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Anti-Inflammatory and Antibacterial Activities of Nanosilver-Treated Cotton Fabric Prepared from Ethanolic Extracts of Three *Terminalia* Species

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NANOTECHNOLOGY, this ample field of the 21st century, is having an exceptionally critical effect on the world's industry and people's lives. This work aims to synthesis of fast green biogenic silver nanoparticles (AgNPs) using the ethanolic extracts of three *Terminalia* spices, namely, *T. catapa* (*T.C*), *T. bellarica* (*T.B*) and *T. mellurie* (*T.M*). AgNPs formed was affirmed by using UV–visible spectrophotometer with the characteristic SPR (Surface Plasmon Resonance) band at 424-430nm and by using FT-IR (Fourier transform infrared spectroscopy) and TEM (transmission electron microscopy). Quantification of carbohydrates, total phenolic compounds, flavonoids, and protein contents of these extracts was appraised since these constituents have effective dual roles for AgNPs synthesis. That is, they reduce Ag^{+} ions to Ag° nanoparticles and thereafter they stabilize these particles. GC/MS analyses of these extracts were also done. The cotton fabrics treated with the synthesized AgNPs showed good antimicrobial and anti-inflammatory activities toward both Gram-positive (S. *aureus* ATCC 25923) and Gram-negative (E. *coli* ATCC 25922) bacteria.

Keywords: Silver nanoparticles, Plant extracts, *Terminalia species*, Cotton fabrics, Antibacterial, Anti-inflammatory study.

Introduction

Many researchers have widely used noble nanoparticles (NPs) in various technological applications because of their unique properties. The noble metal nanoparticles (NPs), in general, and silver NPs (AgNPs), in particular, are known for their versatile applications in food processing industries [1], in medical industries (as ointments to prevent infection of wounds and burns) [2], in textile industries (Ag impregnated fabrics) [3] and in consumer goods being an effective antimicrobial agent [4,5].

Utilizing plant extracts or plant biomass [6-9] could be an exemplary biological method for the production of nanoparticles in an economical and ecofriendly manner compared to chemical and physical methods. The biologically active compounds participated in the green synthesis of NPs deport as functionalizing ligands, making these NPs most relevant for biomedical applications [10]. In consequence, the advancement of such protocols to synthesize nontoxic metal NPs is presently of great importance and there is thus a claim for biosynthetic or green methods for this purpose. It has been noticed that the plant extract of the genus *Terminalia* is mostly rich in

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a variety of biologically active molecules such as phenolics, flavonoids, alkaloids, triterpenoids, tannins and other compounds [11-17]. Because of these phytoconstituents, the majority of *Terminalia* species have a variety of biological, pharmacological and medicinal activities [18–25].

The main objectives of the present study are: (i) to prepare the aqueous ethanolic extract of the leaves of the three *Terminalia* species, (ii) qualitative and quantitative estimation of their phytoconstituents using standard methods and GC/MS, (iii) to synthesize AgNPs using the prepared extracts, (iv) to characterize AgNPs using UV–Vis spectroscopy, FT-IR and TEM, and (v) to check their anti-inflammatory and antibacterial activities towards both Gram-positive and Gram-negative bacteria.

Experimental

Plant material

Green leaves of *Terminalia catappa* (*T. catappa*), *Terminalia bellerica* (*T. bellerica*), and *Terminalia muelleri* (*T. muelleri*) were collected from the Giza Zoo Garden, Cairo, Egypt, in March 2013. These plant leaves were thoroughly washed in running tap water for about 20-30 minutes till all the foreign material and soil particles removed from the surface and then air dried under shade at room temperature. The dried leaves were finely powdered using an electric grinder and used for aqueous ethanolic extraction.

Extraction

Extraction of the plant air-dried powdered leaves was achieved as follows: 5 g of each plant leave was heated with 100 ml aqueous ethanol (80%) at 70°C for 1 hr. in a thermostatic water bath. Each extract solution was filtered and the filtrate was stored at 4°C.

Chemical analysis of the extracts

Phytochemical screening

Considerable significance of phytochemical screening in a given plant is to provide us with information about the chemicals present in this plant in term of their nature and range of occurrence. Thereof, the impacts of these chemicals in biological assays held to investigate a certain bioactivity of a given plant would be well understood. In this study, preliminary phytochemical screening (colour reaction) was conducted on the aqueous ethanolic leaves extract according to standard methods [26–28].

Total phenolic assay

The concentration of phenolics in each plant leaves aqueous ethanolic extract was measured using the Folin–Ciocalteu method [29] as follows: 0.5 ml of the plant leaves extract solution was mixed with 2.5 ml of freshly prepared Folin–Ciocalteu reagent (0.2 N) for 5 min, followed by the addition of 2 ml of Na_2CO_3 solution (75%, w/v). After incubation at room temperature for 2 hrs, the absorbance of the reaction mixture was measured at 760nm against methanol blank. Calibration curve was constructed using gallic acid standards and the total phenolic content was expressed in g of gallic acid equivalents (gGAE) /g of the plant leaves extract.

Total flavonoid assay

The total flavonoid content (expressed as quercetin equivalent, mg QE/g) in the examined plant extracts was estimated using the modified Quettier–Deleu *et al* method [30] as follows: The leaves extract solution for each plant was admixed with 2% $AlCl_3$ solution in methanol. Absorbance readings at 415nm were taken after 10 min against a blank sample. The latter consisting of a 5 ml leaves extract solution with 5 ml methanol without $AlCl_3$. The total flavonoid content (expressed as mg quercetin equivalents (QE) per g plant leave extract) was estimated using a standard curve with quercetin as a standard.

GC-MS Analysis

Quantitative determination of the leaves aqueous ethanolic extract of the three studied plant species were analysed by GC/MS Capillary column of fused silica (5% phenyl methyl polysiloxane), 30m length, 0.25mm I.D. and 0.25 µm thickness, DB-5, carrier gas helium at 13 psi; oven temperature 50-280°C, chart speed 0.5 cm/min; ion source temperature 220°C; ionization voltage 70 ev; accelerated voltage 2000 v; volume injected 1 µl. The results are listed in Table 3, the identification of the compounds was accomplished by comparing their retention times and mass spectral data with those of the library (Wiley Int. USA), NIST (Nat. Inst. St. Technol., USA) and / or published data (Adams 1995) [31].

Synthesis of AgNPs

The leaves aqueous ethanolic extract prepared from the three *Terminalia* species were used for synthesis of AgNPs according to the following procedure:

10 ml of the prepared extract was added to a definite volume of distilled water, the pH of the solution was adjusted to 11, to this solution, 0.017

g of $AgNO_3$ dissolved in 10 ml of distilled water was added and the total volume was adjusted to 100 ml with distilled water. The reaction mixture was subjected to continuous stirring at 60°C using a magnetic stirrer for different durations (15, 30, 45, 60 and 75 min). The visual change of colour from yellow to reddish brown indicates the formation of silver nanoparticles (AgNPs). The coloured colloidal solution formed was checked by diluting HCl for complete conversion of $AgNO_3$ to AgNPs where no white precipitate is formed.

Characterization of AgNPs

UV–Vis spectroscopy

The initial characterization of the synthesized AgNPs was performed using UV–Vis spectroscopy. The reduction of silver ions was monitored using a scan from 250nm to 500nm by JASCO V-670 UV-Vis-NIR double beam spectrophotometer (after 5-fold diluting the sample with distilled water against distilled water as blank. The measurements were carried out as a function of reaction time at room temperature.

Fourier transform infrared (FTIR) spectroscopy

Binding properties of AgNPs biosynthesized by *T. catappa, T. bellerica and T. muelleri* aqueous ethanolic leaves extracts were investigated by FTIR analysis using a JASCO FT/IR 4100 (JASCO, Tokyo, Japan) instrument in the diffuse reflectance mode at a resolution of 4 cm^{-1} in KBr pellets. Prior to analysis, AgNPs were purified, dried and palleted with potassium bromide. For comparison, three *T.* species aqueous ethanolic leaves extract were freeze dried, palletized to be used as a control.

Transmission electron microscopy (TEM)

The morphology and size of the optimal AgNPs solution were identified by Transmission electron microscopy (TEM) (JEOLJEM-1200, Japan). The sample is placed on the carbon coated copper a grid, making a thin film of a sample on the grid and extra sample was removed using the cone of a blotting paper and kept in a grid box sequentially. The instrument was operated at an acceleration voltage of 200 kV.

Fabrics

Desized, scoured, and bleached 100% cotton fabric, was kindly supplied from El-Mahalla Company for Spinning and Weaving, El- Mahalla El-Kubra, Egypt.

Treatment of cotton fabric

Before being utilized, cotton fabric was washed and dried. Experiments were conducted on samples with maximum dimension of 30cm x 15cm. The dried cotton samples were subjected to four separate treatments. Concerning this, the fabrics were, individually, treated with the aqueous ethanolic extract of the three studied *T.* species (set A), aqueous ethanolic extract of each plant and 1% Binder (printo® FX based on acrylate) solution (set B), and AgNPs colloidal solution prepared from each plant extract at a concentration of 108 ppm and 1% Binder (set C). These treatments were carried out using a pad/dry technique and the treated samples were squeezed to 100% wet pick up at constant pressure, then dried at 70◦C for 3 min, followed by curing at 150◦C for 2 min.

Characterization of the treated cotton fabrics

Antimicrobial activity: The antibacterial activities of the untreated cotton fabric (control), fabric treated with aqueous ethanolic extract of each plant species in the absence and presence of a binder (B) and fabrics treated independently with AgNPs synthesized from different *Terminalia* species in the presence of a binder were quantitatively evaluated by using plate count agar according to the AATCC test 100–1999 [32]. The species of microorganisms used were *E. coli* AATCC 2666 (Gram -ve) and *S. aureus* AATCC 6538 (Gram +ve). These bacteria were singly inoculated into tubes containing 5 ml BHI (Brain Heart Infusion Broth) sterile suspension. Such suspension was adjusted spectrophotometrically according to Koo *et al.* 2000 [33], who used the optimal density at 800nm to match the turbidity of 1.5×108 colony forming unit (CFU) ml^{-1} (equivalent to 0.5 Mc Farland standard). A small volume of the previous microorganisms inoculums (10^{-1}) was transferred to a sealed jar containing 1 g of fabric sample in addition to 50 ml normal saline. The jars were incubated at 37◦C for 24 h. 10^{-1} of the previous suspension were transferred on nutrient and Sabouraud dextrose agar for bacterial count. Antibacterial activity was expressed as the percentage of reduction (R%) as follows:

$$R\ (\%) = A-B/A\times 100$$

where R is the reduction rate in the number of colonies, A is the number of bacterial colonies from untreated fabrics, and B is the number of bacterial colonies from treated fabric (Duran *et al.,* 2007) [34].

Wash durability: The washing durability

method for the antibacterial treated fabric was assessed according to the AATCC test 61–1989 [35]. 1 gm sample was soaked in 40 ml solution containing 2 g/l Egyptol PLM (non-ionic detergent). Washing was conducted for 20 min at 40◦C.

Anti-inflammatory activity: Anti-inflammatory activity was identified according to the method described by Winter *et.al.* 1962 [36]. Sixty male albino rats were divided into ten groups, each of six animals. Animals were anaesthetized by the open mask method with anaesthetic ether and their backs were shaved with electric clippers. First group: blank clothes film was applied to the skin of the back and tied firmly. The second group, the reference Indomethacin cream was applied to the blank tissue on film and tied firmly. Third to tenth groups, were applied respectively. One hour later, all the animals have a supplanted injection of 0.1 of 1% carrageenan solution in saline, in the right hind paw and 0.1 ml saline in the left hind paw. Four hours after application of film the rats were sacrificed. Both hind paws were excised and washed separately. The percentage Oedema was calculated according to the following equation:-

$$\frac{\text{wt of right paw} - \text{wt of left paw}}{\text{wt of left paw}} \times 100$$

Results and Discussion

In this context the present study used three *T.* species aqueous ethanolic leaves extract to biosynthesize AgNPs which is rich in polyphenolic compounds, flavonoids and tannins [37], On addition of $AgNO_3$ aqueous solution to the extract the appearance of reddish brown colour appeared within 15 min which was quite faster than the previous bio-based reports [38–41]. Rapid formation of AgNPs was due to higher availability of phytoconstituents and hence higher reduction potential of *T.* species ethanolic extracts. Moreover, these constituents not only reduced Ag+ ions to AgNPs, but acted as capping/ stabilising agents.

The phytochemical preliminary results

Plant ethanolic extracts were characterized by preliminary phytochemical tests for rough ideas of constituents present in the extract. The results obtained showed the presence of some phytochemicals in the three *Terminalia species* under investigation. These embrace carbohydrates, terpenoids, steroids, flavenoids, phenolics, tannins, fats/oils and proteins (Table 1). The quantitative analysis of these constituents (Table 2) revealed that the highest percent yield of the polysaccharides (PS) is present in *T. bellerica*. Data from this table showed also that the percentages obtained from the total protein content is 16, 13.25 and 12.5% for *T. bellerica, T. catappa* and *T. muellere*, respectively. The highest phenolic contents (Table 2) were observed in *T. muelleri* (69.15mg/g GAE), followed by *T. catappa* and *T. bellerica* (46.52 , 37.17 mg/g GAE), respectively. Table 2 shows also that the highest flavonoidal contents were observed in *T. catappa* (70.52 mg/g QE) followed by *T. muelleri* and *T. bellerica* (45.10, 21.29 mg/g QE).

As reported elsewhere [42], these phytoconstituents act as good reductants and stabilizers for Ag ions and AgNPs, respectively.

GC/MS analysis of the crude ethanol extracts (70%) (Table 3) revealed the identification of twenty compounds representing 81.19% of the total peak area of the *T. catappa* extract {the major compounds were germanicol (16.82%), lupeyl acetate (13%) and butylated hydroxytoluene (6.84%)}, fourteen compounds constituting 86.44% of the total peak area of the *T. bellerica* extract {the major compounds were phytol (42.13%), and butylated hydroxytoluene (16.96%)}, and nine compounds representing 72.73% of the total peak area of the *T. muelleri* extract {the major compounds were butylated hydroxytoluene 60.9% and neophytadiene 3.85%}. These major compounds (Fig. 1) have enough capacity to reduce silver ions to silver nanoparticles.

Characterization of Silver Nanoparticles

UV-Vis spectroscopy analysis

Due to excitation of surface plasmon resonance (SPR) in AgNPs, its aqueous suspension appeared as a reddish brown colour. The reduction of Ag^+ ions into AgNPs was monitored using UV–vis spectroscopy. Figure 2 shows the UV–vis spectrum at different time intervals (15-60 min) which confirms the formation of AgNPs. It was observed that a broad plasma resonance appeared at λmax 424-430nm. The intensity of the SPR and absorbance increases from 250nm to 600nm as the reaction time increased up to 60 min, irrespective of the *Terminalia species* used.

The stability of the AgNPs was studied for prolonged periods (3-6 months). The results showed similar monodispersity and chemical stability of the nanoparticles which are well dispersed in the solution without any aggregation.

TABLE 1. Phytochemical constituents detected in 80 % ethanolic leaves extract of *Terminalia* species.

Phytochemical constituents	*T. catappa*	*T. bellerica*	*T. muelleri*
Carbohydrates	+	+	+
Fats/Oils	+	+	+
Saponins	-	-	-
Terpenoids	+	+	+
Steroids	+	+	+
Flavenoids	+	+	+
Phenolics/Tannins	+	+	+
Cardiac glycosids	-	-	-
Proteins/Amino acids	+	+	+
Alkaloids	+	+	+
Anthraquinones	-	-	-

+ = Present ; - = Absent

TABLE 2. Estimation of the various contents of the 80% ethanolic leaves extract of *Terminalia* species.

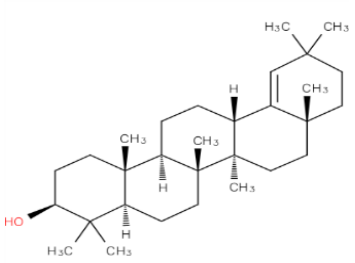
T. species	%Yield of 80 % ethanolic extract	Total phenolic content[b] (mg/g) GAE	Total flavonoid content[c] (mg/g) QE	% of Protein content[a]	% of Total Carbohydrate content[a]
T. catappa	25.2 ± 1.5	46.52 ± 3.2	70.52 ± 3.4	13.20 ± 2.4	13.04
T. bellerica	21.7 ± 1.1	37.17 ± 2.1	21.29 ± 1.4	16.00 ± 1.8	15.57
T. muelleri	24.3 ± 1.8	69.15 ± 4.3	45.10 ± 2.2	12.50 ± 1.4	13.07

The data are expressed as mean ± SD for three replicates[a] % w/w, [b] mg/g extract Gallic acid, [c] mg/g Quarcitine.

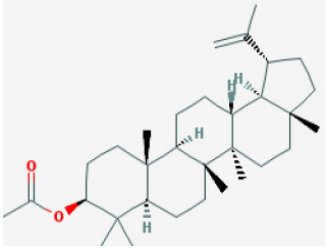
TABLE 3. GC/MS analysis of the 80% ethanolic leaves extract of *Terminalia* species.

RT	M.W	M.F.	Area %			Compound name	B.P.	Main fragments
			T. c	*T.B*	*T. M*			
13.87	170	$C_{12}H_{26}$	---	0.67	1.99	Dodecane	71	43, 85, 99, 127
21.97	190	$C_8H_{40}O_5$	0.65	---	---	Diethyl maleate	117	71, 89, 118, 145
26.51	220	$C_{15}H_{24}O$	6.84	16.96	60.90	Butylated hydroxyl toluene	205	57, 67, 81, 105
32.76	256	$C_{16}H_{32}O_2$	2.77	0.25	---	Tetradecanoic acid ethyl ester	88	101,157, 239, 241
33.68	278	$C_{20}H_{38}$	1.30	0.40	3.85	Neophytadiene	68	57, 82, 95, 123
36.75	284	$C_{18}H_{36}O_2$	6.43	7.27	1.49	Hexadecanoic acid ethyl ester	88	43, 55, 101, 57
38.65	294	$C_{19}H_{34}O_2$	---	---	1.27	8,11-Octadecadienoic acid methyl ester	67	55, 83, 95, 109
38.75	296	$C_{19}H_{36}O_2$	0.45	0.60	1.39	10-Octadecenoic acid, methyl ester	55	69, 83, 97, 264
39.00	296	$C_{20}H_{40}O$	4.64	42.13	---	Phytol	71	43, 81, 83, 95, 123
39.22	298	$C_{19}H_{38}O_2$	---	---	0.57	Octadecanoic acid methyl ester	74	43, 87, 143, 255
39.85	308	$C_{20}H_{36}O_2$	2.46	5.93	0.75	Ethyl linoleate	67	81, 95, 109, 263
39.98	306	$C_{20}H_{34}O_2$	2.90	8.11	---	Ethyl octadecatrienoate	79	67, 95, 108, 121,
40.41	312	$C_{20}H_{40}O_2$	2.79	0.45	---	Octadecanoic acid ethyl ester	88	43, 55, 69, 101,157
41.55	294	$C_{20}H_{38}O$	0.82	---	---	1-Hexadecyn-3-ol-tetramethyl	69	43, 57, 84, 121
42.20	330	$C_{19}H_{38}O_4$	---	0.30	0.52	2,3-Dihydroxypropyl hexadecanoate	55	43, 57, 98, 129,256
42.39	312	$C_{20}H_{40}O_2$	0.45	---	---	Octadecnoic acid ethyl ester	88	43, 55, 69, 101
43.84	366	$C_{22}H_{45}$	0.60	---	---	Docosane	43	57, 71, 85, 99,113
45.73	408	$C_{29}H_{60}$	2.07	0.33	---	Nonacosane	43	57, 71, 85, 99
46.24	390	$C_{24}H_{38}O_4$	1.12	1.29	---	Diisooctyl phethalate	149	57, 70, 83, 167,279
46.45	426	$C_{30}H_{50}O$	5.00	---	---	À-Amyrin	218	44, 55, 189, 203
48.17	410	$C_{30}H_{60}$	---	1.75	---	Squalene	69	81, 85, 137, 191
48.30	426	$C_{30}H_{50}O$	16.82	---	---	Germanicol	204	131, 177, 189, 218
49.84	426	$C_{30}H_{50}O$	5.56	---	---	Lupeol	43	68, 81, 95, 121,218
50.30	426	$C_{30}H_{50}O$	4.52	---	---	Taraxsterol	207	189, 218, 315, 357
53.39	468	$C_{32}H_{52}O_2$	13.00	---	---	Lupeyl acetate	43	95, 109, 189, 218.
Total identified area			81.19	86.44	72.73	---	---	---

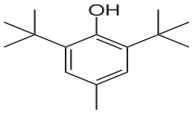
T=Retention time, M.W=Molecular weight, M.F=Molecular formula,
T.C=Terminalia catapa, *T.B=Terminalia bellerica* , *T.M=Terminalia mellueri* , B.P=Base beak



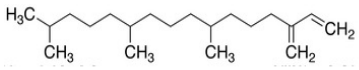
Germanicol



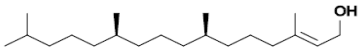
Lupeyl acetate



Butylated hydroxyl



Neophytadiene



Phytol

Fig. 1. The major identified compounds in the three *Terminalia* species.

FTIR analysis

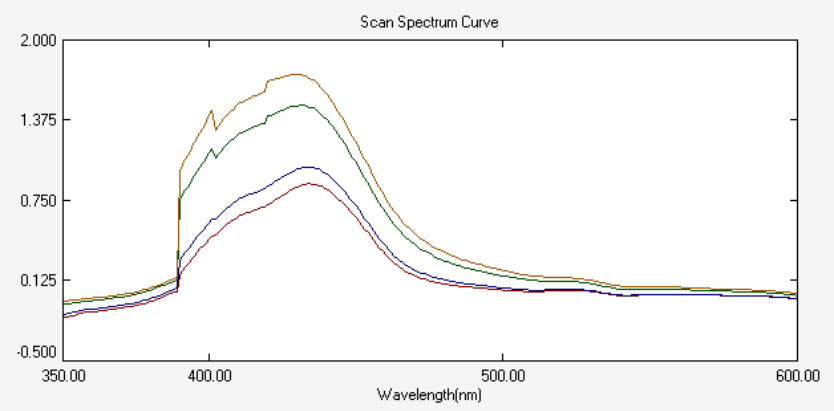
To identify the possible biomolecules responsible for efficient stabilization of the silver nanoparticles, the plant extract and the synthesized nanoparticle were subjected to FTIR spectral studies. Comparative FTIR profiles of ethanolic extracts of *T. bellerica, T. catappa* and *T. muellere* and AgNPs prepared from each, are depicted in Fig. 3a,3b and 3c, respectively. The spectra revealed the presence of prominent peaks at (3462-3472), (2965-2968), (1640-1646), (1460-1465) and (1073-1085) cm−1 corresponding to different functional groups, namely, NH, OH, C≡N, N-O, C=C, C-N, and C=CH_2. These indicate that the polyphenols, protein and polysaccharides present in the aqueous ethanolic extract of the three species were responsible for reduction of Ag^+ to Ag^0 and stabilization of the biosynthesized AgNPs. Hence FTIR study reveals the multi-functionality of the aqueous ethanolic extract of *T.* species. .

Transmission electron microscopy (TEM)

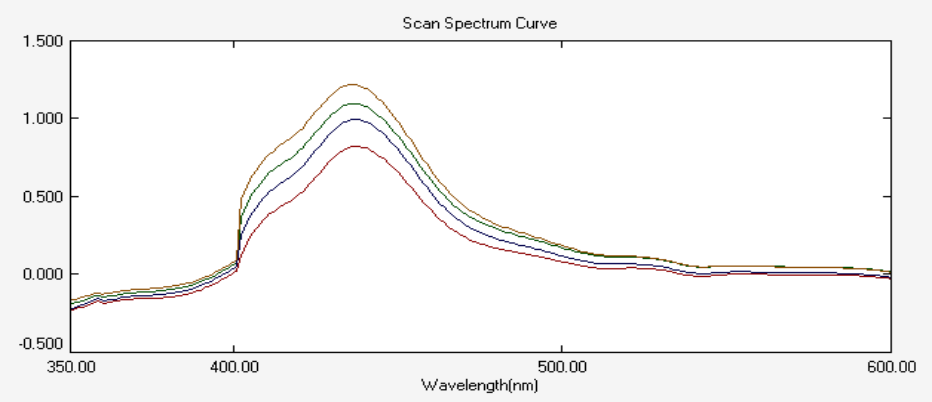
The morphology of the AgNPs was also assessed by TEM**.** The TEM images of AgNPs are shown in Fig. 4a-f. From these images, it is clear that the morphology of AgNPs is polydispersed (Fig.4a-c) and mostly spherical with diameters in the range of 10, 12 and 14nm for AgNPs biosynthesized from *T. bellerica*, *T. muelleri* and *T. catappa*, respectively (Fig. 4d-f). This trend coincides with the findings of the GC/MS and the quantitative analysis of the three plant constituents. That is, because *T. bellerica* contains the highest percentages of phytol, butylated hydroxyl toluene, protein and total carbohydrate, its ethanolic extract showed the highest capacity for reducing and stabilizing AgNPs as compared to the other two plant species under investigation.

Antibacterial activity of the AgNPs-treated cotton fabrics

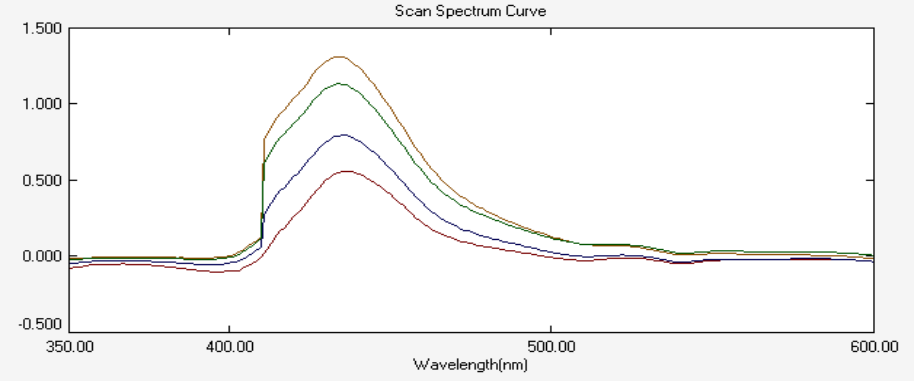
Table 4 shows antibacterial properties (bacterial reduction) of the different treatment sets of the cotton fabrics, as mentioned earlier in the experimental section. Evidently, reduction of the bacterial colonies against either *S. aureus* or *E. coli* was, in most instances, higher than 90% for sets A & B treated samples and reached to 100% in the case of set C treated samples. This holds true before the washing of the treated fabric samples. However, subjecting the treated fabrics for 5 and 10 washing cycles lead to different situations in the reduction of bacterial colonies depending on the treatment conditions as depicted in Table 5. The order of the bacterial colonies reduction follows the order: set A< set B< set C. This confirms that the fabrics treated with a solution containing nano-sized silver particles in the presence of the binder have excellent antibacterial properties as evident before [43,44]. The antibacterial activity may be attributed to the AgNPs itself and/or its surrounding coat which is formed from the major phytoconstituents present in the aqueous ethanolic extract.



(a)



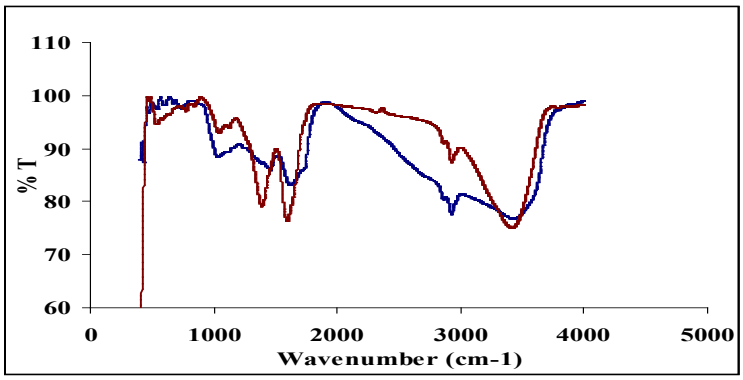
(b)



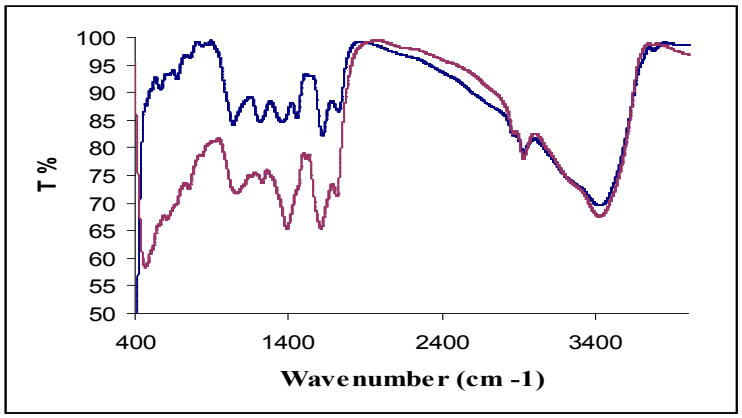
(c)

Fig. 2. UV-Vis absorption spectra of AgNPs synthesized at different time intervals from 80% ethanolic extracts of (a) *T. catapa,* (b) *T. bellerica* and (c) *T. muelleri*

(a) FTIR of *T. B*



(b) FTIR of *T. C*



(c) FTIR of *T.M*

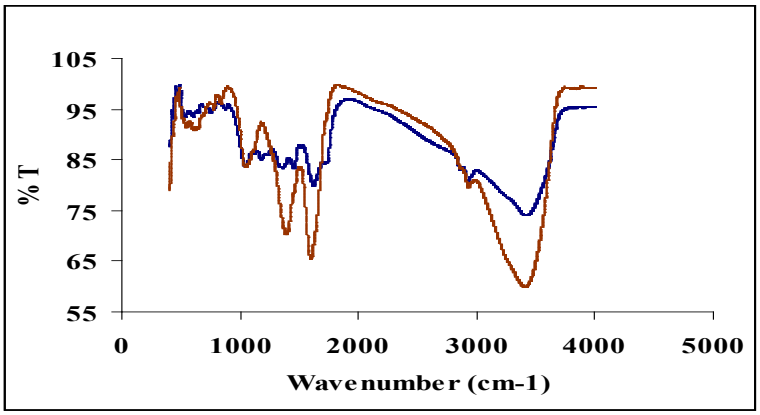


Fig. 3. FTIR Spectrum (transmittance mode) of the three *Terminalia* species extract (red) and the biosynthesized AgNPs (blue).

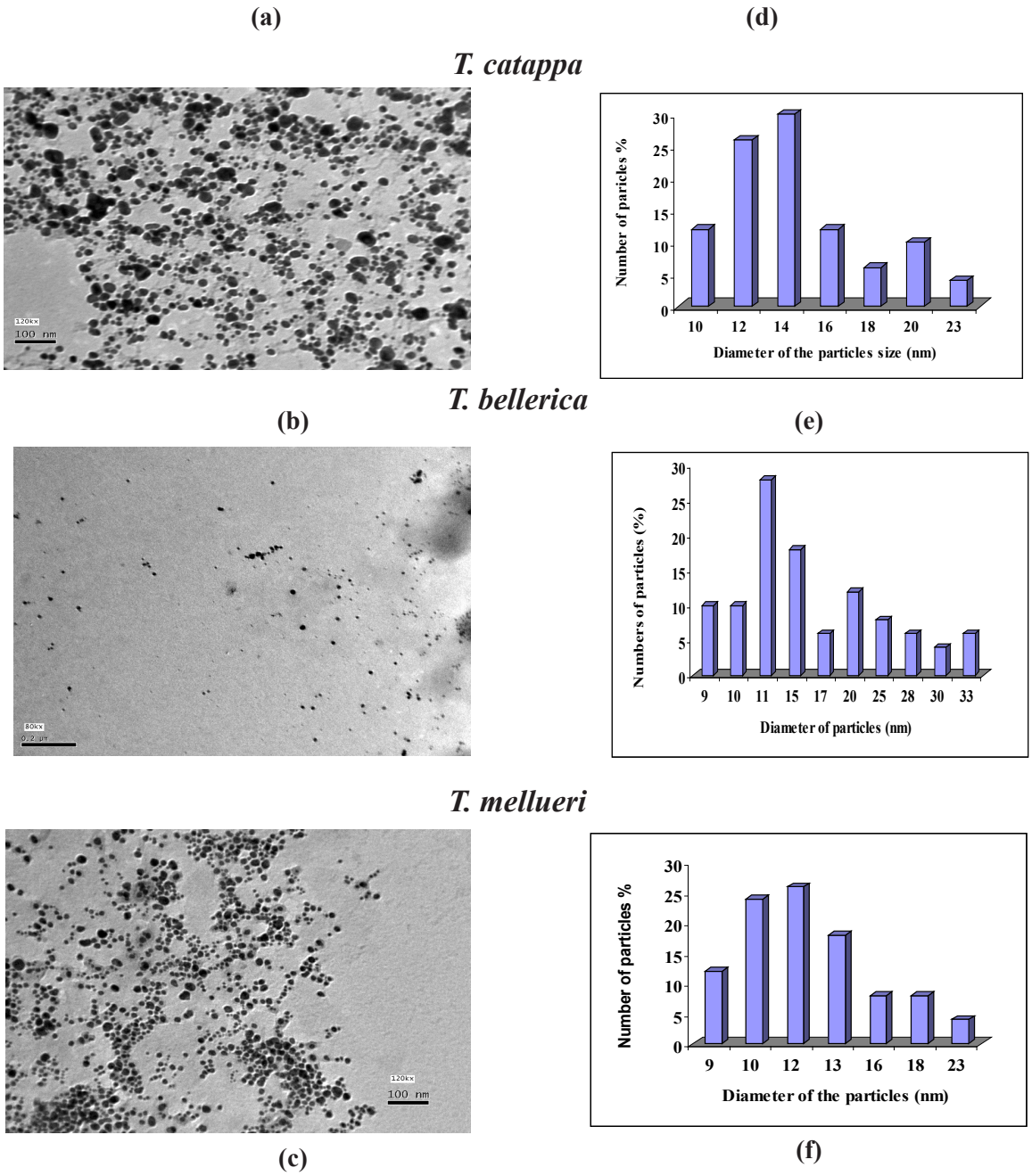


Fig. 4. Representative TEM images of the three *Terminalia species* 80% ethanolic leaf extracts-reduced AgNPs and their corresponding particle size distribution histograms.

TABLE 4. Antibacterial durability of treated fabric.

Treatment set	Sample No.	Treatment solution or Suspension	Reduction ratio	
			E. coli	***Staph***
	1	*T.C*	94.8%	94.8%
A	2	*T.B*	97%	97.6%
	3	*T.M*	95.3%	94.6%
	4	*T.C* + Binder	89.3%	86.2%
B	5	*T.B* + Binder	92.3%	89.8%
	6	*T.M* + Binder	93.5%	91.3%
	10	AgNPs from *T.C* + Binder	100%	100%
C	11	AgNPs from *T.B* + Binder	100%	100%
	12	AgNPs from *T.M* + Binder	100%	100%

T.C= fabric treated with the crud ethanol extract of *Terminalia catapa*
T.B= fabric treated with the crud ethanol extract of *Terminalia bellarrica*
T.M= fabric treated with the crud ethanol extract of Terminalia mellueri

TABLE 5. Antibacterial durability of the treated cotton fabric after 5 and 10 washes.

Treatment set	Sample No.	Treatment solution or Suspension	Antibacterial durability			
			After 5 washes		After 10 washes	
			Staph	***E. coli***	***Staph***	***E. coli***
	1	*T.C*	92.4%	92.2%	79.3%	84%
A	2	*T.B*	93%	92.5%	87.4%	88.7%
	3	*T.M*	92.5%	91.9%	88.6%	83.7%
	4	*T.C* + Binder	94.7%	95.8%	82.6%	87.0%
B	5	*T.B* + Binder	95.9%	94.7%	87.2%	84.9%
	6	*T.M* + Binder	95.3%	95.3%	89.1%	85.8%
	10	AgNPs from *T.C* + Binder	96.2%	96.2%	97.4%	97.6%
C	11	AgNPs from *T.B* + Binder	98.3%	98.5%	96.6%	95.9%
	12	AgNPs from *T.M* + Binder	99.4%	97.3%	97.2%	96.4%

T.C= fabric treated with the crude ethanol extract of *Terminalia catapa*
T.B= fabric treated with the crude ethanol extract of *Terminalia bellarrica*
T.M= fabric treated with the crude ethanol extract of *Terminalia mellueri*
T.C AgNPs = fabric treated with silver nanoparticles prepared from *Terminalia catapa*
T.B AgNPs = fabric treated with silver nanoparticles prepared from *Terminalia bellarric*
T.M AgNPs = fabric treated with silver nanoparticles prepared from *Terminalia mellueri*

Anti-inflammatory activity of the treated cotton fabrics

Apart from being an excellent antibacterial agent, AgNPs appears to have anti-inflammatory properties as well. Concerning this, the plant extracts as well as the AgPNs prepared from each plant extract was independently applied to cotton fabric and all treated samples are examined towards anti-inflammatory activity in male-albino rats. The results obtained are tabulated in **Table 6**. Untreated fabric sample as well as the fabric treated with indomethacin cream as a reference drug is also depicted in this table.

The results obtained indicated that: (a) the untreated cotton fabric (control) sample has no anti-inflammatory activity, (b) the fabric samples singly treated with the three ethanolic plant extracts imparted lower anti-inflammatory activity as compared to that of the standard indomethacine-treated fabric sample, and (c) the fabric samples severally treated with AgNPs, prepared from different ethanolic plant extracts, showed comparable anti-inflammatory activity as that of the reference sample. This holds true, irrespective of the type of the plant leave used.

The aforesaid outcomes confirms the roles of both phytochemical constituents present in the ethanolic plant extracts as well as the AgNPs for enhancing the anti-inflammatory activity of the treated fabric samples.

TABLE 6. Anti-inflammatory activity of treated samples and indomethacin drug in male albino rats.

Treatment of Fabric	% Oedema Mean ±S.E	% of change	Relative potency
Untreated fabric (Control)	62.3±2.1*	-------	-----
Indomethacin	21.4 ±0.3*	65.65	100
Fabric samples treated with extracts only			
T.C. extract	34.7±1.2*	43.82	66.74
T. M extract	36.8±1.3*	40.77	62.10
T. B extract	37.3±1.4*	40.13	61.13
Fabric samples treated with AgNPs prepared from different extracts			
AgNPs from *T.C*	26.9±0.7*	56.82	86.55
AgNPs from *T.B*	28.2±0.6*	64.36	98.03
AgNPs from *T.M*	30.2±0.8*	51.52	78.47

Values are expressed as mean ± SEM of n = 6 animals in each group.
* Significantly different from control at $p<0.01$

Conclusion

This manuscript is a considerable effort in the production of environmentally safe natural based antibacterial and anti-inflammatory finished fabrics using the aqueous ethanolic extracts of the three *Terminalia* spices, namely, *T. catapa* (*T.C*), *T. bellarica* (*T.B*) and *T. mellurie* (*T.M*). The formation of silver nanoparticles was confirmed by Surface Plasmon Resonance (SPR) at (424-430) nm and by using Fourier transformed infrared spectroscopy (FT-IR) and transmission electron microscopy (TEM). Quantification of total phenolic compounds, flavonoids, carbohydrates and protein contents of these extracts were estimated since these constituents have dual effects as reducers for silver ions as well as stabilizers for the synthesized AgNPs. The cotton fabrics treated with the synthesized AgNPs showed good antimicrobial and anti-inflammatory activities toward both Gram-positive bacteria (S. *aureus* ATCC 25923) and Gram-negative bacteria (E. *coli* ATCC 25922).

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الأنشطة المضادة للبكتيريا والإلتهابات لقماش القطن المعالج بالفضة النانوية المحضرة من المستخلصات الكحولية لثلاث فصائل من نبات الترمناليا

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لأن تكنولوجيا النانو، هذا المجال الواسع فى القرن ال ٢١ ، له تأثير حاسم بشكل استثنائي على الصناعة فى العالم وعلى حياة الناس، لذا فإن هذا العمل يهدف إلى تخليق حيوى سريع بطريقة خضراء لجسيمات فضة نانوية (AgNPs) بإستخدام مستخلصات الإيثانول لثلاث فصائل من نبات الترمناليا ، وهي ترمناليا كاتابا (T.C)، ترمناليا بيلاريكا (T.B) وترمناليا ميلوري . (T.M)تم التأكد من تكوين جسيمات الفضة النانوية (AgNPs) باستخدام طيف الأشعة فوق البنفسجية المرئية مع خاصية (رنين البلازمون السطحى) عند قيم تتراوح بين ٤٢٤-٤٣٠ nm ، كما تم تأكيد تكوين هذه الجسيمات أيضاً بإستخدام جهاز تحويل الاشعة تحت الحمراء (FTIR) والمجهر الإلكتروني النافذ ((TEM. تناول البحث أيضاً التقدير الكمى لمكونات هذه المستخلصات مثل الكربوهيدرات ، المركبات الفينولية الكلية ، الفلافونويدات ، البروتين – حيث يلعب كل من هذه المكونات أدواراً مزدوجة مؤثرة فى تخليق جسيمات الفضة النانوية (AgNPs) من حيث كونها تختزل كاتيونات الفضة الى فضة معدنية نانوية ثم تعمل على تثبيتها فى حالة معلقة بعد تكوينها. وتم أيضاً تحليل كل مستخلص نباتى عن طريق كروماتوجرافيا الغاز المقترن بمطياف الكتلة (GC/MS). أوضحت نتائج هذا البحث أن الأقمشة القطنية التى تمت معالجتها بجسيمات الفضة النانوية والمحضرة من المستخلصات المذكورة – كل على حدة- تعطى نشاطاً جيدا كمضادات للبكتيريا والإلتهابات سواء ضد البكتيريا موجبة الجرام مثل (S. *aureus* ATCC 25923) أو البكتيريا سالبة الجرام مثل (E. *coli* ATCC 25922)