Current Chemistry Letters 12 (2023) 375–382

Contents lists available atGrowingScience

Current Chemistry Letters

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Thermal and morphological studies of chitosan and agar-agar blends

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C H R O N I C L E

Article history:
Received July 12, 2022
Received in revised form
August 2, 2022
Accepted November 30, 2022
Available online
December 1, 2022

Keywords:
Chitosan
Agar-agar
Blends
Compositions
Glass transition
SEM
TGA

A B S T R A C T

Many researchers are attracted to Chitosan based blends due to its properties and potential applications in various fields. The advanced development of Chitosan blends integrates the science and technology of blended materials. The present experimentation is reporting the preparation of Chitosan and Agar-Agar blends (CCA) by chemical mixing of these materials at different compositions. The thermal studies of the prepared blends were studied by differential scanning colorimeter (DSC) and thermogravimetric analysis (TGA) tools. Thermal studies reveal that the lowest degradation temperatures of blends might be attributed to the partial miscibility of CAA blends at particular composition and miscibility due to single glass transition temperature (T_g) between Chitosan and Agar-agar. Bonding nature of sample blends were carried out by Fourier transform infrared (FT-IR) instrumentation. This study reveals the interaction between Chitosan and Agar-agar is partial miscibility. Morphological study reveals that a few aggregated particles, which suggest the partial miscibility of CAA blends. Homogeneity of blend compositions and specific intermolecular interactions of hydrogen bonding type is also observed.

1. Introduction

Generally, agar occurs as a structural carbohydrate in the cell walls of agarophytes algae, possibly existing in the form of its calcium salt or a mixture of calcium and magnesium salts. It is a complex mixture of polysaccharides composed of two major fractions namely agarose, a neutral polymer, and agaropectin, a charged sulfated polymer[1-2]. Agarose is a gelling fraction and also a neutral linear molecule essentially free of sulphates, consists chains of repeating alternate units of β-1, 3-linked- D-galactose and α-1, 4-linked 3, 6-anhydro-L-galactose. Agaropectin is a non-gelling fraction and is a sulfated polysaccharide (3% to 10% sulphate) composed of agarose and varying percentages of ester sulphate, D-glucuronic acid, and small amounts of pyruvic acid. The proportion of these two polymers varies according to the species of seaweed. In fact, Agarose normally represents at least two-thirds of the natural agar-agar. Regarding its gelling power, agar-agar is outstanding among other hydrocolloids. Agar-agar gels can be molded in very dilute solutions, containing a fraction of 0.5% to 1.0% of agar-agar. These gels are rigid, brittle and they have well defined shapes as well as sharp melting and gelling points[3-6].

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doi: 10.5267/j.ccl.2022.12.002

Agar or agar-agar is a substance resulting from boiling a polysaccharide in red algae, where it is accumulated in the cell walls of agarophyte and serves as the primary structural support for the algae's cell walls. Moreover, Agar is a mixture of two components: the linear polysaccharide agarose and a heterogeneous mixture of smaller molecules called agaropectin[7-8]. It has a distinctive smell, one can easily distinguish agar from the other materials commonly found in a laboratory. Chemically, agar is a polymer made up of subunits of the sugar galactose, and is a component of the cell walls of several species of red algae that are usually harvested in eastern Asia and California. Although agar's chief use is as a culture medium for various microorganisms, particularly for bacteria. In addition, well-known uses include serving as a thickening for soups and sauces, in jellies and ice cream. It finds many applications in cosmetics, for clarifying beverages, and for sizing fabrics[9-10]. It is a known fact that, agar is generally resistant to shear forces; however different agars may have different gel strengths or degrees of stiffness. Chitin has a crystalline structure and it constitutes a network of organized fiber structure conferring rigidity and resistance to organisms.

The principal derivative of chitin is chitosan (poly [β-(1 4) -2-acetamido-2-deoxy-D-glucopyranose]) a copolymer of D-glucosamine (GlcN) (80%) and N-acetyl-D-glucosamine (20%) (GlcNAc) units, which are produced by alkaline deacetylation of chitin. Chitosan (CHI) also occurs naturally in some fungi; however, its occurrence is much less widespread than that of the chitin. In fact, Chitosan is a collective name representing a family of de-N-acetylated chitins deacetylated at different degree levels. These are well distinguished by their insolubility or solubility in dilute aqueous acid solutions. Chitosan properties are dependent on the relative proportions of N-acetyl-D-glucosamine and D-glucosamine residues on the basis of their biodegradability and biological role.

Santosh kumar et.al and Al-Naamani et.al highlighted that, the Chitosan blends/composites with numerous natural antioxidants, antimicrobial components, and nanomaterials have also attracted significant research focus in recent years. The recent advancement in chitosan and chitosan based nanocomposite, their fabrication and applications in food packaging and preservation for shelf life extension of fruits, vegetables, meat and fish products are reviewed[11-12]. With reference to the above, many applications can expect by moulding chitosan for its blends with Agar-Agar. Hence, the present work is an attempt for a new way to the chemical blending of the chitosan and Agar-agar gel in different proportions. Miscibility of the blends is characterized by thermal studies like DSC and TGA. Further, they are correlated with FT-IR. Their surface morphology of the film is studied by SEM. Such a blended agar /chitosan can show variable high thermal stability, hydrophilicity and swelling behavior etc.

2. Experimental

2.1 Material and Methods

All chemicals used in the experimentation are AR grade and solvents used are distilled completely. The main source of commercial chitosan and agar is from Merck (Mumbai, India). Double distilled water is used for the preparation of required solutions. Chemical homogeneous mixing is used for the preparation of CAA blends

2.2 Synthesis of Chitosan /Agar-agar blends

Prepared stock solutions of Chitosan and Agar-Agar (1% w/v) was prepared by dissolving l gm of chitosan in 100 cm^3 of 0.5% acetic acid and 1 gm of agar -agar dissolved in 100 cm^3 of distilled water. This solution was stirred 40 ^{0}C for 13horse and was filtered. Blends of Chitosan/Agar-agar (CAA) of different composition like100/0, 90/10, 80/20, 70/30, 60/40, 50/50, 40/60, 30/70, 20/80, 10/90, 0/100) were prepared from the stock solution by mixing these polymers with suitable proportions and stirred for 30 minutes under the influence of magnetic stirrer. Agar is used in a final concentration of 2% for solidifying culture media. However, 0.45% is used in media for motility studies (0.5% w/v) and for growth of anaerobes (0.1%) and microaerophiles. Further, specifications for bacteriological grade agar include good clarity, controlled gelation temperature, controlled melting temperature, good diffusion characteristics, absence of toxic bacterial inhibitors and relative absence of metabolically useful minerals.

2.6 Characterization

Morphology and bonding of the blends was studied by Philips XL 30 FESEM and Perkin–Elmer 1600 spectrophotometer in KBr medium tools respectively. Thermal behaviors of the blend samples were studied by thermo gravimetric analysis (TGA) of the sample and were recorded using NETZSCH STA 409PC tool. Heating rate of 10^0C/min under Argon atmosphere is maintained to obtain TGA data of the sample. Metllor Toledo star instrument was used for the thermal characterization.

3. Results and Discussion

3.1 DSC Studies

The thermal behavior of CAA and their blend compositions (100/0, 90/10, 80/20, 70/30, 60/40, 50/50, 40/60, 30/70, 20/80, 10/90, 0/100) were studied by means of DSC tool and the corresponding thermo grams are given below from **Fig. 1,**

Fig. 2 and **Fig. 3,** respectively. The composition of these three figures indicates that, different composed blends showing similar glass transition temperature (Tg) between the pure chitosan and agar-agar which indicates the partial miscibility of the blends. The glass transition temperature was taken as the mid-point of the change of slope in the DSC curves[13]. The observed experimental T_g values for all the composed blends are in good agreement with that of theoretical values which indicates an intermolecular interaction between the polymers[14-15].

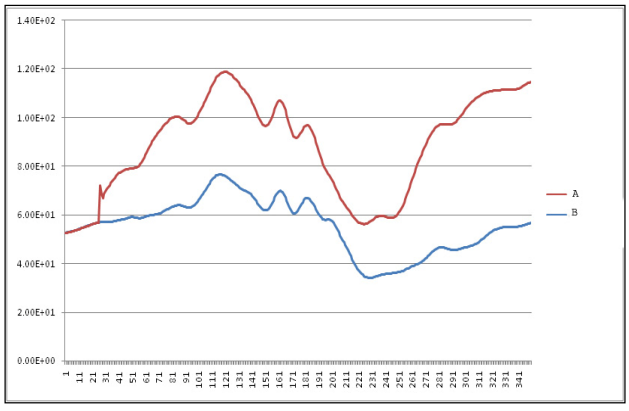

Fig. 1. DSC traces of (A) pure CHI and (B) Agar-agar

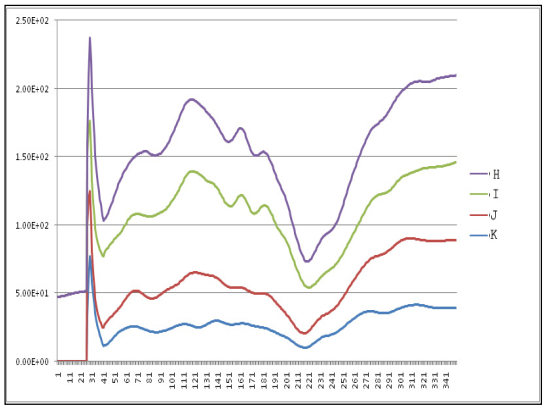

Fig. 2. DSC traces of (H) 10/90 CHI/Agar-agar blend, (I) 20/80 CHI/Agar-agar blend, (J) 40/60 CHI/Agar-agar blend and (K) 50/50 CHI/Agar-agar blend

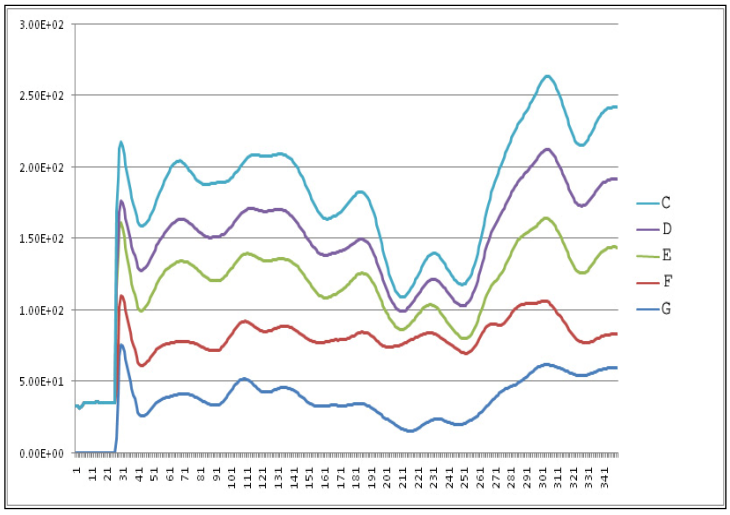

Fig. 3. DSC traces of (C) 60/40 CHI/Agar-agar blend, (D) 70/30 CHI/Agar-agar blend, (E) 75/25 CHI/Agar-agar blend (F) 80/20 CHI/Agar-agar blend and (G) 90/10 CHI/Agar-agar blend

3.2 FT-IR Spectroscopic study

FT-IR spectra of pure chitosan, Agar-agar and their blend films (100/0, 90/10, 80/20, 70/30, 60/40, 50/50, 40/60, 30/70, 20/80, 10/90, 0/100) were recorded and are given in **Fig. 4, Fig. 5** and **Fig. 6**, respectively. It is observed from figures that various vibrations are viewed at 2000-500cm^{-1} range. The strong absorption at 1554.67 cm^{-1} is due to N-O asymmetric stretch and the other strong absorption at 1375.03 cm^{-1} indicates the chitosan has the C-H bending[16]. The strong absorptions at 1243.16 cm^{-1} and 1072.49 cm^{-1} indicate the C-N stretch. Similarly, the absorption at 995.62 cm^{-1} is due to =C-H bond. Consequently, the FTIR study proved that the pure chitosan has N-O asymmetric stretch, C-H bending, C-N stretch and =C-H in its structure[17-19]. On the other hand, Agar-agar spectra at 1645.59 cm^{-1} is due to the stretching of conjugated peptide bonds formed by amine (NH) and acetone (CO) groups[20]. The peak at 1419.62 cm^{-1} indicates C-H stretching and the peak at 1378.90 is due to C-C stretch. The peaks at 1161.65 cm^{-1}, 1040.07 cm^{-1} and 930.79 cm^{-1} are indicated as the 3, 6 anhydrogalactone bridges[21]. The characterized spectrum of CAA blend films in the presence of absorption bands typical of the pure components, with the intensity roughly proportional to the blending ratio. It is noticed that, depending on the composition, the peak attributed to the methoxy group in CAA blended the films. This indicates that some interaction has been developed between them. Likewise, some extra peaks are also noticed in the blended films due to the weak secondary force of attraction. This feature confirmed that some interaction has been formed between chitosan and agar-agar as shown in the **Fig. 5** and **Fig. 6.** Further, it was distinctly observed that all blends are homogeneous suggesting that these blends are partially miscible.

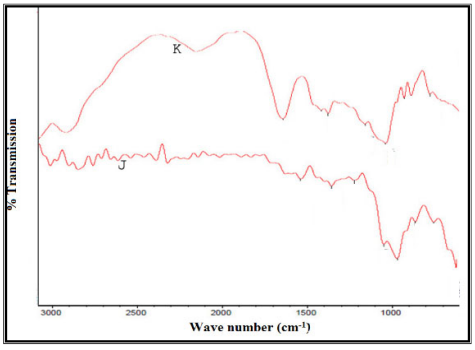

Fig. 4. FTIR spectra of (J) Chitosan and (K) pure Agar-agar

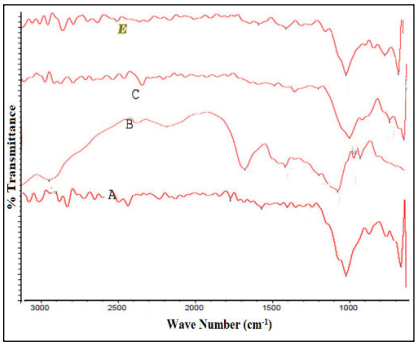

Fig. 5. FTIR spectra of (A) 10/90 CHI/Agar-agar blend (B) 20/80 CHI/Agar-agar blend, (C) 40/60 CHI/Agar-agar blend, and (E) 50/50 CHI/Agar-agar blend

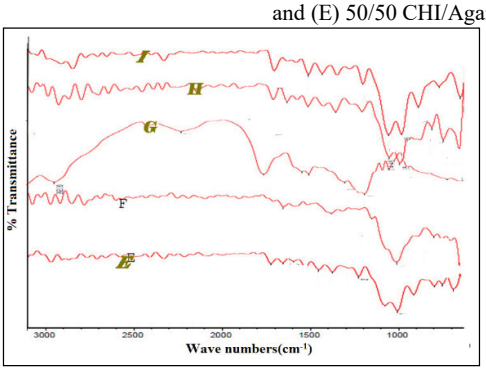

Fig. 6. FTIR spectra of (E) 50/50 CHI/Agar-agar blend,(F) 70/30 CHI/Agar-agar blend, (G) 80/20 CHI/Agar-agar blend , (H) 90/10 and (I) 75/25 CHI/Agar-agar blend

3.3 Morphology Studies by SEM

Figs. (7-10) show SEM images of solution cast films of 20/80, 40/60, 80/20 and 50/50 CHI/Agar-agar nontransparent blends respectively. Almost the images show similar matrix mixed formation of blends. Most of the images show uniform streak morphology with masking particle arrangement. In addition, the fine dispersed visibility with amorphous nature can be viewed. These dispersed images show the partial miscibility of the blends[22].

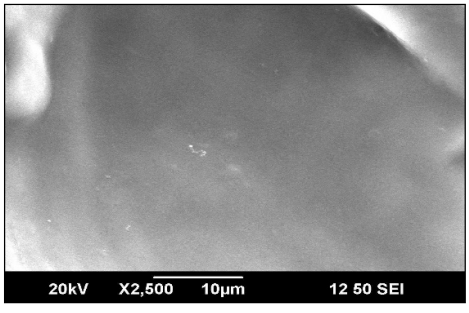

Fig. 7. SEM image of 20/80 CHI/Agar-agar blend

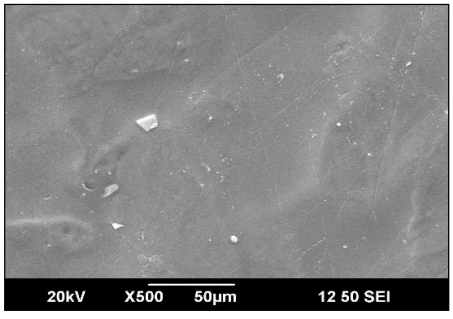

Fig. 8. SEM image of 40/60 CHI/Agar-agar blend

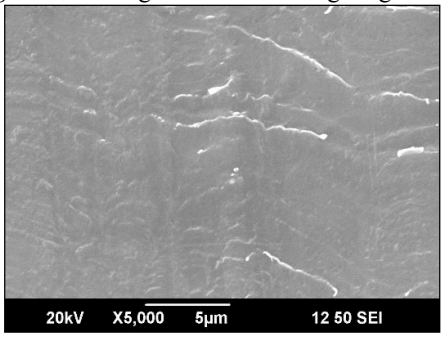

Fig. 9. SEM image of 80/20 CHI/Agar-agar blend

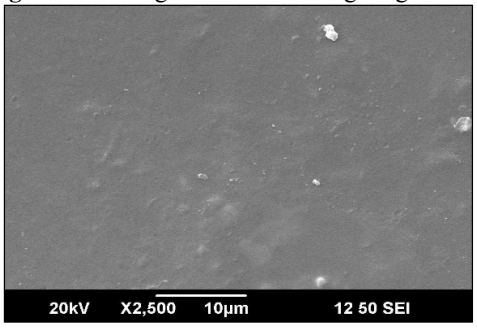

Fig. 10. SEM image of 50/50 CHI/Agar-agar blend

3.4 TGA studies

It is found that TGA studies help in determining the thermal stability of Chitosan and their blend films[23-24]. The TGA curves of homopolymer and blends of Chitosan/agar-agar are given in **Fig. 11, Fig. 12 and Fig. 13**, respectively. In order to determine thermal stability trend, the temperature characteristics such as T_0 (Temperature of onset of decomposition), T_{20} (Temperature for 20% mass loss), T_{30} (Temperature for 30% mass loss), T_{50} (Temperature for 50% mass loss), T_{max} (Temperature for maximum mass loss), ash content and oxidative index (OI) were calculated and are presented in tables 1 and 2. Here, T_0, T_{20}, T_{40}, T_{50}, T_{60} and T_{max} are the main criteria of the thermal stability of blends and signal features of the TGA curves. The OI was calculated based on the weight of carbonaceous char (CR) as in the empirical equation.

$$OI \times 100 = 17.4 \times 0.4\ CR$$

The blend samples were prepared in different compositions as 100/0, 90/10, 80/20, 70/30, 60/40, 50/50, 40/60, 30/70, 20/80, 10/90, 0/100 of Chitosan and thermo gravimetric analysis were performed using thermo gravimetric analyzer under nitrogen environment. The TG curve shows that the Chitosan and Agar-agar films have only one degradation stage. This degradation stage is due to the loss of water in the blend followed by complete weight loss of the sample films. The initial degradation of the homo polymers Chitosan agar-agar and their blends appeared faster and proceeded at a moderate rate in the latter stage. The decomposition temperature of the blends shows that the 10/90 blend possesses the lowest value and the maximum value is observed in the 20/80 Chitosan/Agar-agar blend. **Table 1** and **Table 2** show that when Chitosan content in the blend is in less percentage, the degradation temperature of the blend is almost nearer to the degradation temperature of pure Chitosan. However, the lowest degradation temperatures of 90/10, 80/20 and 70/30 compared to other compositions may be attributed to the partial miscibility of Chitosan/Agar agar at that composition.

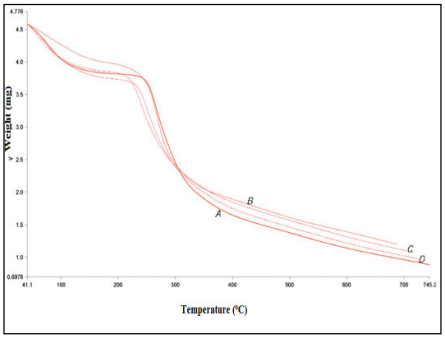

Fig. 11. TGA curves of (A) Agar-agar, (B), Chitosan, (C) 10/90 Chitosan/Agar-agar blend (D) 20/80 Chitosan/Agar-agar blend

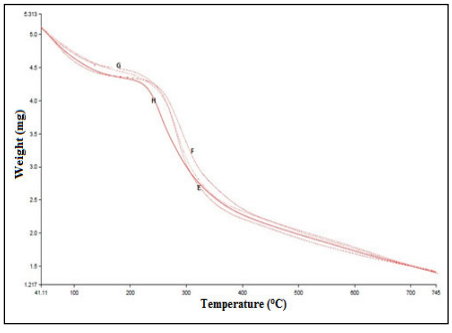

Fig. 12. TGA curves of (E) 40/60 Chitosan/Agar-agar blend, (F) 50/50 Chitosan/Agar-agar blend, (G) 60/40 Chitosan/Agar-agar and (H) 70/30 Chitosan/Agar-agar blend

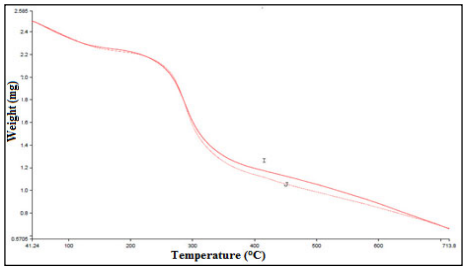

Fig. 13. TGA curves of (I) 80/20 Chitosan/Agar-agar blends, (J) 90/10 Chitosan/Agar-agar blend.

Table 1. Thermal data obtained from TGA scans for CAA blends

Sample Chitosan /Agar-agar	Temperature at which different weight						Ash Content		Oxygen Index(OI)
	T_0	T_{10}	T_{20}	T_{30}	T_{50}	T_{max}	%	mg	
100/0	41.11	140	275	295	380	745.8	31.6	1.7314	0.1205
90/10	41.86	168	270	295	370	700	28.26	1.36	0.094656
80/20	41.24	198	262	288	360	713.8	26.47	0.6624	0.0461
70/30	40.01	142	260	290	375	700	31.7	0.815	0.056724
60/40	40.93	147	260	280	360	734	29.94	1.36	0.094656
50/50	41.06	95	255	293	350	745.6	23.05	0.89114	0.062023
40/60	41.11	110	240	270	347	745	27.362	1.4029	0.09764
20/80	41.2	95	240	260	325	746.2	24.71	1.4515	0.1010244
10/90	40.87	90	240	260	355	745	24.762	1.2921	0.08993
0/100	41.1	95	255	270	310	745.2	19.237	0.8832	0.06147

Table 2
Temperature range of thermal degradation of Chitosan/Agar-agar blends from derivative curve of TGA

CAA Sample	Transition range (^{0}C)		
	T_i	T_P	T_C
100/0	211	289.19	418
90/10	214	290.09	357
80/20	223	287.90	362
70/30	200	287.36	413
60/40	200	285.08	400
50/50	190	280.80	400
40/60	211	291.74	438
30/70	212	252.04	324
20/80	208	247.09	334
10/90	205	247.67	339
0/100	224	265.83	346

(Ti—Temperature at which decomposition starts, Tp—Temperature at which decomposition ratio is max, Tc —Temperature at which decomposition is complete)

4. Conclusions

The blend compositions show single glass transition temperature between Chitosan and agar-agar due to miscibility. Interaction between Chitosan and Agar-agar besides partial miscibility was confirmed by bonding study. TGA studies have also shown the lowest degradation temperatures of 90/10, 80/20 and 70/30 and these blends might be attributed to the partial miscibility of Chitosan/Agar agar at that composition. Morphological studies emphasizing that the blends are homogeneous and agar-agar was well distributed in the Chitosan matrix. In addition to this, a few aggregated particles suggest the partial miscibility of Chitosan and Agar-agar blends. The same method may be applied for the synthesis of different Chitosan.

5. Funding

This work is not financially supported by any funding agency

6. Conflicts of Interest

The authors declare no conflict of interest.

Acknowledgement

Authors thank DST-FIST (SR/FST/CSI-003/2016) grant provided for instruments and infrastructure facilities at Department of studies in Chemistry, Vijayanagara Sri Krishnadevaraya University, Ballari. Thanks to Prof. A.Venkataraman, Professor, Department of Chemistry, Gulbarga University, Kalaburagi, Karnataka, India for useful discussion in spectral analysis.

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