Available online on www.ijcpr.com
International Journal of Current Pharmaceutical Review and Research; 8(1); 31-39
doi: 10.25258/ijcprr.v8i01.9086
ISSN: 0976 822X

Research Article

The Antibacterial of Essential Fatty Acid Semicarbazide Extracted from Flaxseed Oil Against Some Nosocomial Infection Bacteria in Iraq

Ahmad Ayad Hady*, Mayada F Darweesh, Ahmad A Motar

Department of Biology, College of Sciences, Kufa University, Najaf, Iraq

Available Online: 25th February, 2017

ABSTRACT

During the period from December 2014 to July 2015, isolation and identification of pathogenic bacteria from clinical sources attending to AL-Hakeem Hospital and AL-Sadder Medical City in AL-Najaf province. A total of One hundred clinical samples from various sources included: UTI - patients, Burn, postoperative surgery site –Wound, The results revealed the occurrence (52) bacterial isolate were isolated, 37(71%) were gram– positive bacteria that was divided into *S. aureus* 20(39%), *S. pyogen* 9 (17%), and *S. pneumonia* 8 (15%). But only 15 (29%) was gram – negative bacteria which represented by *E. coli* 7(13%), and *P.aeruginasa* 4 (8%), *Protuse mirabilis* 4(8%). The susceptibility of *S. aureus* and *E.coli* bacteria which are consider the most common G+ve and G-ve nosocomial pathogen to routenly used antibiotic in Najaf hospitals were tested by using antibiotic susceptibility pattrons, in order to select one isolate from *S.aureus* and other one from *E.coli* which had the higher resistance to most antibiotic for continue the other steps of these study. The result illustrated that *S.aureus* No.5 and *E. coli* No.3 were greatly resistant to antibiotics, so regard as multi-drug resistant(MDR). Crud extracts of *Linum usitatissimum* which had been extracted by three solvents (petroleum ether, hexan and aqueous), for studying the chemical contents of these extracts by using suitable reagents and tested antibacterial activity on the growth of studied *S.aureus* and *E. coli* bacteria. The extraction of Essential fatty acid Semicarbazide (omega-3-6-fatty acid) from *Linum usitatissium* (Flax seed) and the chemical and physical properties of isolated oil compounds were studied by using thin layer chromatography (TLC), ultraviolet spectrum (UV) and FTIR spectrum. The result revealed that shown high antibacterial activity against studied (*S.aureus, E.coli*). results explained broad spectrum antibacterial property of oil compounds against studied bacteria nosocomial.

Keyword: *S. aureus* and *E.coli* bacteria , clinical infection , antibiotic sensitivity , *Linum usitatissimum* seed oil , TLC, UV and FTIR.

INTRODUCTION

Nosocomial infection, is an infection received at some point of sanatorium care which become no longer present or incubating at the time of admission, the infections which arise extra than 48 hours after admission are also taken into consideration nosocomial[1]. Nosocomial infections are as a result of pathogens that without problems spread thru the body. Many clinic patients have compromised immune systems, so they're much less capable of combat off infections. In a few cases, patients increase infections due to bad situations at a medical institution or a healthcare facility, or because of clinic workforce no longer following proper procedures[2].

The increase in prevalence of multiple drug resistance has slowed down the development of new synthetic antimicrobial drugs and has necessitated the search for new antimicrobials from alternative sources[3].Due to the widespread and often indiscriminate use of antimicrobial drugs, many bacteria have the genetic ability to transmit and acquire resistance to drugs and these strains are particularly evident in the hospital environment[4]. Bacterial infections are world- wide problem and resistant bacteria is increasing worldwide in both outpatients as well as hospitalized patients, which are considered as a focus of infection[5].

Many of plants are used in medicine for treatment of disease such as Flax (Linum usitatissimum L.) is a multi-purpose crop. Have long been used in human and animal diets and in industry as a source of oil. Recently there has been a growing interest in the probiotic properties of flax and in its beneficial effects on coronary heart disease, brest and prostate cancer and neurological and hormonal disorders[6]. As well as have antibacterial activity against several pathogenic bacteria[7]. Flaxseed is abundant in many nutrients, such as polyunsaturated fatty acid, protein, and lignans[8].

Flaxseed is now attracting expanded interest as a meals because of its high fibre content, high content of bioactive phenolic compounds (specifically lignans) and excessive content material of the important omega-3 fatty acid alphalinolenic acid with ordinary stages of 55% within the oil[9]. Omega-3-fatty acids have anti-inflammatory, antithrombotic antiarrhythmic and hypolipidaemic effects[10]. due to this, those fatty acids are useful in the prevention and treatment of coronary heart disease[11], atherosclerosis, and typeII diabetes[11], rheumatoid

*Author for Correspondence: mayadajalala@yahoo.com

arthritis[10], homo sapiens evolved in an omega-3-rich nutritional environment[12] also for brain development and function[13].

MATERIALS AND METHODS

Isolation and Identification

A total of One hundred clinical samples from various sources included: UTI - patients, Burn, postoperative surgery site -Wound. All isolates were identify by morphology properties after culturing on MacConkey, Manitol salt agar and Blood agar, conventional biochemical test then finally confirmed by using Vitek-2 system gram– positive (G+ve) and gram–negative (G-ve) card. These samples were collected from patients attending to Al-Sadder Medical City, Al-Hakeem General Hospital.

Antibiotic susceptibility testing

Antimicrobial susceptability testing was accomplished by the "Kirby-Bauer disc diffusion method using Mueller-Hinton agar" as recommended by Clinical Laboratory Standard Institute. The antimicrobial agents tested and their corresponding concentrations were as follows: Vancomycin (VA) (30µg), Amikacin (AK) (30µg), Ampicillin (AM)(10µg), Cefotaxime(CTX)(30µg), Ciprofloxacin(CIP)(5µg), Chloramphenicol(C)(30µg), Imipenem (IPM)(10µg), Gentamicin(GM)(10µg), Penicillin (P)(10µg), Rifampin(RIF)(5µg), Tobromycin (TOB)(10µg), Erythromycin(E)(15µg), Tetracycl-ine (T)(30µg).

A small inoculum of bacteria suspension (which prepared by inoculate 5 isolated grown on BHI agar to 5 ml of tryptic soy broth then incubated for 2hr. to produce a bacterial suspension of moderate turbidity that compared with turbidity of ready–made 0.5 McFaerland tube standard) was inoculated on Mueller-Hinton plates and antibiotic discs were placed on the plates, spacing them well to prevent the overlapping of inhibition zones. After incubating the inoculated plates aerobically at 37 C° for 18 to 24h, the susceptibility and resistance of the bacteria isolates to each antimicrobial agent was measured and the results were interpreted in accordance with criteria provided by (CLSI,[14]).

Preparation of Plants Extracts

The plants were extracted with three types of solvents, which were cold, hot distilled water and alcohol (hexan, Petroleum ether). In both cases, the extracted parts of plants were *Linum usitatissium (*Flax seed)[15,16].

preparation of oil

The preparation of oil from flaxseed according to Borhade[16].

Phytochemical screening

Chemical detection of the active components in plant extracts[7].

preparation purification and Identification of Essential Fatty Acid Semicarbazide of Flaxseed(oil) compounds

To determine the purity and Identification of Flaxseed(oil) compounds according to thin layer chromatography(TLC)[15]. Ultra violate (UV) spectra, FT-IR spectra[17].

Preparation of Concentration different from crud plant extract

Stock solution was prepared for each extract by dissolving 500 mg of dried extract with 1 ml of distilled water for aqueous extract and haxan, ptrolum ether for alcohol extract, so the final concentration of extract would be 500 mg/ml, from this stock solution other concentration were prepared 250, 125, 62.5 mg /ml which was used against bacteria[18].

Determination of antibacterial activity

Agar well diffusion method was used to determine the antibacterial activity of EFASC of Flaxseed (oil) compounds according to Egharevba *et al*.[19].

RESULTS AND DISCUSSION

Identification of nosocomial infections bacterial

Out of 100 clinical specimens, only 52 bacterial isolates were giving positive and negative results on general and selective media[9]. as shown in the fig. (1).

The G+ ve bacteria 37(71%) that was divided into *S. aureus* 20(39%), *S. pyogen* 9(17%), and *S.pneumonia* 8(15%). But only 15 (29%) was G –ve bacteria which represented by *E.coli* 7(13%),and *P.aeruginasa* 4(8%), *Protuse mirabilis* 4(8%). According to these results G +ve bacteria were considered to be the dominant bacteria isolated in this study, also this study founded that *S.aureus* and *E.coli* were the commonest offending isolated bacterial pathogen in gram positive and gram negative frequency; this observation is in the same line with Oluwole and Victoria[20] who reported that *S. aureus* and *E. coli* were most common urinary tract pathogen. Dessie et al.[21] who stated that most common organisms associated with postoperative surgical site infections were *S. aureus* followed by *Klebsiella* species and *E. coli*. According to Dessie et al.[21]. *S.aureus* is the most prevalent organism associated with surgical wound infections. It is worth mentioning that all the bacteria isolated from surgical wound infections belonged to *S.aureus, E.coli and P.aerogenosa* . *S. aureus* was the most frequently reported pathogen associated with nosocomial surgical wound infections by Church et al.[22]. Multidrug resistant bacteria have frequently been reported as the cause of nosocomial outbreaks of infection in burn units or as colonizers of the wounds ofburn patients[23].

Antibiotic susceptibility of pathogenic bacteria

As shown in Table (1) and (2) the results of antibiotic resistance profile for twenty *S.aureus* isolates and seven *E.coli* isolates to 13 commonly antibiotics used in hospitals by using Kirby-Bauer disk diffusion method[8] and all results for the diameter of inhibition zone are compared with standard zone of inhibition determined by CLSI[14].

The antibiotic susceptibility results appear high resistance towards Penicillin, Ampicillin, Tetracycline, Rifampin, Gentamicin, and Amikacin *S. aureus* were resistance with percentage 100%, 100%, 95%, 90%,90% and 85% respectively, while *E .coli* resistant with percentage 100% for Penicillin , Ampicillin , Tetracycline and 95 %, for Rifampin and Gentamicin 85% for Amikacin, The bacterial chosen isolates showed a varying resistance to each of Chloramphenicol 85%, Cefotaxime 80%, Ciprofloxacin 70%, Tobromycin 80 % and Erythromycin 75 % for *E.coli* bacteria and Cefotaxime

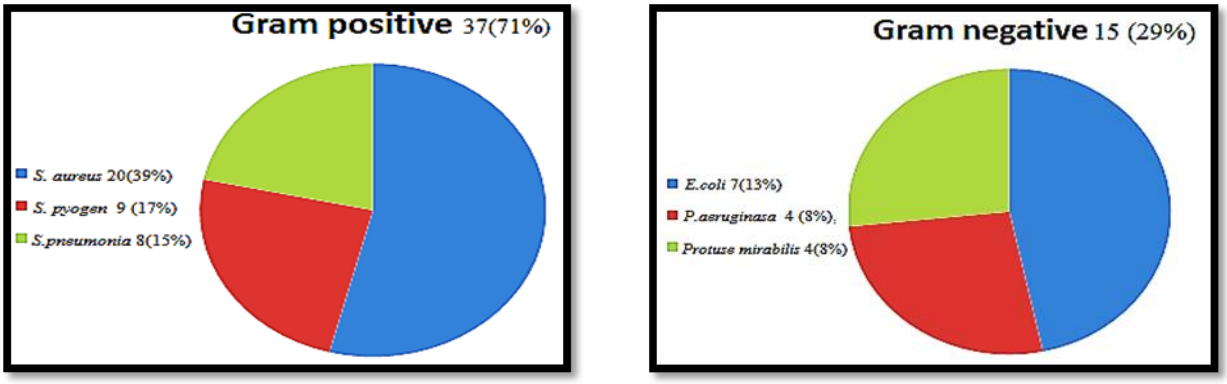


Figure 1: The Gram positive and Gram negative.

Table 1: Antibiotic resistance test of *Staphylococcus aureus* bacteria.

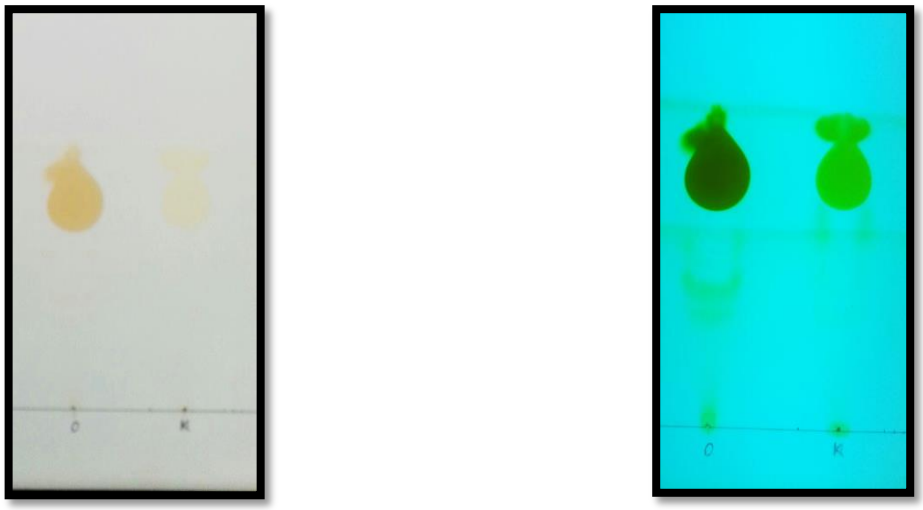
S.aureus	AK	VA	AM	TOB	CIP	C	CTX	GM	P	RIF	T	E	IPM
1	+	-	+	+	-	+	-	+	+	+	+	-	-
2	+	-	+	-	+	+	+	+	+	+	+	+	-
3	+	-	+	-	+	-	+	+	+	+	+	+	-
4	+	-	+	+	-	-	+	-	+	+	+	-	+
5	+	-	+	+	+	+	+	+	+	+	+	+	+
6	+	-	+	+	-	+	-	+	+	-	-	+	-
7	+	-	+	-	+	-	+	+	+	+	+	+	-
8	+	-	+	+	-	+	-	+	+	+	+	+	-
9	+	-	+	-	+	-	+	+	+	+	+	+	-
10	+	-	+	+	-	-	-	+	+	+	+	+	-
11	+	-	+	-	+	+	+	+	+	+	+	+	-
12	+	-	+	+	-	-	+	+	+	+	+	+	-
13	+	-	+	+	+	+	+	-	+	+	+	+	-
14	-	-	+	-	+	+	-	+	+	+	+	+	-
15	+	-	+	+	-	+	+	+	+	-	+	-	-
16	+	-	+	-	+	-	+	+	+	+	+	+	-
17	-	-	+	+	-	+	-	+	+	+	+	+	-
18	+	-	+	-	+	-	+	+	+	+	+	-	-
19	+	-	+	+	-	-	+	+	+	+	+	+	-
20	-	-	+	+	-	+	+	+	+	+	+	+	-
percent	15 %S 85%R	100 %S	100 %R	40 %S 60%R	50 %S 50%R	45 %S 55%R	30 %S 70%R	10 %S 90%R	100 %R	10 %S 90%R	5 %S 95%R	25 %S 75%R	90 %S 10%R

Resistant = R(+) ,and Sensitive=S(-)

Table 2: Antibiotic resistance test of *Escherichia coli* bacteria.

E.coli	AK	VA	AM	TOB	CIP	C	CTX	GM	P	RIF	T	E	IPM
1	+	-	+	-	-	+	+	+	+	+	+	+	-
2	+	-	+	+	+	-	+	+	+	+	+	+	-
3	+	-	+	+	+	+	+	+	+	+	+	+	-
4	+	-	+	+	-	+	+	-	+	+	+	+	-
5	+	-	+	+	+	+	-	+	+	+	+	-	-
6	+	-	+	+	+	+	+	+	+	-	+	+	-
7	-	-	+	+	+	+	+	+	+	+	+	-	-
percent	14.3% S 85.7% R	100 %S	100% R	14.3% S 85.7% R	28.6 %S 71.4 %R	14.3% S 85.7% R	14.3% S 85.7% R	14.3 %S 85.7 %R	100 %R	14.3 %S 85.7 %R	100 %R	28.6 %S 71.4 %R	100 %S

Resistant = R(+) ,and Sensitive=S(-)



A- TLC plates by the daylight. B-TLC plates under UV-Light.

Figure 2: Thin layer chromatography for oil extracts , system hexan / diethyl ether (93/7,v/v). stander EFA(Omega 3) = o , extract =k).

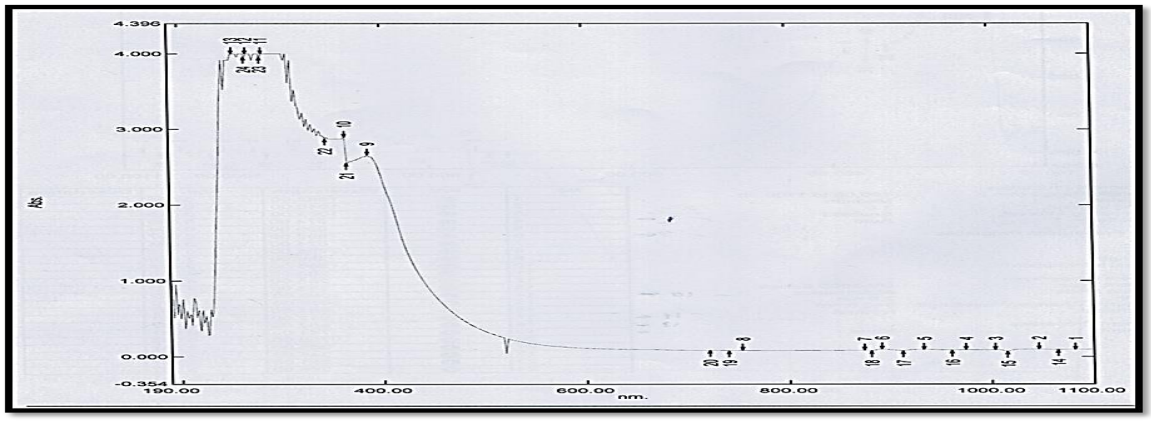


Figure 3: UV spectrum of EFASC of Linseed (oil).

70%, Chloramphenicol 55 %, Ciprofloxacin 50%,Tobromycin 60% and Erythromycin 75% for *S.aureus* on the other hand, *E.coli* bacteria were very sensitive for Imipenem100%whereas *S.aureus* appear100%and90%susceptible for Vancomycin and Imipenem. Isolated *E.coli* were resistant to the most antimicrobial agents tested, whereas most of them were sensitive for Imipenem. These had been correlated properly with the ones received with the aid of Bayram et al.[24] they found that E.coli incredibly sensitive to Imipenem.The results of Cefotaxime susceptibility were similar with that reported by Balakit[25] in Iraq.Gram-negative bacteria produce big portions of type1 cephalosporinase when exposed to first- generation cephalosporin, Ampicillin,and Penicillin G,those antimicrobials are quite simply hydrolyzed by using this enzyme, and inducible organisms are intrinsically resistant to these retailers[26]. The present study revealed that isolate were confirmed β-lactam resistant. Such resistance may be emerged because of the wide spread use of these drugs in Iraqi hospitals. As shown in Table (4) all isolates of *E. coli* were resistant to both Ampicillin and Penicillin. The result fitted with local study in Najaf Abeer[27] who reported that all the clinical isolates of *E.coli* were resistant to Ampicillin.Result also were consistent with an investigation done by Ahmed[23] who stated that more than 72% of the Gram-bad isolates of burn wound had been immune to Gentamicin. The results of present study agree with Rubin[25] who explained that *E.coli* 100% resist to Tetracycline and moderate resistance for Gentamicin and Amikacin, at the same time who found that *S.aureus* have moderately resistance for Ciprofloxacin and high unsusceptibility for both Gentamicin and Amikacin.

The results of this study revealed that most of the *S.aureus* were resistant to many antibiotics. Antimicrobial resistance among nosocomial pathogens is a big problem in scientific settings that can be added to the fee of hospital treatment then the morbidity and mortality of sufferers[26]. Mansour et al.[28] mention that emergence of antibiotics unsusceptible *staphylococcal spp* strains may become a big trouble in failure therapy of infection in future by *Staphylococcus spp*.

The result is similar with a local study done by Ahmed[26] who stated that 92% of *S.aureus* , were resistance to Amikacin. These findings were in accordance with many

Table 3: The inhibition zone of flaxseed by use three different solvent extracts (hexan , petroleum ether and water) against *S.aureus*.

Pathogenic bacteria	Inhibition zone on *S.aureus*			
Type of extracts Concentration	petroleum ether	hexan	C.A	H.A
500 mg /ml	33±0.577	30±0.57	28±0.57	24±0.577
250 mg /ml	32±0.5	29±0.57	22±0.57	20±0.57
125 mg /ml	21±0.57	20±0.5	20±0.577	18±0.5
62.5 mg /ml	20±0.5	18±0.57	18±0.5	15±0.57
Control	-	-	-	-
LSD (0.05) = 3.765	Cold aqueous= C.A, Hot aqueous= H.A			

Table 4: The inhibition zone of flaxseed by use three different solvent extracts (hexan , petroleum ether and water) against *E.coli*.

Pathogenic bacteria	Inhibition zone on *E.coli*			
Type of extracts Concentration	Petroleum ether	hexan	C.A	H.A
500 mg /ml	33±0.577	28±0.57	26±0.57	24±0.577
250 mg /ml	29±0.57	26±0.5	20±0.5	19±0.57
125 mg /ml	18±0.57	19±0.577	19±0.57	17±0.577
62.5 mg /ml	17±0.5	16±0.5	16±0.57	15±0.5
control	-	-	-	-
LSD (0.05) = 3.765	Cold aqueous= C.A , Hot aqueous= H.A			

Table 5: UV spectral data of EFA (Omega3) compounds by using Haxan as solvent, λ max (nm).

Compounds 0.05 mg/ml		EFA (Omega 3)	EFASC
Band I	λ max	386	366
	Abs	2.659	0.069
Band II	λ max	363	305
	Abs	2.893	0.169

Abs. = Absorption , λ max = wavelength maximum

Table 6: Thin Laye Chromatography results of Essential Fatty Acid Semicarbazide of Linseed(oil) compounds:

Properties	EFA (Omega 3)	EFASC
R*f*	0.36	0.36
Color by daylight	Dark Brown	Light Brown
Color under UV-Light	Dark Green	Light Green

local studies reported by Tayh[29] who founded that *S.aureus* have 100% resistance to Pencillin and 40% resistant for Ciprofloxacin while Huda[30] In Karbala observed that *S.aureus* were 100% resistance for Ampecillin, 95.8% to Pencillin, moderate resistant for Gentamicin and high sensitive for Chloramphenicol. Rağbetli et al.[31] In Diyala explained that the resistance rates to Erythromycin among hospital inpatient isolates were 56.1% compared with 11.4% in nonhospital patient and 13.6 % of the hospital isolates were resistant to Rifamicin while 100% sensitive for Vancomycin. Bashaer[32] in Karrbala observed the Ampicillin, Erythromycin do not have effect against both *E.coli* and *S.arueus* where as they were sensitive to Tobtramycin and Chloramphincol. The high antimicrobial resistance of most bacterial isolates is possibly promoted due to a selective stress exerted on micro organism for numerous motives like non-adherence to health center antibiotic policy and immoderate and indiscriminate use of huge-spectrum antibiotics. those multi drug resistant lines set up themselves in the health facility environment in areas like sinks, taps, railing, bed, lavatories and thereby spread from one affected person to any other[33]. The results illustrated that *E.coli* No.3 and *S.aureus* No.5 were resistant to all antibiotics this may be due to multiple resistance of nosocomal infections, these two isolates had been chosen to continue and complete other steps of the study.

Phytochemical screening

Phytochemical screening was done using color forming and precipitating chemical reagents on the seeds of *Linum usitatissimum* for alcoholic and cold– hot aqueous extracts which contain alkaloids and flavonoids, phenolic compounds, resins, saponins, Amino acid. But Glycosides and tannins were found to be negative according to Amin and Thakur[7].

Antibacterial activity of plants extracts

Agar -well diffusion method was used to detect the antibacterial activity for studied flaxseed extracts against *E. coli* and *S. aureus* selected isolates.

The result of this study for the antibacterial activity of three different solvent extracts (hexan, petroleum ether and water)prepared from flaxseed which were screened against pathogenic bacteria *S.aureus* and *E.coli*, explaining that Petroleum ether crude extract expressed maximum inhibitory zone at concentration 500 and 250 mg/ml which was 33±0.577, 32±0.57 mm against *S. aureus* and *E. coli* was 33±0.577 , 29±0.57 mm respectively but in low concentration 62.5 mg/ml was 20±0.5 mm against *S. aureus* and 17±0.5 mm against *E. coli* . while the inhibition zone diameter reach to 30±0.57 and 29±0.57

Table 7: FT-IR spectral data of EFA (Omega3) compounds recorded as KBr discs (cm^{-1}).

Compounds	C-H stretching vib. Str.	CH_2 - asym str.	CH_2 - symmetric str.	O=C-O Str.	Solid fatty acid (CH_2 Vib.) Str.	-C=O Str.	R-$COOCH_3$ Str.	R-CH=CHR_3 Str.	H atoms Str.
EFASC	3009.9	2923.1	2853.4	1459.6-1743	1374.8	1258.3	1159.3	1023.5	792.4
Standard EFA (Omega3)	3011.6	2923.7	2853.1	1459.9-1742.8	1375.2	1230.5	1144.7	1096.4	716.8

mm against *S.aureus* in 500 and 250 concentration of hexan extract and 28±0.57 , 26±0.5 mm against *E. coli* respectively, the low concentration of hexan recorded 18±0.57 mm for *S. aureus* and 16±0.5 mm for *E .coli*. On the other hand the inhibition zone diameter of cold extract reached to 28±0.57 mm for *S.aureus* and 26±0.57 mm for *E.coli* at the concentration 500 mg/ml and 18±0.577 mm for *S. aureus* 16±0.57 mm for *E.coli* at 62.5 mg/ ml concentration. While the hot aqueous extract recorded the weak inhibition activity in growth both of *S. aureus* and *E.coli* bacteria which were 24±0.577, 15±0.5 mm in the high and low concentration respectively .As shown in the table (3),(4). These results are compatible with Al-bayatee[32] who observed that antibacterial screening from crude extracts of *Linum usitatissimum* seed showed that the petroleum ether, ethanol, aqueous and chloroform extracts have antibacterial activity against most tested bacteria, and concluded that Petroleum ether extract demonstrated more effective in inhibition growth of *S. aureus* and *E.coli* while Aqueous extract demonstrated moderate activity against most tested bacteria. Ozkan et al.[33] observed that alcoholic extract of plant had strong antibacterial activity against both G+ ve and G- ve while aqueous extract had moderate antibacterial activity, and this activating of plant depends largely on solvent type. In this regard Amin and Thakur[7] found that Chloroform extracts of flaxseeds confirmed antimicrobial interest against all examined microorganisms (*Salmonella typhii, Enterococcus, E.coli, Bacillus subtilis and S.aureus*) while ethanolic extract was effective against all tested bacteria except *E.coli* that showed unsusceptability towared this extract. These results match with the results obtained by Simon et al.[34] who revealed that alcohol extract activity against tested bacteria which can be attributed to the linseed content of saponins, gum, cyanogenic and glycosides which have been reported to contain compounds that have antifungal and antibacterial effect.

Identification of Essential Fatty Acid Semicarbazide(EFASC)

Thin Layer Chromatography

The analysis of TLC chromatography of oil compounds Fig. (2) shows the presence of several spots corresponding to solvent system hexan / diethyl ether (93/7,v/v) appeared light Brown spot on day light and light Green by using UV-light deeper after spraying with (ioden spray). This compound has R*f* approximately which is equal to 0.36. The compound 2 on thin layer chromatography for stander EFA (Omega 3) has R*f* value 0.36 appear as dark Brown at day light and dark Green under UV-light. This compound is stander of the compound numbered 1 on TLC for Linseed oil extract extracts as shown in the table (6) that explain properties the band on TLC plates.

These results approached with Pandya *et al*.[35] who explained that flaxseed (oil) by use hexan solvent gave R*f* value 0.34. Small R*f* value indicated low dissolvability of compound in mobile phase therefore the compound slowly moves to up. Big R*f* value indicated high dissolvability of compound in mobile phase therefore the compound readily moves to up[36]. *Ultraviolet spectra (UV)*

Ultraviolet spectra by Hexan solvent of essential fatty acid semicarbazide(EFASC) (Omega3) compounds were determined. The qualitative UV spectra profile of Standard EFA(Omega3) and Linseed (oil) that isolated from seed of *Linum Usitatissimum* were selected at wavelength from 600.0-1100.0 nm due to sharpness of the peaks and proper baseline. The electronic absorption data of the investigated compounds are gathered in Table (5) and the spectra of these compounds are shown in Figure (3 and 4) the Standard and Linseed (oil) are characterized by two bands, the short bands at 363 nm of Standard and 305 nm of Linseed (oil) . The appearance of the short bands in the electronic absorption spectra of isolated EFA (Omega 3) compounds is ascribed to the locally excited by $n\rightarrow\sigma^*$, $n\rightarrow\pi^*$ transition with the double band (C=N) of the compounds. The spectra of long bands at 386, 366 nm of standard and Linseed (oil) respectively. These results approached with Borhade[16] who explained that Absorption spectra of essential fatty acid semicarbazide (EFASC) of *Linum usitatissimum* (Linseed) seed oil shows maximum absorption 3.182 at 370 nm.

FTIR Spectra

The FTIR spectrum was used to identify the functional group of the active components based on the peak value in the region of infrared radiation. The IR spectra of the studied essential fatty acid EFA (Omega 3) as KBr discs and of their representative spectra are shown in Table (7) and Figure (5 and 6). The spectrum of Linseed (oil) is characterized by nine bands corresponding to the stretching vibrations of the C-H stretching vibration of the cis- double (=CH), CH_2 - asym stretching, CH_2 - symmetric stretching, O=C-O, Solid fatty acid (CH_2 Vib.), -C=O, R-$COOCH_3$, R-CH=CHR_3, and H atoms, which occur in (3009.9), (2923.1), (2853.4) (1459.6-1743),

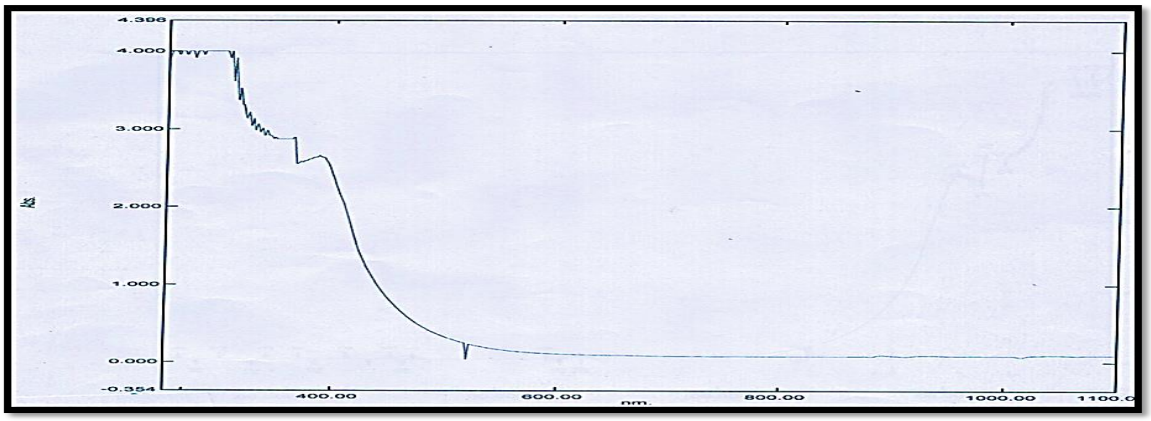


Figure 4: UV spectrum of Standard EFA (Omega3).

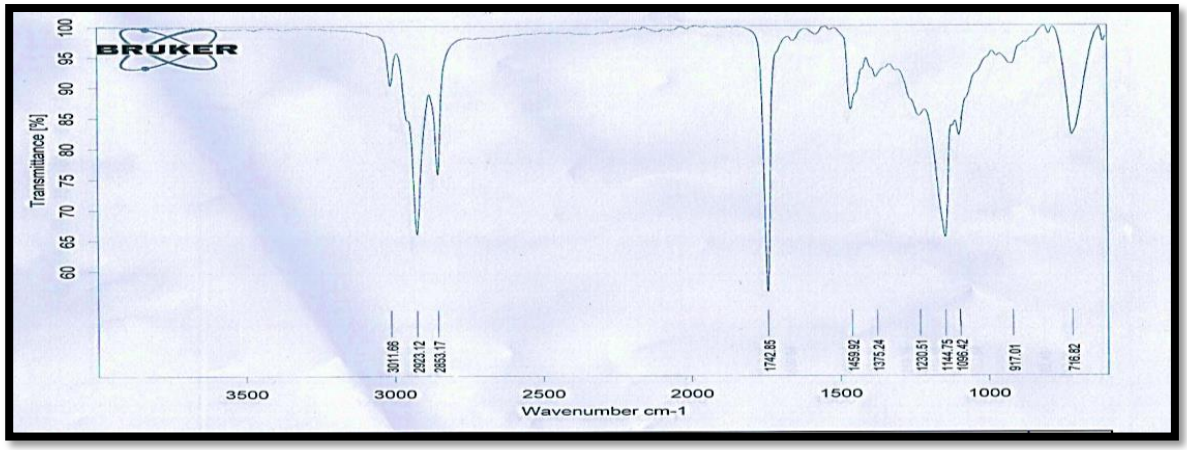


Figure 5: FT-IR Spectrum of essential fatty acid (SFA) (Omega 3).

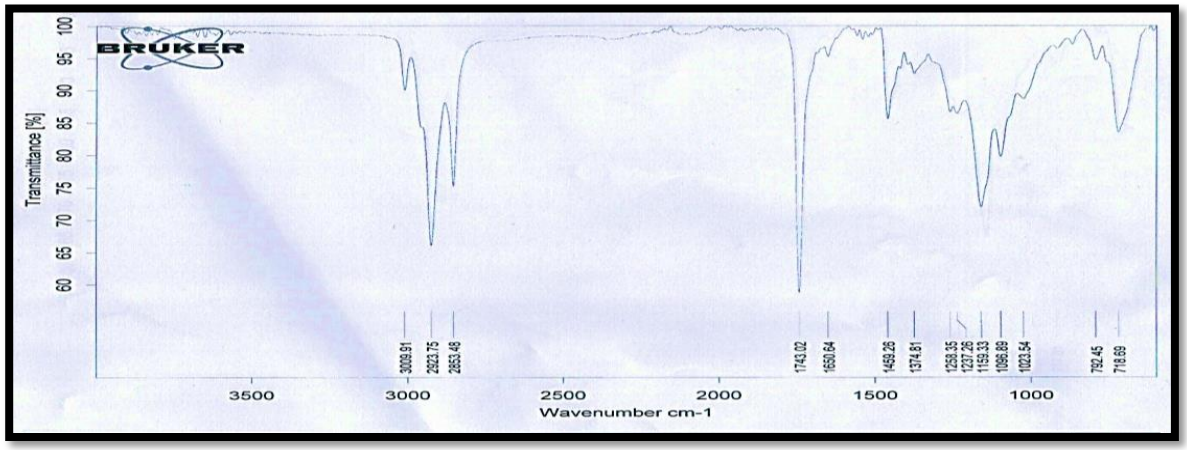


Figure 6: FT-IR Spectrum of EFASC of Flaxseed (oil).

(1374.8), (1258.3), (1159.3), (1023.5) and (792.4) cm-1 respectively. The absorption data of IR of standard showed seven stretching vibration bands, which confirmed the correctness of the proposed structure. These bands C-H stretching vibration of the cis- double (=CH), CH_2 - asym stretching, CH_2 - symmetric stretching, O=C-O**,** Solid fatty acid (CH_2 Vib.), -C=O, R-$COOCH_3$, R-$CH=CHR_3$, and H atoms which occur in (3011.6), (2923.7), (2853.1), (1459.9-1742.8), (1375.2), (1230.5), (1144.7), (1096.4), and (716.8) cm-1 respectively. The results of functional groups of FTIR of isolated EFA(Omaga3) compounds are in agreement with EFASC of flaxseed(oil) structure. The results are nearly similar with study by Chauhan et al.[37] they founded that oil of Linseed have the characteristics bands associated with 2924 cm^{-1} (CH_2 - asym stretching), 2856 cm^{-1} (CH_2 - symmetric stretching). The band at 3008 cm^{-1} assigned to the C-H stretching vibration of the cis-double (=CH). The strong peak

Table 8: The inhibition zone of essential fatty acid semicarbazide (EFASC) extracts against *S.aureus* and *E.coli*.

Bacteria Concentration Of EFASC	Inhibition zone on *S.aureus*	Inhibition zone on *E.coli*	Control (DMSO)
500mg /ml	34 ± 0.577	31 ± 0.57	-
250 mg /ml	30 ± 0.57	29 ± 0.577	-
125 mg /ml	24 ± 0.577	23 ± 0.57	-
62.5 mg /ml	20 ± 0.57	19 ± 0.577	-
Tetracycline	16 ± 0.5	15 ± 0.5	-

LSD (0.05) = 3.638 Not inhibition (-)

demonstrated at 1743 cm^{-1} is attributed to ester carbonyl functional group of triglyceride and fitted with results done by Borhade[16] who observed present seven functional group in flaxseed oil with the nearly same stretching.

The antibacterial activity of essential fatty acid semicarbazide (EFASC) extracts against chosen bacteria

Antibacterial Activity of EFASC of Linseed oil was analyzed. The EFASC were evaluated for their ability to inhibit the growth against bacteria *E. coli* and *S. aureus* by the agar-well diffusion inhibition test contrast to tetracycline which is regarded as standard antibiotic as preliminary test, no references for the inhibitory effect of our EFASC compounds were found in the Iraqi searchers, the results were explained in Table (8) that show the effect of different concentrations (62.5, 125, 250 and 500 mg/ml) of EFASC which increased the inhibition zone against bacteria. The inhibition zone of EFASC of flaxseed (oil) was 34± 0.577 mm against *S. aureus* and 31± 0.57mm against *E. coli* in concentration 500 mg/ml, whereas the inhibition zone diameter against *S. aureus* and *E. coli* at low concentration came to be between 20± 0.577, 19± 0.577 mm respectively. These results agree with Borhade[16] who shows that EFASC of *Linum usitatissimum* (Linseed) seed oil posses good antibacterial activity against *E.coli* and *S.aureus* at varied level and found that *S.aureus* more active in inhibition zone than *E.coli* .

The results of this study are correlated well with those obtained by Mokbel and Hashinaga[38].They illustrated that linseed oil content is essential fatty acid which included palmatic acid, linoliec acid and oleic acid and are reported to contain antibacterial properties and compatible with study by Seidel and Taylor[39] they found that the antibacterial action of fatty acids is usually attributed as being a property of the long-chain unsaturated fatty acids, including oleic acid, linoleic acid, and linolenic acid, while long-chain saturated fatty acids, including palmitic acid and stearic acid, are reported as showing less antibacterial activity.

REFERENCES

1. Milton, A.; Priya, G.; Aravind, M.; Parthasarathy, S.; Saminathan, M.; Jeeva, K. and Agarwa, R. Nosocomial Infections and their Surveillance in Veterinary Hospitals. DOI | http://dx.doi.org/10.14737/journal.aavs/3.2s.1.24 ISSN (Online) 2015, | 2307-8316; ISSN (Print) | 2309-3331.
2. Aljanaby, A.A.J.J. Antibacterial activity of an aqueous extract of Petroselinum crispum leaves against pathogenic bacteria isolated from patients with burns infections in Al-najaf Governorate, Iraq. Res Chem Intermed. 2012;38(9).
3. Chanda, S.; Vyas, B.R.M.; Vaghasiya, Y. and Patel, H. Global resistance trends and the potential impact of Methicillin Resistant Staphylococcus aureus (MRSA) and its solutions. Department of Biosciences, Saurashtra University, Gujarat, India.2010.
4. Shahid, M. and Malik, A. Resistance due to aminoglycoside modifying enzymes in *Pseudomonas aeruginosa* isolates from burns patients. *Indian J Med Res* 2005; 122; 324-9.
5. Ventola, C The Antibiotic Resistance Crisis. P T.; 2015;40(5): 344–352.
6. Aljanaby, A.A.J.J and Gafil, F.A.A, Comparative study among different antibiotics effect on aerobic pathogenic bacteria that cause otitis media and urinary tract infection in Al-Manathera city in Iraq. Res Chem Intermed. 2012;38(9).
7. Amin, T. & Thakur, M. *Linum usitatissimum* L. (Flaxseed)- A Multifarious Functional Food. Online Int. Interdisciplinary Res. IV(I), 2014; 220-238.
8. Mishra, S. and Verma,P. Flaxseed- Bioactive compounds and health significance. 2013;17(3), PP 46-50.
9. Kajla, P.; Sharma, A. and Sood, D. Flaxseed—a potential functional food source. J Food Sci Technol. 2015;52(4): 1857–1871.
10. Simopoulos, A. Omega-3 Fatty Acids and Athletics. Current Sports Medicine Reports, 200; 6:230–236 Current Medicine Group LLC ISSN 1537-890x.2007.
11. Mostafa, M.and Ayimba, E. Effect of omega 3 fatty acids family in human health. International Journal of Advanced Research, 2014; 2(3), 202-211, ISSN 2320-5407.
12. Peet, M. and Stokes, C. Omega-3 Fatty Acids in the Treatment of Psychiatric Disorders. LEADING ARTICLE Drugs 2005; 65 (8): 1051-1059.
13. Lauritzen L; Brambilla, P.;Mazzocchi, A.; Harsløf, LB.; Ciappolino ,V. and Agostoni ,C.(2016). DHA Effects in Brain Development and Function. Nutrients.4;8(1). pii: E6. doi: 10.3390/nu8010006.
14. CLSI. Performance Standards for Antimicrobial Susceptibility Testing: Twenty – Fourth Informational Supplement M02- A11, M 0-A11, and M11-A8. Wayne, PA, USA .2014.
15. Harborne, J. B. (1984). Phytochemical methods.; A Guide to Modern Techniques of Plant Analysis, 2nd ed. Chapman and Hall, London.

16. Borhade, S. (2014). Synthesis, Characterisation and Antimicrobial Activity of Essential Fatty Acid of Semicarbazide. ISSN 0976-2590, Online ISSN 2278 – 6015 .2014;5(2), pp 46-55.
17. Donald, L.; Pavia, G. M.; Lampman, G.S.; Kriz, J. R. & Vyvyan. Introduction to Spectroscopy. 4th ed. Western Washington University U.S. A.: 656 pp.2009.
18. Nwachukwu, E. & Uzoeto, H. O. Antimicrobial activities of leaf of Vitex doniana and Cajanus on some bacteria; Researcher 2010;2 (3): 2460–2465.
19. Egharevba, H. O.; Kunle, O. F.; Iliya, I.; Orji, P. N.; Abdullahi, M. S.; Okwute, S. K.; Okogun, J. I. Phytochemical analysis and antimicrobial activity of Punica granatum L. (fruit and leaves). New York Scie. J.2010; 3 (12): 91-98.
20. Oluwole, O.M. and Victoria, A Asymptomatic bacteriuria: Occurrence and antibiotic susceptibility profiles among students of a tertiary institution in Ile-Ife, Nigeria. 2016; 10(15), pp. 505-510. Article Number: DB8ECAE58164 ISSN 1996-0808. http://www.academicjournals.org/AJMR.
21. Dessie, W.; Mulugeta, G.; Fentaw, S.; Mihret, A.; Hassen, M. and Abebe E. Pattern of Bacterial Pathogens and Their Susceptibility Isolated from Surgical Site Infections at Selected Referral Hospitals, Addis Ababa, Ethiopia. International Journal of Microbiology. Article ID 2016; 2418902, 8 pages.
22. Church, D; Elsayed, S.; Reid, O.; Winston, B. and Lindsay,R . Burn Wound Infections. Clin Microbiol Rev.2006; 19(2): 403–434.
23. Agnihotri, N.; Gupta, V. and Joshi, RM. Aerobic bacterial isolates from burn wound infections and their antibiograms: A five-year study. Burn. 2004;30: 241 243.
24. Bayram, Y.; Parlak, M.; Aypak, C. and Bayram, I. Three-year review of bacteriological profile and antibiogram of burn wound. isolates in Van, Turkey. Int. J. Med. Sci., 2013;10(1): 19-23.
25. Balakit, H.A.J. Clinical Bacteriology and Immunology of Burn Victims. M.Sc. Thesis, Collage of Medicine, University of Babylon. 112 pp. 2007.
26. Ahmed, A. Mohammed. Isolation and Identification of some Microbes and Estimation of some Immunological Aspects in Burn Patients. Master ThesisFaculty of Science, University of Kufa.2013.
27. Abeer, A. Al-Hasnawi. Comparison of biochemical tests, Api system, Vitek 2 system and PCR of the enteropathogenic bacteria isolated from children with persistent diarrhea. And the occurrence of virulence factors and antibiotic resistance in the isolates.Master Thesis. Faculty of Science, University of Kufa .2014.
28. Mansour FA,Mohamed Zaky MM, and El Morsy T (2009). Multi- drug resistance and slime production in coagulase negative staphylococci implicated in nosocomial infection in mansoura Hospitals, mansoura, Egypt. Egypt.J.Exp. Biol.(Bot), 5:207-214.
29. Tayh, G.A. Risk Factors and Antimicrobial Resistance of Pathogens Isolated from Burn Units at Local Hospitals in Gaza Strip, Palestine. M.Sc. Thesis, Collage of Science, University of Gaza, Palestin. 86 pp.2011.
30. Huda, M. Jawad. Assessment of Some immunological aspects in Immunodeficiency with skin infection in Karbala. Master Thesis. Faculty of Science, University of Karbala .2014.
31. Rağbetli, C.; Parlak, M.; Bayram, Y. Guducuoglu, H. and Ceylan, N. Evaluation of Antimicrobial Resistance in *Staphylococcus aureus*Isolates by Years. Interdiscip Perspect Infect Dis; 2016: 9171395.
32. Bashair, S. Hadi. Effect of some fruits peel extracts on the virulence of some pathogenic bacteria isolated from urinary tract infection in pregnant women in Karbala. Master Thesis. Faculty of Science, University of Karbala. 2013.
33. Omran L., E Askar. Antibiotic Sensitivity Patterns of the most common bacteria isolated from Al-Mouwasat University Hospitalin, Syria, International Jounal of Pharm Teach Research .2016;9(1).
34. Simon, N.; Santhoshkumar, R. and Kumark,N. (2016). Phytochemical analysis and antimicrobial activities of Annona squamosa (L) leaf extracts. E-ISSN: 2278-4136 P-ISSN: 2349-8234 JPP; 2016;5(4): 128-131.
35. Pandya,P.; Harisha ,CR; Shandla,V.J. & Chandola, H. M. Pharmoacogostical and Photochemical evaluation Atasi(Linum ustatissimmum L.).Indian Journal of Tradition Knowledge. 2013; 12(4), PP. 688-692.
36. Keroynz, N. & Anthrykin, J. Identidcation organic compounds. Translated by Yasin shandallah and Nazar AL Jubory. *AL-Mousel Univ*.1956; 225 (In Arabic).
37. Chauhan,R.; Chester,K. ; Khan, Y.; Tamboli, E. and Ahmad, S. Characterization of Linum usitatissimum L. oil obtained from different extraction technique and in vitro antioxidant potential of supercritical fluid extract. J Pharm Bioallied Sci. 2015; 7(4): 284–288.
38. Mokbel, M., and Hashinaga, F. Antibacterial and antioxidant activities of Banana (Musa AAA cv. Cavendish) Fruits Peel. Am. J. Biochem. Biotechnol. 2005;1(3), 126-132.
39. Seidel, V.and Taylor, PW. In vitro activity of extracts and constituents of Pelagonium against rapidly growing mycobacteria. Int. J. Antimicrob. Agents, 2004;23: 613-619.