibration of 2,5-disubstituted thiophene	Terthiophene and BBTOH
1041	-CH ₂ -CH ₂ -OH group	BBTOH
841	Angular deformation out of the plane of thiophene ring	Terthiophene and BBTOH
796	C-H deformation vibration of 2,5-disubstituted thiophene	Terthiophene and BBTOH
721	C-S bend	Thiophene

The availability of thiophene rings in HSF can be confirmed by FTIR absorption bands observed at 721 cm⁻¹ for C-S bending and aromatic C-C ring stretching bands at 1642 cm⁻¹ and 1377 cm⁻¹. FTIR absorption bands observed at 1457 cm⁻¹, 1241 cm⁻¹ and 796 cm⁻¹ show the connectivity of thiophene rings through 2,5 coupling. (Marani, & Entezami, 1993), (Camurlu, Cirpan, & Toppare, 2005), (Gnanakan, Rajasekhar, & Subramania, 2009), (Karim, 2012), (Sakil, Singh, & Roy, 2013) Other absorption bands may correspond to the side chains attached to thiophene and the impurities present in HSF.

3.5 TLC of the HSF

Several solvent systems with different polarities were used to separate the components available in HSF using TLCs. Those solvent systems are summarized in Table 2. The gas chromatogram obtained for the component extracts from the upper band of TLC is shown in Figure 5. However, BBTOH was not able to observe from GC. This might be due to the very low concentration of BBTOH in the extract.

Table 8. Results of the TLC analysis for the HSF

R _f of the separated spot	Under illumination of 254 nm	Under illumination of 366 nm
0.83	Brown	Yellow
0.78	Brown	Blue
0.70	Brown	Absent
0.48	Brown	Absent
0.33	Brown	Pale yellow
0.12	Brown	Blue

Literatures have been reported that n-hexane: acetone 10:0.1 solvent system was used to develop TLCs for the plant extract, which was extracted from the flowers of *Tagetes* species such as *Tagetes tenuifolia*, *Tagetes patula* and *Tagetes erecta*. Reported R_f value for thiophene derivatives was around 0.9. (Deineka, Tret'yakov, Lapshova, & Deineka, 2014)

In this study, only two separated spots for the plant extract were observed on the TLC plate for *n*-hexane: acetone 10:0.1 solvent system. Out of those solvent systems hexane: acetone (9:1) had shown the maximum number of spots with better separation among the spots. The polar stationary phase retards the movement of polar compounds due to better intermolecular interaction between compounds and the stationary phase. Alpha - T (terthiophene) is a non- polar compound and has less preference to the stationary phase. Therefore, it travels easily with the mobile phase to give high R_f value ($R_f = 0.83$). Thiophenes isolated from *Tagetes patula* had given brown color spots under UV radiation at 254 nm. Further visual observation of thiophene compounds can be obtained by illuminating those with UV radiation at 366 nm, which gives a characteristic yellow color for alpha - T. (Adekunle, 2007) The summary of this TLC analysis is shown in Table 8. Through UV illumination it has been confirmed that the thiophene derivatives can only be available in the spot with the highest R_f value.

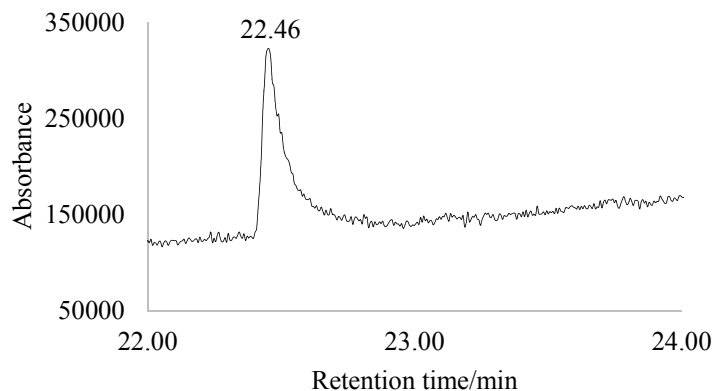


Figure 5. GC of the upper band in preparative TLC of HSF

These bands observed in preparative TLC plates were separately dissolved in hexane and subjected to GC-MS analysis. It was confirmed that the presence of Alpha - T in the upper band of the TLC.

3.6 Polymerization of Pure Thiophene Monomers and Plant Based Thiophene Derivatives

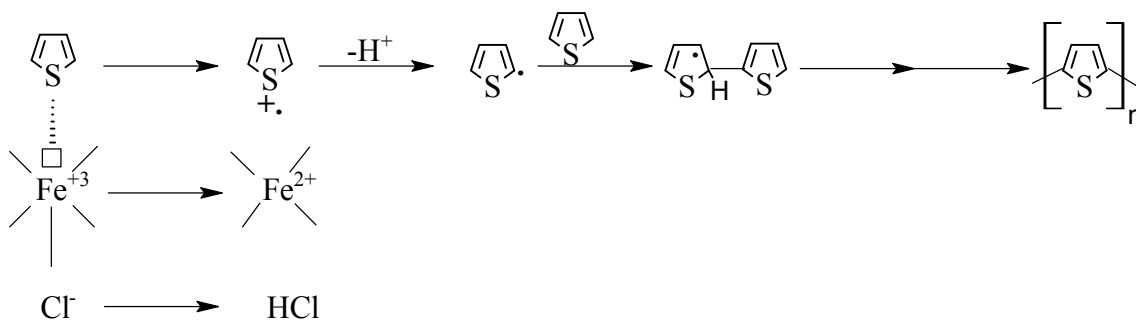


Figure 6. The possible polymerization mechanism of thiophene in the presence of FeCl_3 catalyst

Chemical oxidative polymerization was followed to obtain the polymerized product from the plant based thiophene derivatives and pure thiophene monomer separately. These thiophene polymerization reactions were catalyzed by anhydrous FeCl_3 . In this process, polymerization starts from solid FeCl_3 , due to the surface crystal Fe^{3+} ions being the active sites, which has one unshared chloride and one vacant orbital that has the Lewis acidity to accept electrons from sulphur atom of thiophene. This ability of Fe^{3+} ions is enough to oxidize the thiophene molecule into a radical cation. In parallel, Fe^{3+} reduces to Fe^{2+} . Therefore, ferric chloride acts as an oxidant and it initiates the radical oxidative cationic polymerization of thiophenes. (Niemi, Knuutila, & Osterholm, 1992)

One of the possible mechanisms of thiophene polymerization is shown in Fig. 6. Chloroform is used as the medium of polymerization. The excess amount of FeCl_3 was dissolved in CHCl_3 prior to adding thiophene monomer or plant based thiophene derivatives. After polymerization, the related mixture was filtered, and the remaining crude was washed with the excess volume of chloroform to remove residual unreacted alpha - T and other organic impurities. Thereafter, the product was washed with excess methanol in order to remove the remaining ferric chloride catalyst.

The best average percentage yield of 89% for the pure polythiophenes was obtained for 1:7 weight ratio of pure thiophene monomer to oxidant. The weight ratios of HSF to oxidant for the polymerization was altered to obtain the highest average percentage yield, which was 21.8 %, received for weight ratio of HSF to oxidant 1:7. When compared with the percentage yield of pure polythiophene, the plant based product is less. The yield can reduce due to the other side products of ferric chloride with aromatic compounds and the effects of iron chelating.

3.7 FTIR Analysis for the Polymerized Product

The FTIR spectra for the pure polythiophene and the plant based polythiophene are shown in Figure 7 (a) and (b) respectively.

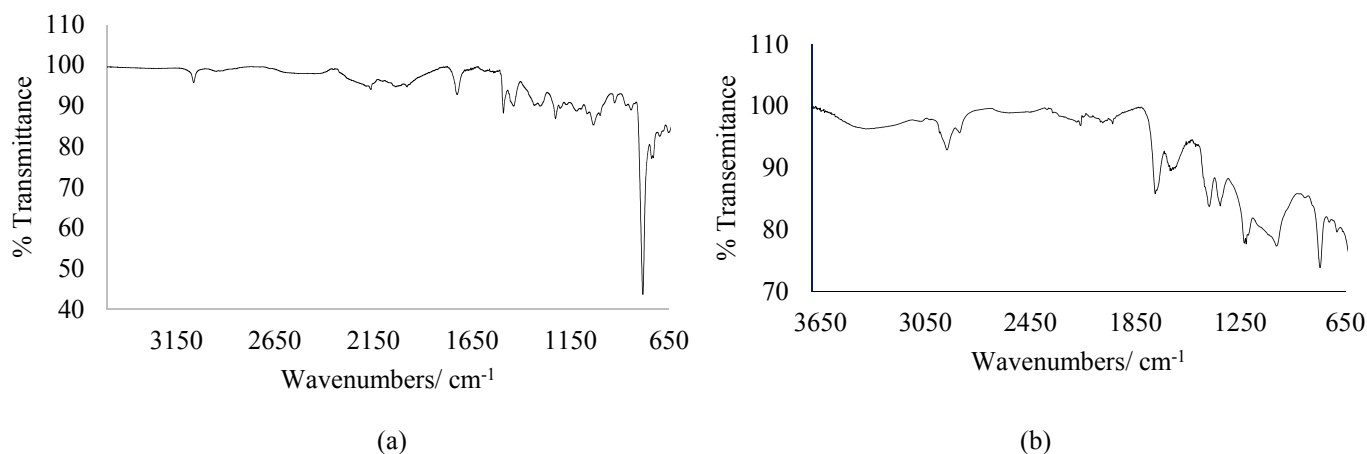


Figure 7. FTIR spectrum of (a) pure polythiophene, (b) plant based polythiophene

The common FTIR bands for both polythiophenes are summarized in Table 9.

Table 9. Summary of the common FTIR band analysis for polythiophenes made from pure thiophene monomer and plant based thiophene derivatives

Wavenumber/cm ⁻¹	Bond	Possible origin of functional groups
2950-3050	ν_{as} -CH ₃ stretch	Aromatic CH ₃
1029-1086	C-S stretch	Thiophene
781-801	α, α' - coupling of poly-2, 5-thiophene	Thiophene

The most significant band for identifying polythiophene is the band between 781 cm⁻¹ – 801 cm⁻¹ due to 2, 5-disubstitution of thiophenes. According to the polymerization mechanism, polymerization prefers to occur via 2, 5 coupling, which is also known as α, α' coupling of monomers. (Kelkar, & Chourasia, 2011) This is a very strong and narrow band. The FTIR absorption bands at 1029 and 1086 cm⁻¹, show C – S stretching. The asymmetric vibration of the -CH₃ group shows in between 2950 – 3050 cm⁻¹. When the FTIR bands of the plant-based product was compared with the pure polythiophene, it confirms the presence of similar functional groups in the plant-based product.

Table 10. Summary of FTIR band analysis of the plant based polythiophene

Wavenumber/cm ⁻¹	Bond	Possible origin of functional groups
3390	-OH stretch, H-bonded	Alcohols from BBTOH
2924 – 2854	aliphatic -CH ₂ stretch	Alkane from BBTOH
1456	-CH ₂ bend	Alkane from BBTOH
1376	-CH ₃ bend	Alkane
833	ω/τ thiophene ring	Terthiophene from BBTOH

In addition to common FTIR bands of polythiophenes, other bands present in the plant based polythiophene are tabulated in Table 10. The band at 3390 cm⁻¹ shows the availability of hydroxyl groups in the plant based polythiophene.

These hydroxyl groups might come from the polymerization of BBTOH in the plant extract. Other bands for aliphatic hydrocarbon chains may correspond to side chains attached to thiophenes.

3.8 Uv-vis Analysis of the Polymerized Products

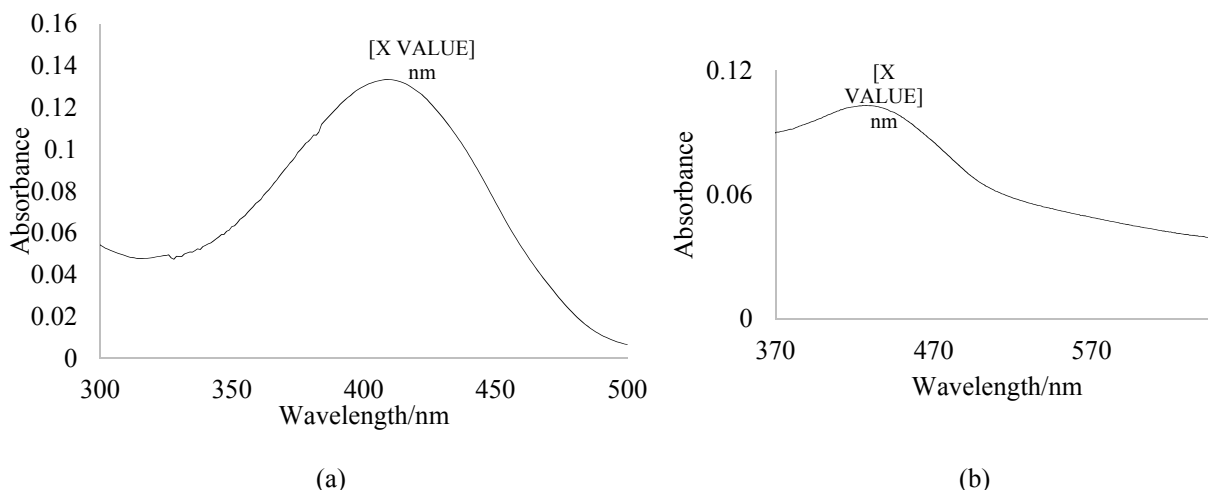


Figure 8. Uv-vis spectrum for (a) the pure polythiophene, (b) plant based polythiophene

UV spectroscopy looks at double bonds and lone pairs in compounds. Electrons can be promoted from the HOMO level to the LUMO level by absorbing radiation. The energy gap of bonding to antibonding (π - π^*) transition is higher than that of non-bonding to antibonding (n - π^*). Therefore, π - π^* transition requires short wavelengths compared to n - π^* transition. When the length of conjugation increases the gap for π - π^* , the transition narrows down gradually. The Uv-vis spectrum for the pure polythiophene and the plant based polythiophene are shown in Fig.8 (a) and (b) respectively.

The pure polythiophene obtained the absorption band at 413 nm and also the plant based polythiophene associated the absorption band at 432 nm. Therefore, the conjugation length of the plant based polythiophene is a little higher than that of pure polythiophene. The literature reported that these λ_{max} values may correspond to π - π^* transitions due to the main charge carriers such as polarons and bipolarons in polythiophenes. (Gnanakan, Rajasekhar, & Subramania, 2009)

3.9 XRD Analysis of the Polymerized Product

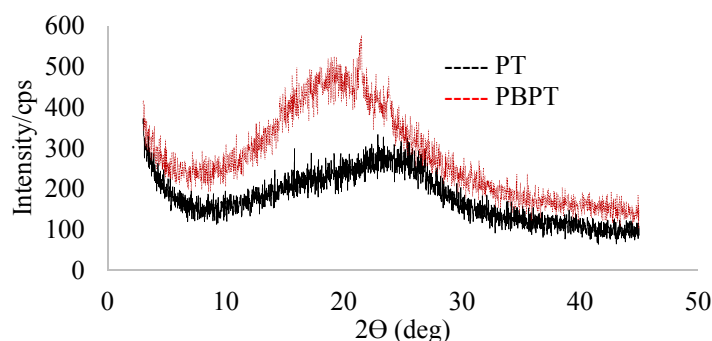


Figure 9. XRD spectrum for both polythiophenes

The XRD spectra showed for both pure polythiophene (PT) and plant based polythiophene (PBPT), which revealed three peaks near 2θ values of 19.61° , 21.34° and 23.79° . Among them the first large broad peak (at $2\theta = 19.61^\circ$) relates to amorphous peak and other two at $2\theta = 21.34^\circ$ and 23.79° small peaks indicates two crystalline peaks. These three peaks are overlapped together and the overall peak is given on Figure 9. But the pure polythiophene has shown only the amorphous peak. Polymer chain packing of polythiophenes made from pure thiophene monomers and the thiophene derivatives of the plant extract are different. Plant based polythiophenes may contain other side chains due to the polymerization of BBTOH with Alpha-T. Therefore, peak center of the amorphous diffraction may slightly shifter to lower theta (higher d values).

The peak deconvolution and degree of crystallinity of plant based polythiophene was calculated and summarized in Table 11.

Table 11. XRD peak characteristics for plant based polythiophene

Peak position at 2 θ /degree	Nature of peaks	Inter planar distance/ Å	Area of peaks	Degree of crystallinity
19.61	Amorphous	2.29	2690.1	Amorphous
21.34	Crystalline	2.10	88.6	3.1
23.79	Crystalline	1.91	103.6	3.6
Total			2882.3	6.7

4. Conclusions

The study had confirmed the possibility of polymerizing plant based thiophene derivatives in their crude extract into polythiophenes. This study was the first green polymerization reported in literature to direct polymerization of the crude extract of plant based thiophene derivatives to get the final polymerized product. Polythiophene formation from HSF confirms the minimal effect from other impurities present in the plant extract on polymerization of thiophene derivatives.

Thiophene derivatives in the roots of *Tagetes erecta* were extracted from methanol-water solvent system and partitioned into the hexane fraction. By analyzing the HSF through GC-MS, it was confirmed that two thiophene derivatives (α -T and BBTOH) were available with many other impurities like hydrocarbons, fatty acids, esters and carbonyl compounds in the root extract of *Tagetes erecta*. The FTIR and Uv-Vis spectra recognized the existence of connected thiophene rings while TLC results confirmed that the presence of thiophene derivatives in the HSF.

Polythiophene was obtained through direct cationic oxidative polymerization of thiophene derivatives in the HSF in the presence of anhydrous FeCl₃ catalyst. The percentage yields for plant based polythiophenes and pure polythiophenes concluded that the monomer to the oxidant weight ratios affect the overall yield. Synthesized plant based polymer and pure polythiophene were characterized by using FTIR, XRD and Uv-Vis techniques. The FTIR spectra also confirmed that certain other groups have been attached to the polythiophene chain apart from α -T, which were mainly aliphatic side chains, and hydroxyl groups that come from BBTOH and organic impurities present in the partially purified extract. XRD studies have shown that the resulting plant based polythiophenes were assembled into a semi-crystalline form. Through the analysis of XRD spectrum by Peak fit software, it was calculated that the degree of crystallinity of the prepared polymer was about 6.7%.

Acknowledgement

This work was financially supported by the Research Grant (ASP/06/RE/SCI/2013/07), University of Sri Jayewardenepura, Sri Lanka. UV-Visible studies were carried out with the instrument provided by the NSF, Sri Lanka (NSF Grant RG/2013/EQ/07)

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