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Preparation and Characterization of NanohybridCompound from Tannic Acid and Zinc Oxide

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Abstract

A nanohybrid tannic acid was prepared by intercalation bulk tannic acid (Tan) into zinc oxide layers. The prepared compound was given the symbol Tan-ZnO and subjected to identification FT-IR spectra showed the appearance of some functional groups and had a shift towards higher and lower frequencies indicating the preparation of new compound. X-Ray Diffraction (XRD) results supported the results obtained from FT-IR and showed the appearance of two new planes in the spectrum at (9.99° and 21.15°)2θ with crystalline distance of (0.88 and 0.42) nm, respectively. By using Atomic force microscope, results showed that (54.44%) of the total particles were nanoparticles with diameter range (30 – 100) nm. The loading efficiency of Tan on Zinc oxide was around 27.68%. Ferric reducing antioxidant power (FRAP) method was used to evaluate the antioxidant activity of the prepared compound.

Keywords: Tannic acid ,Zinc oxide, FT-IR,XRD,Antioxidant activity.

Introduction

Tannic acid (Penta-m-digalloyl glucose) has a molecular formula $C_{76}H_{52}O_{46}$ and belongs to gallotannins group which represents an important class in hydrolysable tannins. This organic acid is widely distributed in the plant kingdom and found in leaves and bark of many plant species [1].It is a water soluble tannin and consist of glucose molecule which occupies the central core position attached with five chains formed by two esterified gallic acid molecules [2].

Tannic acid received more attention in the last decades due to its wide applications, such as antimutagenic, anticarcinogenic, antioxidant, antibacterial, antienzymatic and astringent[3,4].Duemtomthemrapidmdevelopmentmofmnanotechnology,nanomaterials have emerged and have attracted lots of attention because of their novel properties including a large specific surface area and high reaction activity[5,6]Nanocomposites represent hybrid materials possess different phases in which one phase at least withmthemnanoscale.Themlayeredmnanocompositesare formed through the intercalation of guest anion into the interior part of inorganic layer without any change in the layer structure [7].Zinc oxide was used successfully as a nanoparticles because it is non-toxic, has good biocompatibility to human cells as well as it is listed as safe material by FDA [8,9].The current study aimed to prepare and characterize a nanohybrid antioxidant from tannic acid and evaluation its antioxidant activity.

Materials And Methods

preparation of nanohybrid tannic acid

Nanohybrid tannic acid was prepared according to the method described by [10]with some modifications. Briefly, (1.2) (1.2gm) of bulk tannic acid was dissolved in (50) ml deionized water, (1 gm) of zinc oxide was dissolved in (50ml) of (50%) ethanol. The tannic acid solution was dropped within two hours on the zinc oxide solution under stirring. The mixture was placed in a shaker incubator at (37 C°) for (18) hours and then incubated in a static incubator and left for aging at (40 C°) for (24) hours. The mixture was centrifuged, washed four times with deionized water, dried at (40 C°) and stored for further uses. The prepared nanocompound was given the symbol Tan-ZnO while bulk tannic acid was given Tan.

Characterization OfNanohybrid Tannic acid

FT-IR

Infrared spectra of Tan-ZnO, Tan and zinc oxide were recorded in the region (400 – 4000) cm^{-1}. Samples were grinded well and mixed with potassium bromide (KBr) and then pressed into thin pellet.

X-Ray diffraction (XRD) spectrum

In order to measure the difference in the thickness of layer before and after intercalation process for tannic acid according to Brack's low ($n\lambda = 2d\sin\theta$), XRD spectrum was recorded in the range (2 – 50) 2θ.

Atomic force microscope (AFM)

Tan – ZnO was prepared and characterized by AFM to measure the diameters, sizes and aggregation of the nanoparticles.

Loading efficiency of Tan-ZnO

The loaded Tan on zinc oxide was determined as total phenolic content according to the Folin – Ciocalteu method described by [11] with slight modification using Tan-ZnO concentrations in the range (10 – 100) µg/ml. 1 ml from each concentration was diluted with (7.5ml) deionized water and mixed well with ((0.5ml) of Folin-Ciocalteu reagent (1:1) and (1ml) of (7%) Na_2CO_3. After (30) min incubation at room temperature, absorbance was measured at ((600 nm)) using spectrophotometer. Tan in the same concentrations range was subjected to the same reaction and conditions for comparison.

Invitro antioxidant activity of Tan-ZnO

Ferric reducing antioxidant power (FRAP) was used to evaluate the antioxidant activity of the Tan-ZnO and Tan according to the method described by [12,13]. 10 mM of 2, 4, 6, --- Tripyridyl –s-Triazine (TPTZ) was prepared by dissolving in (40) mMHCI., Ferric chloride ($FeCl_3$) was prepared in deionized water (20 mM), while sodium acetate buffer (PH 3.6) was used in (0.3 M), The three solutions were mixed well in the ratio (1:1:10) to prepare FRAP working solution (3.6ml) from FRAP solution were added to (200) µl of different concentrations from Tan-ZnO and Tan, separately, mixed well and left at room temperature for (4) min. Absorbance was measured using spectrophotometer at (593) nm. $FeSO_4.7H_2O$ was chosen to prepare standard curve.

Results And Disscussion

Characterization of nanohybrid tannic acid(Tan_ZnO)

FT-IR:

FT-IR analysis was performed to identify the functional groups in the bulk tannic acid (Tan),Tan-ZnO and Zinc oxide. Fig.1 presents the FT-IR spectrum of Tan. The appearance of the broad band at (3354) cm^{-1} was attributed to the stretching vibration of phenolic hydroxyl group (OH) which seem to be interfered with aromatic CH stretching. The band at (1705) cm^{-1}indicates the presence of ester carbonyl (C═O) group. The spectrum included the appearance of two bands at frequencies (1612 and 1537) cm^{-1}which due to the skeletal stretching of benzene ring (C┅C), while phenolic stretching of (C─O) can be observed (1205)cm^{-1}. Finally, out of plane bending of aromatic CH can be seen at frequencies (869 and 759) cm^{-1}[14].

FT-IR spectrum of zinc oxide layers can be observed in Fig. 2. Many weak bands were observed in the region (400-500) cm^{-1} which can be attributed to the metal bond vibration (Zn-O) [15] . Fig. 3 shows the FT-IR spectrum of Tan-ZnO which confirm the preparation of new hybrid compound. The appearance of the band at (3410) cm^{-1} refers to stretch of phenolic hydroxyl group (O-H) and had a shift towards a higher frequency. The band appearance at (1637) cm^{-1} is due to stretch of ester carbonyl group (C═O) and has a shift to lower frequency. Two bands were appeared at frequencies (1575 and 1487) cm^{-1} which refer to the skeletal stretching of benzene ring and had a shift to lower frequency. Phenolic stretching of (C─O) can be observed at (1207) cm^{-1}, while bending of aromatic CH bonds were observed at (829 and 754) cm^{-1} and had shift to lower frequency [14].

XRD

XRD spectra were performed to confirm the crystalline nature of the prepared nanohybrid tannic acid (Tan-ZnO). This analysis was very useful to determine the differences in the thickness of layer before and after intercalation between zinc oxide layers according to Brack's law.

XRD spectrum of zinc oxide layers was illustrated in Fig. 4. Three diffraction planes were observed included the first plane (100) which appeared at the angle (31^o 2θ) with crystalline distance of (0.281) nm, the second plane (002) which appeared at the angle (34^o 2θ) with crystalline distance of (0.259nm), while the appearance of the third plane (101) was appeared at the angle (36^o 2θ) with crystalline distance of (0.247nm).

As seen in Fig. 5, two new diffraction planes were appeared in the XRD spectrum of Tan-ZnO that confirm the intercalation of Tan into Zinc oxide layers. The first plane was appeared at (9.99^o2θ) with crystalline distance of (0.88 nm) while the other was appeared at (21.15^o 2θ) with crystalline distance of (0.42nm).

AFM

AFM was useful tool to measure diameters, sizes and aggregation of nanomaterial. It is nondestructive for the sample and provide good three dimensional spatial resolution.

Two dimensional image of Tan-ZnO can be seen from Fig. 6a. the particles are aggregated in a spherical forms. Fig. 6b illustrates the three dimensional image of a section of the nanohybrid tannic acid where the elevation of particulate assemblies of up to (0.44 nm). This result confirm the formation of a nanohybrid compound from Tan and Zinc oxide layers and supported the results of FT-IR and XRD for the nanohybrid compound under study.

Results from table 1 revealed that (54.44%) of the total particles were nanoparticles with diameter range (30 – 100 nm). The lowest and highest percentage of nanoparticles were (1.11 and 11.11 %) for particles with diameters (30 and 80 nm), respectively. The results also showed that the means of particle size was (95.16) nm. This result was in agreement with those found by [16] where the means of nanohybrid Amphotericin B (loaded in layered double hydroxides) was (96.18 nm). Similar results were reported by [17]where the means of the nanohybrid levofloxacin was ((94.83 nm).

Loading efficiency of Tannic acid on Zinc oxide

By observing table 2 the loading efficiency of tannic acid on zinc oxide were (32.3, 27.8, 29.6, 26.1, 25.0, and 25.3 %) for the concentration (10, 30, 50, 70, 90, and 100) µg/ml, respectively with an average of (27.68%).

The results obtained from this study were less than those reported by [18]where the loading efficiency of tannic acid on layered double hydroxides reached (33.08%).

Tannic acid reacts with Folin-ciocalteu reagent to form a blue chromophore (phosphotungstic-phosphomolybdenum complex) where the maximum absorption depends on tannic acid concentration [19,20]

Antioxidant activity of Tan-ZnO by FRAP method

The reducing power exhibited by Tan-ZnO and Tan was measured using $FeSO_4.7H_2O$ as a standard. Results in Fig. 7 and 8 revealed an increasing in the reducing power of the compounds understudy with the increasing in concentrations indicating that Tan-ZnO and Tan have the ability to donate hydrogen atom to break free radical chains. Results also illustrated that Tan exhibited higher reducing power than Tan-ZnO.

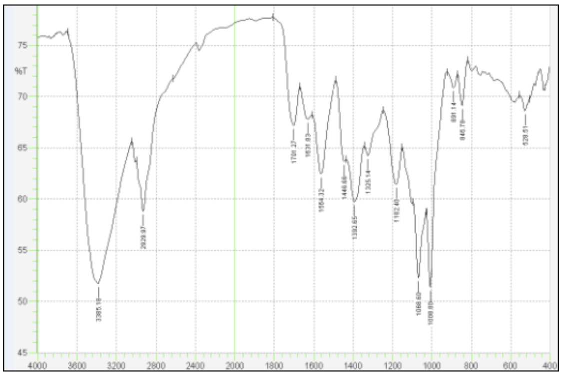
Our results supported those found by [18]where the reducing power of Tannic acid was higher than that exhibited by the nanohybrid tannic acid loaded on layered double hydroxides.In comparison with other methods, FRAP is considered as a simple, fast and not expensive method as well as there was no need to specific tools and can be achieved by manual and mechanical methods[21].

Table(1):Diameters ,volume and accumulationof Tan –ZnO from atomic force microscope

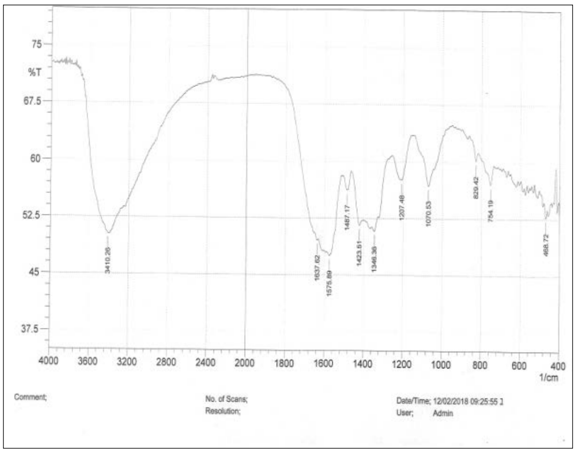
Diameter(nm)<	Volume(%)	Accumulation(%)	Diameter(nm)<	Volume(%)	Accumulation(%)	Diameter(nm)<	Volume(%)	Accumulation(%)
30.00	1.11	1.11	80.00	11.11	39.44	130.00	6.11	78.33
40.00	2.78	3.89	90.00	9.44	48.89	140.00	7.22	85.56
50.00	7.78	11.67	100.00	5.56	54.44	150.00	8.89	94.44
60.00	7.22	18.89	110.00	9.44	63.89	160.00	2.78	97.22
70.00	9.44	28.33	120.00	8.33	72.22	170.00	2.78	100.00

Table (2):Loading efficiency of Tan on Zinc oxide

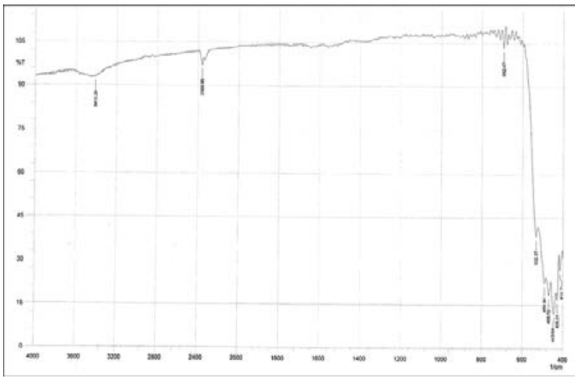
Acid Con. (µg/ml)	A_{600} for Tan-ZnO (A_1)	A_{600} for Tan (A_2)	Loading efficiency (A_1/A_2) x100
10	0.01	0.031	32.3
30	0.032	0.115	27.8
50	0.061	0.206	29.6
70	0.079	0.303	26.1
90	0.100	0.400	25.0
100	0.112	0.442	25.3



Figure(1) :Infrared spectrum for Tannic acid (Tan)



Figure(3): Infrared spectrum for nanohybrid tannic acid (Tan -ZnO)



Figure(2):Infrared spectrum for Zinc oxide

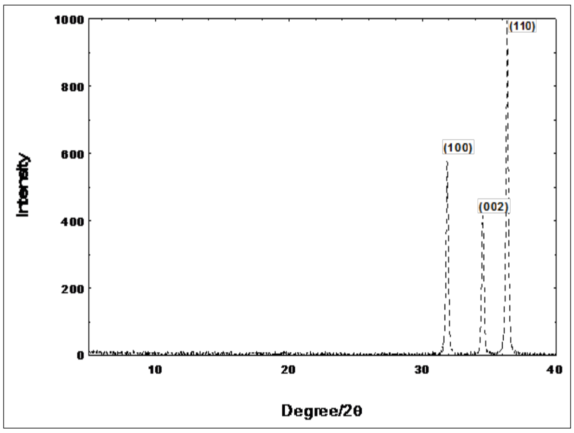
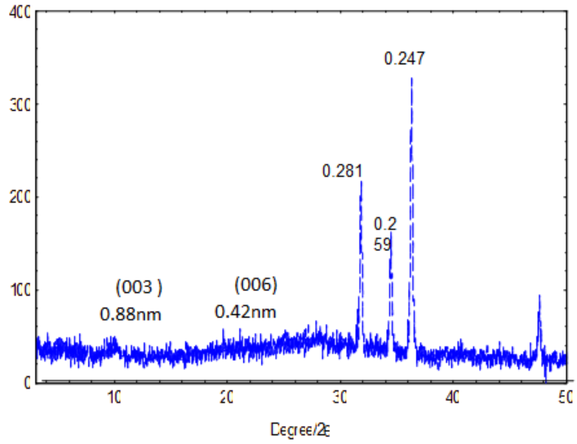
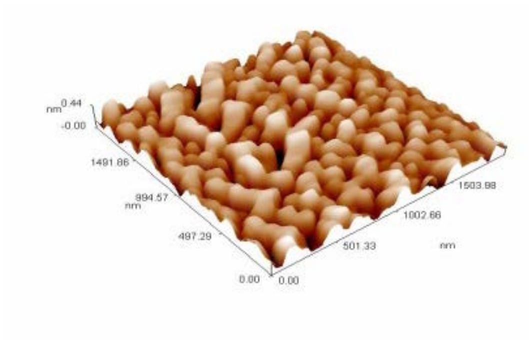


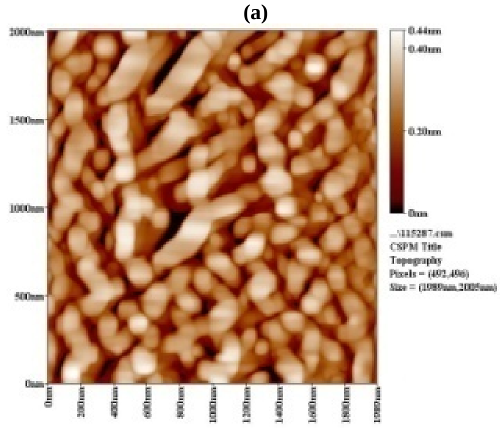
Figure (4): X-ray diffraction spectrum (XRD) for Zinc oxide layers



Figure(5):X-ray diffraction spectrum of nanohybridtannic acid(Tan-ZnO)



(a)



(b)

Figure 6:(a) Two-dimensional, (b) three–dimensional, image of the nanohybrid Tannic acid (Tan-ZnO)

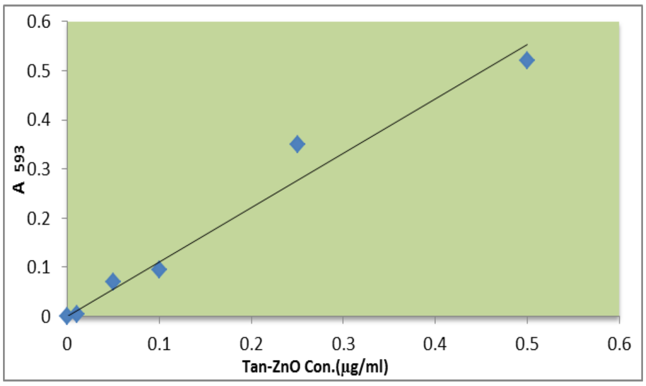
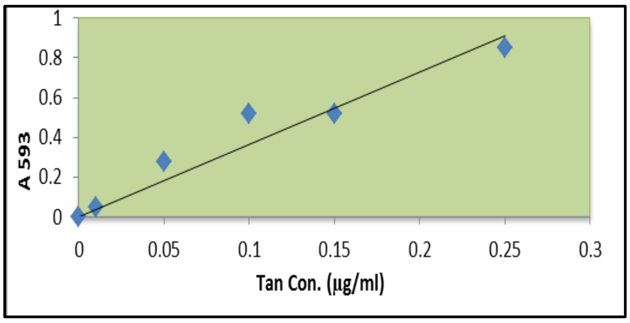


Figure (7):Antioxidant activity of Tan –ZnO by FRAP method



Figure(8):Antioxidant activity of Tan by FRAP method

CONCLUSION

A nanohybrid antioxidant can be prepared by intercalation of tannic acid on zinc oxide layers (Tan-Zno).The prepared compound showed good antioxidant activity by Ferric reducing antioxidant power(FRAP)method.

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