EVALUATION OF YIELD PERFORMANCE AND ITS QUALITY CHEMICALLY AND BIOLOGICALLY IN CANOLA UNDER SALINITY STRESS CONDITIONS

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This study focused on evaluating canola (*Brassica napus* L.) productivity under salt stress conditions and its impact on oil quality, chemically and biologically. The experiment was conducted during the 2022/2023 season in the salinity-affected North Sinai region, where two canola cultivars (Giza 1 and Gemmiza 1) were grown to evaluate yield and oil quality. The results showed significant differences between both the seed and oil yield of the two tested cultivars where, the Gemmiza 1 cultivar outperformed the Giza 1 cultivar (1869.84, 1683.60 and 753.55, 670.07 kg/ha, respectively). Chemical investigation canola seeds of two cultivars (Giza 1 and Gemmiza1) indicated that, Gemmiza 1 cultivar outperforms Giza 1 cultivar in raising the acid and peroxide values by 3.28 and 1.19 times, respectively. The GC-Mass technique lead to thirteen fatty acids in the oil and four of them are saturated fatty acids. The IC_{50} exhibited by cytotoxic activity against colon carcinoma cell line (Caco2), canola oil of both two cultivars (Giza 1 and Gemmiza 1) was 118.77±2.96 and 110.16±1.34 µg/ml, respectively. Network pharmacology identified some important phytoconstituents linolenic acid and palmitic acid, related to colon cancer targets. The chemical oleic acid and linoleic acid exhibited degree of connection in the network, suggesting that they may act as curative agents against antifungal targets. This study provides valuable information for evaluating oil yield, monitoring the nature of cultivars under a biotic condition and the future trend of canola adoption and the quality of chemically and biologically extracted under salinity stress.

Keywords: salt stress*, Brassica napus,* peroxide, antimicrobial activity**,** pharmacology networking

INTRODUCTION

Salinity is a significant cause of environmental stress and can significantly restrict plant development and crop yield (Koca et al., 2007). Elevated salinity inhibits stomata closure, repeatability and leaf expansion, which is critical for successful germination and plant growth. High salt concentrations in soil and water hurt several physiological processes in plants, including photosynthesis, respiration, transpiration, water conductivity, membrane permeability, nutritional balance, enzyme activity, metabolic activities, cellular homeostasis and hormone regulation (Mahmoud and Abdelhameed, 2021). Abiotic stresses also affect seed yield and its components, as well as oil content, with differences between different crop genotypes (Safavi et al., 2018; El-Sayed and Badran, 2020 and El-kadi et al., 2022).

One of the most significant oil crops in the world; canola has seen a notable increase in production in recent years. Soil salinity is a key barrier to canola seed germination and seedling establishment. This issue has a negative impact on the canola plant growth, development and its yield, which lowers agricultural output. One of the most important times for a crop exposed to salinity is germination. The germination of canola seeds can be impacted by soil salinity in two ways: either by generating an osmotic potential outside the seed that inhibits water uptake or by the poisonous effects of Na and Cl ions on the growing seeds (Khajeh-Hosseini et al., 2003)

Rapeseed (*Brassica napus* var. napus), an annual oilseed crop also known as rape and oilseed rape, belongs to the Brassica family. It is grown worldwide in winter or spring and in Egypt it is grown in winter. In the early 1970s, to ensure that rapeseed was safe for eating by humans and animals, Canadian plant breeders used plant breeding methods to eliminate erucic acid and glucosinolates (the anti-nutritional elements). Canola is also influenced by environmental conditions to a greater extent than by heritability (Brandle and McVetty, 1989). Thus, the reaction of direct choice to seed yield may be unexpected unless environmental alteration is well handled. Erucic acid is the major component of rapeseed oil, so the international standard definition of canola oil distinguishes it from rapeseed oil by stating that it contains less than 2% erucic acid and less than 30 μmol/g glucosinolates (Kondra and Steffanson, 1970). Oils low in erucic acid, such as canola oil, has been associated with a decrease in heart muscle injury (Bauer et al., 2015). Canola oil is composed of low levels of saturated fatty acids (SFAs) (just 7%); it is also characterized by comparatively high levels of monounsaturated fatty acids (MUFAs), moderate levels of polyunsaturated fatty acids (PUFAs) and a good balance between omega-6 and omega-3 fatty acids (Raatz et al., 2018). MUFAs, like oleic acid, are the most prevalent type of fatty acid found in nature. It is the main fatty acid in oils of canola and olive (Nigam et al., 2014 and Raatz et al., 2018). Oleic acid comprises sixty-one percent of the fatty

acids in canola oil. Canola oil comes in second place after olive oil among common vegetable oils concerning oleic acid concentration (Liu and Iassonova, 2012; Nigam et al., 2014 and Liu et al., 2016). Research conducted over the last fifteen years has illustrated that dietary oleic acid, as well as linoleic acid, is just as beneficial as dietary PUFAs in decreasing plasma cholesterol levels. Oleic acid is not needed in the diet because humans and other species can synthesize it; it is not an essential fatty acid. Moreover, canola oil has a moderate amount of PUFAs compared to other vegetable oils (Aslam et al., 2009). Since plasma cholesterol is a significant risk factor for coronary heart disease and is lowered by PUFAs, there is interest in these fats due to their role as necessary fatty acids. It has long been known that linoleic acid is a necessary fatty acid. Since animals and humans cannot synthesize it must be included in their diets. They can transform linoleic acid into other members of the omega-6 family of fatty acids, such as arachidonic acid. Prostaglandins, thromboxanes, prostacyclins, and leukotrienes are examples of "hormone-like" compounds that are synthesized from arachidonic acid, which is also essential for membrane structures. These chemicals are known as eicosanoids collectively and they play a crucial role in a multitude of physiological responses, such as immunological response and blood coagulation (Benito et al., 2001).

It is well-recognized that consuming too much SFAs is linked to an increase in plasmatic cholesterol and obesity (Ristic and Ristic, 2003). Conversely, PUFAs and MUFAs consumption has been suggested to enhance the lipid profile as opposed to SFAs. According to Yu-Poth et al. (2000), PUFAs-rich meals may cause LDL cholesterol oxidation to increase while HDL cholesterol levels decrease. Increased recommendations for MUFAs consumption are common; these recommendations appear to not affect HDL levels and may even lower blood levels of triacylglycerol and LDL, making them more beneficial in preventing heart disease. Canola oil variations had a high MUFAs and PUFAs content (Bauer, et al., 2015). The iodine value in canola oil ranges from 120.41 g I_2/100 g; being the greatest, to 83.14 g I_2/100 g, being the lowest (Roiaini et al., 2015).

Antimicrobial activity is primarily dependent on two types of fatty acids (hydrocarbon chains with a carboxylic acid functional group) and monoglycerides (esterified from a fatty acid and glycerol molecule). Fatty acids are produced from lipids by the action of enzymes, to release free fatty acids, which have important biological activities (Desbois and Smith, 2010). The biological activities of free fatty acids have roles in host defenses against potential opportunistic or pathogenic microorganisms. This is carried out by growth suppression or eradication of bacteria. (Desbois and Smith, 2010). Many scientists evaluated the antimicrobial activity of fatty acid derivatives. However, they gave weak antimicrobial activity against *Escherichia coli* and *Staphylococcus aureus* (Shukla et al., 2018). Many fatty acids, such as lauric, palmitic, linolenic, linoleic, oleic, stearic and myristic acids are noted to

possess antimicrobial activity mainly against bacteria and fungi (Seidel and Taylor, 2004).

Concerning the anticancer effect, fatty acids diminish the proliferation of cancer cells. It has been demonstrated that a mixture of fatty acids, lowers the proliferation of human melanoma cancer cell line WM793, mostly liable for mitochondrial pathway-dependent apoptosis (Domagała et al., 2021). Latest study proves the valuable impacts of dietary PUFAs on dimension tumor development and safe in the treatment of cancer. Long chain fatty acids adjust the tumor cell reaction to chemotherapeutic drugs. Many researches proved that fatty acids not only enhance the tumoricidal effect of anticancer drugs but also increase the ingestion of anticancer medicine leading to an increase in the intracellular aggregation of the anticancer agents (Selvaraj, 2017).

Good seedling establishment requires seeds with a high tolerance to environmental stresses, particularly salinity. These seeds must also be able to supply the seedlings with essential nutrients until they are well-established and capable of independent photosynthetic growth, which is positively correlated with germination rate, germination speed, and seedling growth (Bewley and Black, 1994).

This study set out to assess the biological and chemical aspects of canola crop performance and quality under salt stress.

MATERIALS AND METHODS

1. Experimental Conditions and Plant Materials

Canola seeds of two cultivars (Giza 1 and Gemmiza1) were obtained from the Agricultural Research Center, Giza, Egypt and were sown during the winter season 2022/2023. The study site was located in the Central North Sinai region, represented by the coordinates: 30° 56’ 41.3’’ N and 32° 28’ 02.0’’ E. The surface layers of soil (0 to 40 cm) were analyzed to determine the percentage of available elements, organic matter, salinity and pH in the soil, in addition to analyzing the water used for irrigation during the season.

2. Seed and Oil Yield of Canola Cultivars under Experimental Conditions

At harvesting some measurements were recorded of two tested cultivars as follow: seed yield (kg/ha), oil yield (kg/ha) and oil percentage under experimental conditions in the study region.

3. Chemical Evaluations

3.1. Acid value assay

The acid value was estimated by titration method (Rao et al., 2009 and Neagu et al., 2013) by titration of the oil in an alcoholic medium against a standard 0.1 N potassium hydroxide solution using phenolphthalein indicator.

3.2. Peroxide value assay

Peroxide value was conducted according to Pearson (1976) and Board method earlier research and procedure Japanese Association of Oil Chemists (1972).

3.3. Iodine value assay

The Iodine value was conducted based on the Hanus method is a technique for the determination of the iodine value of oils and fats. It involves the reaction of a sample with Hanus solution (iodine monobromide in glacial acetic acid) and then titration the remaining iodine with sodium thiosulfate, and the calculations approach according to Neagu et al. (2013).

3.4. Preparation and quantification of fatty acids using GC-Mass

Saturable fatty acid methyl esters and oil extraction were made using the techniques explained by El-Kashef et al. (2014) and Mahmoud et al. (2015). In the lab of the National Research Centre, fatty acid was separated and quantified using gas chromatography-mass spectrometry as follows: After extraction, the samples were resuspended in 50 µl of BSTFA (N, O-Bis(trimethylsilyl) rifluoroacetamide) and heated to 70°C for 30 minutes in a dry block heater

3.5. Gas chromatography–mass spectrometry (GC-MS)

The GC-MS apparatus (Agilent Technologies) was outfitted with a gas chromatograph (7890B) and mass spectrometer detector (5977A) at the North Laboratories Network, National Research Centre, Cairo, Egypt. The GC was outfitted with an HP-5MS column (30 m x 0.25 mm internal diameter and 0.25 µm film thickness). Analyses were performed using hydrogen as the carrier gas at a flow rate of 1.0 ml/min at a splitless, injection volume of 1 µl and the following temperature program: 50°C for 1 min; rising at 10°C/min to 300°C and held for 20 min. The injector and detector were held at 250°C. Mass spectra were obtained by electron ionization (EI) at 70 eV; using a spectral range of m/z 30-700 and solvent delay 9 min. The mass temperature was 230°C and Quad 150°C. Identification of different constituents was determined by comparing the spectrum fragmentation pattern with those stored in Wiley and NIST Mass Spectral Library data.

4. Network Pharmacology Profiling of Compounds

4.1. Potential target screening of active compounds and anti-colon cancer, antibacterial and antifungal activity

The data of proper targets for effective phytoconstituents were restored from SwissTargetPrediction (http://www.swiss targetprediction.ch/) and HIT (http://www.badd-cao.net:2345/search) and SEA Search Server (https://sea.bkslab.org/) by inserting the SMILES and point out species as “Homo sapiens”. The colon cancer, antibacterial and antifungal targets were downloaded from GeneCard (http://www.genecards.org/) and OMIM (https://www.omim.org/). The targets of these databases were unified and withdraw duplications in targets. The mutual targets were also explored from

UniProtKB (https://www.uniprot.org/). Venn diagram formation utilizing Bioinformatics means (https://bioin formatics.psb.ugent.be/webtools/Venn) was used to obtain the common targets of phytoconstituents and colon cancer, antibacterial and antifungal (Tabassum et al. 2022 and Shahzadi et al., 2024).

4.2. Construction of compound-target network

To inspect the relationship of active phytoconstituents inside the biological system, the compound-targets network was developed by using Cytoscape V3.10.3 (https://cytoscape.org/). Nodes stand for the phytoconstituents and targets, with edges elucidating their interactions. The network analyzer function was utilized to evaluate the fundamental characteristics of the network. Following this, the network underwent filtering based on the "degree," which represents the number of connected nodes linked to a specific network node as nodes attribute (Ram et al., 2023).

5. In-Vitro Cytotoxic Activity

5.1. Measurement of the potential cytotoxic activity

The potential cytotoxic activity was carried out on colon carcinoma cell lines (Caco2), utilization the oil produced from two canola cultivars (Giza 1 and Gemmiza 1), while the positive control utilized was doxorubicin. The methods were in accordance with the (MTT protocol) viability assay (Slater et al., 1963 and Van de Loosdrecht et al., 1994).

5.2. Morphological assay

Large-scale, morphological changes that occur at the cell surface, or in the cytoskeleton, can be followed and related to cell viability. Damage can be identified by large decreases in volume secondary to losses in protein and intracellular ions due to altered permeability to sodium or potassium. Necrotic cells: nuclear swelling, chromatin flocculation, loss of nuclear basophilia. Apoptotic cells: cell shrinkage, nuclear condensation and nuclear fragmentation (Alley et al., 1988).

6. Antimicrobial Activity

6.1. Pathogenic microorganisms

Gram positive bacteria: *Bacillus subtilis* ATCC 6633 and *Staphylococcus aureus* ATCC 6538

Gram negative bacteria: *Escherichia coli* ATCC 7839 and *Pseudomonas aeruginosa* ATCC 9027

Fungi: *Candida albicans* ATCC 10231

6.2. Antimicrobial activity

The tested pathogenic microorganisms were inoculated into Mullar hinton broth medium with a concentration of 4×106 CFU/ml for bacteria (24h at 37^{o}C) according to McFarland standard (McFarland, 1907). Oil samples were tested for their antimicrobial potentiality determined by well diffusion method as the presence of inhibition zones (Perez et al., 1990).

6.3. Minimum inhibitory concentration (MIC)

Minimum inhibitory concentration (MIC) of the oil samples was dissolved in the least volume of dimethyl sulfoxide (DMSO) and was tested against different pathogenic microorganisms. In this respect different concentrations of the oil were used (5.0, 25.0, 50.0, 75.0, 100, 125, 150, 175, and 200 µg/ml), control hole was loaded with dimethyl sulfoxide (DMSO). The standard used was Imipenem (for bacteria) and fluconazole (for fungi). The MICs (µg/ml) were determined (Tripathi, 2004).

7. Statistical analysis

The data were statistically analyzed by the usage of Minitab software's one-way analysis of variance (ANOVA) approach. There were found to be significant (p<0.05) variations between the means.

RESULTS AND DISCUSSION

1. Experimental Site Conditions

The results of water and soil analysis indicate that the average salinity concentration in irrigation water (Peace Canal) is in the low concentration range, while the average salinity concentration in groundwater and soil is in the high concentration range (Tables 1 and 2). In the same manner, the results obtained indicate that the concentration of all microelements as well as heavy metal concentration was within the permissible limits, while the concentration of boron in ground water was relatively high (Table 3).

2. Seed Yield, Oil Yield and Oil Percentage under Experimental Conditions

The data in Table (4) show significant differences between the seed yield of the two tested cultivars and also the oil yield per hectare, where Gemmiza 1 appears superior to the Giza 1 cultivar. While there are no significant differences between the two tested cultivars in the percentage of oil extracted from the seeds.

Table (1). Some chemical properties of water used in the study rejoin (irrigation water and groundwater).

Irrigation water										
pH	**E.C. (dS/m)**	**T.D.S. (ppm)**	**Soluble cations (meq/L)**				**Soluble anions (meq/L)**			
			Ca^{++}	**K^{+}**	**Na^{+}**	**Mg^{++}**	**Cl^{-}**	**SO_4^{--}**	**HCO_3^{-}**	**CO_3^{--}**
7.1	1.712	977.8	3.35	0.41	9.51	3.95	8.99	3.93	4.00	0.40
Groundwater										
pH	**E.C. (dS/m)**	**T.D.S. (ppm)**	**Soluble cations (meq/L)**				**Soluble anions (meq/L)**			
			Ca^{++}	**K^{+}**	**Na^{+}**	**Mg^{++}**	**Cl^{-}**	**SO_4^{--}**	**HCO_3^{-}**	**CO_3^{--}**
7.6	8.57	5488	21.25	1.28	37.71	26.09	41.05	33.08	11.15	0.70

E.C.: electric conductivity; T.D.S.: total dissolved solids

Table (2). Some microelements and heavy metal concentration of irrigation and water table of water used in the study rejoin (irrigation water and groundwater).

Irrigation water	**Microelements concentration (mg/L)**						
	Cu	Fe	Mn	Zn	Ni	B	Mo
	<0.002	0.039	0.026	0.040	<0.0005	0.101	0.005
	Heavy metal concentration (mg/L)						
	Al	Cd	Co	Cr	Pb	Sr	V
	1.385	<0.0002	0.0003	<0.001	0.032	0.866	0.016
Groundwater	**Microelements concentration (mg/L)**						
	Cu	Fe	Mn	Zn	Ni	B	Mo
	<0.002	0.173	0.010	0.028	<0.0005	1.885	0.012
	Heavy metal concentration (mg/L)						
	Al	Cd	Co	Cr	Pb	Sr	V
	0.356	<0.0002	0.0004	0.004	0.037	9.922	0.032

Table (3). pH, electrical conductivity, organic material and calcium carbonate content in the soil.

pH	E.C. dS/m	O.M (%)	$CaCO_3$ (%)
8.56	6.464	0.79	3.45

Table (4). Seed, oil yield (kg/ha) and oil (%) under experimental conditions.

Cultivars	Seed yield (kg/ha)	Oil yield (kg/ha)	Oil (%)
Giza 1	1683.60[b]	670.07[b]	39.8[a]
Gemmiza 1	1869.84[a]	753.55[a]	40.3[a]

3. Chemical Evaluations

The data in Table (5) presents the significant differences of some of the physical and chemical properties (acid value, peroxide value and iodine value) of oil extracted from two canola cultivars (Giza 1 and Gemmiza 1). According to the results, the Gemmiza 1 cultivar outperforms the Giza 1 cultivar in raising the acid and peroxide values by 3.28 and 1.19 times, respectively. Under growth conditions in North Sinai, the percentage of iodine value reduces by 3.69% in the Gemmiza 1 cultivar relative to the Giza 1 cultivar. The outcomes are entirely in line with earlier research by Neagu et al. (2013) and Roiaini et al. (2015); canola oil has a higher peroxide value according to AbdulKarim et al. (2007), who confirmed that the oil containing a high percentage of linolenic acid, the peroxide value must be high. This suggested that the oxidation process was accelerated by the canola oil's inherent antioxidants Roiaini et al. (2015). Oils' peroxide value increases when they are exposed to light and room temperature air (Siddique et al., 2010). The augmentation of the peroxide value can also be positively impacted by trace levels of heavy metals. Furthermore, because canola oil has a high percentage of linoleic acid, the degree of unsaturation of the oil changes, which causes a rise in iodine values. The amount of iodine in canola oil increases with its linoleic acid level (Siddique et al., 2010). Also, depending

on variations in the cultivar and purity of the canola oil supply, the iodine value may change (Long et al., 2005 and Abdel-Razek et al., 2011).

Table (5). The physiochemical properties in canola oil produced from two cultivars (Giza 1 and Gemmiza 1) grown under Central North Sinai conditions.

Canola cultivars	Acid value (mg/g)	Peroxide value ($meqO_2$ /kg)	Iodine value (gI_2 100/g)
Giza 1	2.77^b	15.98^b	102.55^a
Gemmiza 1	9.10^a	19.10^a	98.76^b

In canola oil derived from two cultivars (Giza 1 and Gemmiza 1) under agricultural conditions in Central North Sinai, Table (6) displays the quality and quantity of fatty acids. There are thirteen fatty acids in the oil, and four of them (palmitic, stearic, arachidic, and lignoceric acids) are SFAs. Compared to the Giza 1 cultivar, the Gemmiza 1 cultivar has a 28% higher amount of SFAs. Nine USFAs, including MUFAs, such as palmitoleic, oleic, *cis*-11-eicosenoic, erucic, and nervonic, are also present in canola oil. The four types of PUFAs are linoleic, linolenic, *cis*-11, 14-eicosadienoic, and *cis*-13, 16-docosadienoic. Additionally, the results show that in Central North Sinai's agricultural settings, the Giza 1 cultivar outperforms the Gemmiza 1 cultivar in terms of raising the percentage of USFAs by 2.65%. Under growth conditions in Central North Sinai, the Gemmiza 1 cultivar was found to have a lower proportion of erucic acid (14.72%) and a higher percentage of oleic acid (1.27%) compared to the Giza 1 cultivar. the acquired data were backed up by (Zambiazi et al. 2007 and Bauer et al., 2015), A sample of canola oil containing several fatty acids, such as caproic acid, caprylic acid, capric acid, lauric acid, myristic acid, palmitic acid, stearic acid, arachidic acid, behenic acid, lignoceric acid, oleic acid, linoleic acid, linolenic acid, and erucic acid, was subjected to both quantitative and qualitative estimation.

The results presented in Table (7) indicate that the Giza 1 cultivar outperforms Gemmiza 1 in terms of improving the ratio of USFAs to SFAs by a factor of 1.31. Additionally, the research indicates that under agricultural conditions in Central North Sinai, the Giza 1 cultivar is defined by an elevation in the percentage of MUFAs, which is 2.38 % higher than that of the Gemmiza 1 cultivar. Likewise, the data demonstrated that the oil from the Giza 1 cultivar had 3.21% more PUFAs acids than that from the Gemmiza 1 cultivar. Furthermore, the percentage of MUFAs and PUFAs compared to the percentage of SFAs for the same cultivar shows that the Giza 1 cultivar is superior to the Gemmiza 1 cultivar by 31.21% and 32.07%, respectively. Under farming conditions in Central North Sinai, there were no appreciable differences between the Giza 1 and Gemmiza 1 cultivars regarding the ratio of MUFAs to PUFAs.

Table (6). Fatty acid composition of oil produced from two canola cultivars (Giza 1 and Gemmiza 1) grown under Central North Sinai conditions.

		Fatty acid name	Carbon	Giza 1	Gemmiza 1
Fatty acids composition (%)	**Saturated**	Palmitic acid	$C_{16:00}$	5.13	7.21
		Stearic acid	$C_{18:00}$	2.42	2.23
		Arachidic acid	$C_{20:00}$	0.81	1.18
		Lignoceric acid	$C_{24:00}$	0.16	0.29
	Total saturated			8.52	10.91
	Unsaturated	Palmitoleic acid	$C_{16:1}$	0.38	0.61
		Oleic acid	$C_{18:1}$	43.99	44.55
		Linoleic acid	$C_{18:2}$	19.22	18.72
		Linolenic acid	$C_{18:3}$	9.75	9.51
		cis-11-Eicosenoic acid	$C_{20:1}$	14.77	12.91
		cis-11,14-Eicosadienoic ac	$C_{20:2}$	0.65	0.58
		Erucic acid	$C_{22:1}$	1.97	1.68
		cis-13,16-Docosadienoic ac	$C_{22:2}$	0.24	0.12
		Nervonic acid	$C_{24:1}$	0.49	0.42
	Total unsaturated			91.46	89.10

Table (7). The ratios between saturated, monounsaturated and polyunsaturated fatty acids in oil produced from two canola cultivars (Giza 1 and Gemmiza 1) grown under Central North Sinai conditions.

Ratios between fatty acids	Giza 1	Gemmiza 1
TUSFAs: TSFAs	10.73:1	8.17:1
TSFAs	8.52	10.91
TMUSFAs	61.60	60.17
TPUSFAs	29.86	28.93
TMUSFAs: TSFAs	7.23:1	5.51:1
TPUSFAs: TSFAs	3.50:1	2.65:1
TMUSFAs: TPUSFAs	2.07:1	2.08:1

TUSFAs: Total unsaturated fatty acids; TSFAs= Total saturated fatty acids, TMUSFAs= Total monounsaturated fatty acids and TPUSFAs= Total polyunsaturated fatty acids.

4. Network Pharmacology Analysis

4.1. Identification of potential targets

Network pharmacology is an interdisciplinary method that investigates complex interactions between biotic systems, compounds and diseases to gain a comprehensive interpretation of drug actions and discover newfound curative targets (Nogales et al. 2022 and Yuan et al. 2022). The 88 targets were collected from 10 phytoconstituents through the SwissTargetPrediction, HIT and SEA. The potential targets of colon cancer, antibacterial and antifungal found in the databases GeneCard and OMIM were (981, 985 and 1169, respectively). Following the withdrawal of replication

and the incorporation of different targets, 14 common targets were identified for colon cancer, 35 common targets for antibacterial and 43 common factors for antifungal activity. Implying possible intersections between phytoconstituent targets and those correlated with these activities. These shared targets were regarded as possible targets for the selected plants in their colon cancer-related, antibacterial and antifungal actions (Figs. 1, 2 and 3).

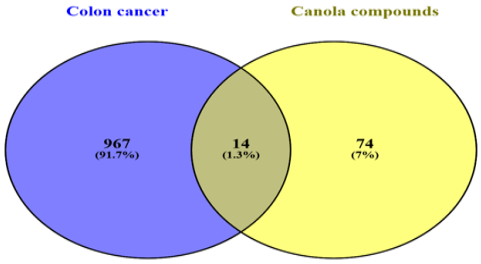

Fig. (1). Venn diagram of potential protein targets between canola oil phytoconstituents and colon cancer.

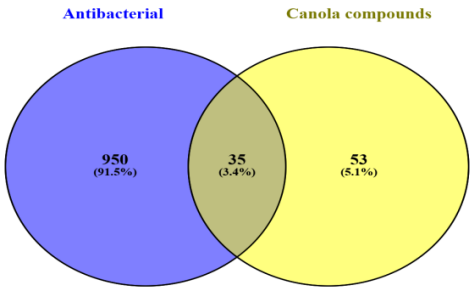

Fig. (2). Venn diagram of potential protein targets between canola oil phytoconstituents and antibacterial activity.

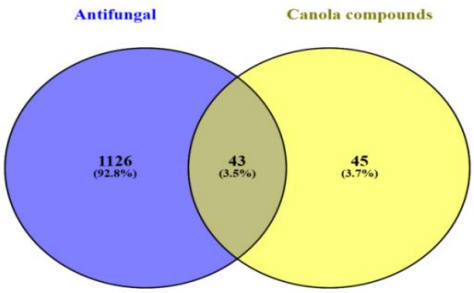

Fig. (3). Venn diagram of potential protein targets between canola oil phytoconstituents and antifungal activity.

4.2. Construction of compound-target network

4.2.1. Colon cancer activity

Compound-target network was developed by using Cytoscape to study the connection among the 7 active phytoconstituents and 14 possible targets (Fig. 4). In Fig. (4), the network pink-colored nodes represent the phytoconstituents; yellow-colored nodes show the possible targets of colon cancer. The active phytoconstituents and possible target protein were assessed, yet as the mode of action is still a question. To reveal the molecular mechanism, a network of 7 bioactive phytoconstituents and 14 possible target proteins was formed. The network included 22 nodes and 38 edges. The topological examining of the network discovered it consists of 0.165 density, 0.822 heterogeneity, 0.552 northization, 2.156 characteristic path length and 3.455 average number of neighbors.

The chemical linolenic acid (6), palmitic acid (4), oleic acid (4) and stearic acid (3), laid out larger degree of connectivity in the network, suggest that these phytoconstituents may act as curative agents (Table 8).

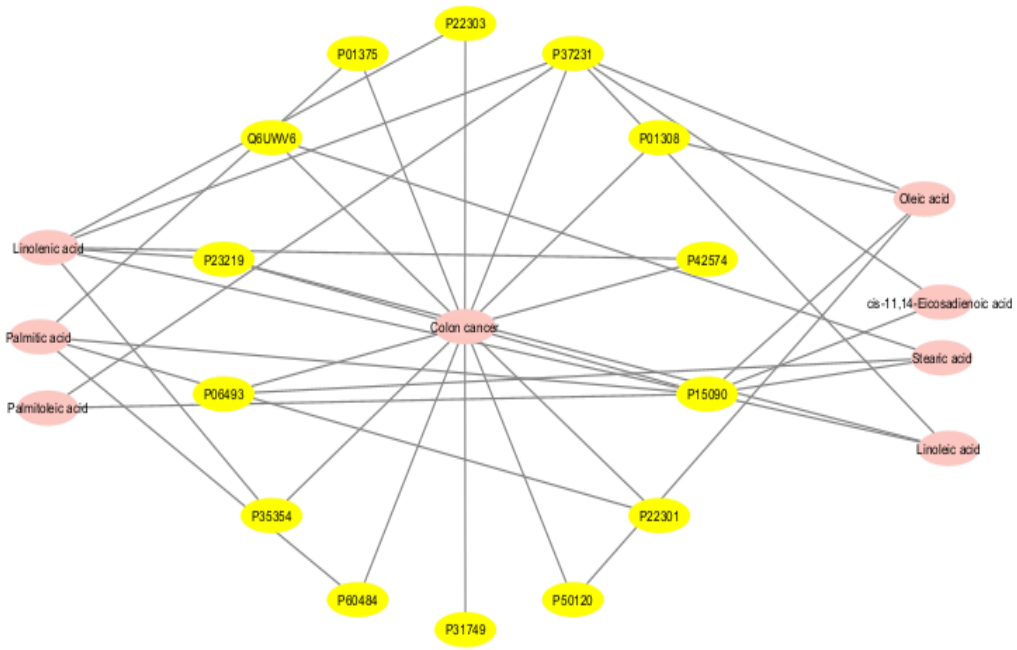

Fig. (4). Compound–target network. pink circles represent the compounds of canola, yellow circles represent targets of colon cancer and edges represent interaction between ingredients and targets.

Table (8). Degree of connectivity of compounds and targets of colon cancer via Cytoscape.

Name	Degree	Closeness centrality	Betweenness centrality
Colon cancer	14	0.750000	0.607564
P15090	8	0.617647	0.248033
P37231	6	0.552632	0.115743
Linolenic acid	6	0.477273	0.072394
Palmitic acid	4	0.437500	0.035850
Oleic acid	4	0.437500	0.033823
P23219	3	0.477273	0.023560
Stearic acid	3	0.420000	0.021043
Linoleic acid	3	0.420000	0.011204
P50120	2	0.456522	0.010119
P01308	2	0.456522	0.010119
P22303	2	0.456522	0.006179
P01375	2	0.456522	0.010370
Q6UWV6	2	0.456522	0.014246
P06493	2	0.456522	0.014246
P35354	2	0.456522	0.006179
P60484	2	0.456522	0.010370
P22301	2	0.456522	0.010370
P42574	2	0.456522	0.006179
Palmitoleic acid	2	0.403846	0.001918
cis-11,14-Eicosadienoic acid	2	0.403846	0.001918
P31749	1	0.437500	0.000000

4.2.2. Antibacterial activity

Compound-target network was developed by using Cytoscape to study the connection between the 8 active phytoconstituents and 35 possible targets (Fig. 5). In Fig. (5), the network pink-colored nodes represent the phytoconstituents; yellow-colored nodes show the possible targets of colon cancer. The network included 44 nodes and 82 edges. The topological analysis of the network revealed that it consists of 0.087 density, 1.437 heterogeneity, 0.762 Northization, 2.174 characteristic path length and 3.727 average number of neighbors.

The chemical oleic acid (12), linoleic acid (11), palmitic acid (8) and linolenic acid (7) laid out larger degree of connectivity in the network, indicating that these phytoconstituents may act as curative agents (Table 9).

4.2.3. Antifungal activity

Cytoscape was used to create compound-target network to analyze the interaction between the 8 active compounds and 43 possible targets (Fig. 6). In Fig. (6), the network pink-colored nodes represent the phytoconstituents; yellow-colored nodes show the possible targets of antifungal activity. The network contained 52 nodes and 100 edges. The algebraic inquiry of the network revealed that it consists of 0.075 density, 1.621 heterogeneity, 0.798

northization, 2.152 characteristic path length and 3.846 average number of neighbors. The chemical oleic acid (18), linoleic acid (13), palmitic acid (8) and linolenic acid (8), laid out larger degree of connectivity in the network, indicating that these phytoconstituents may act as curative agent (Table 10).

5. Cytotoxic activity

As shown in Fig. (7) and Table (11) the cytotoxic activity against colon carcinoma cell line (Caco2), canola oil of both two cultivars (Giza 1 and Gemmiza 1) caused inhibition of cell viability which was correlated to the concentration of the oil (IC_{50} 118.77 ± 2.96 and 110.16 ± 1.34 µg/ml, respectively). This is also proven by morphological assay as illustrated in Figs. (8, 9 and 10).

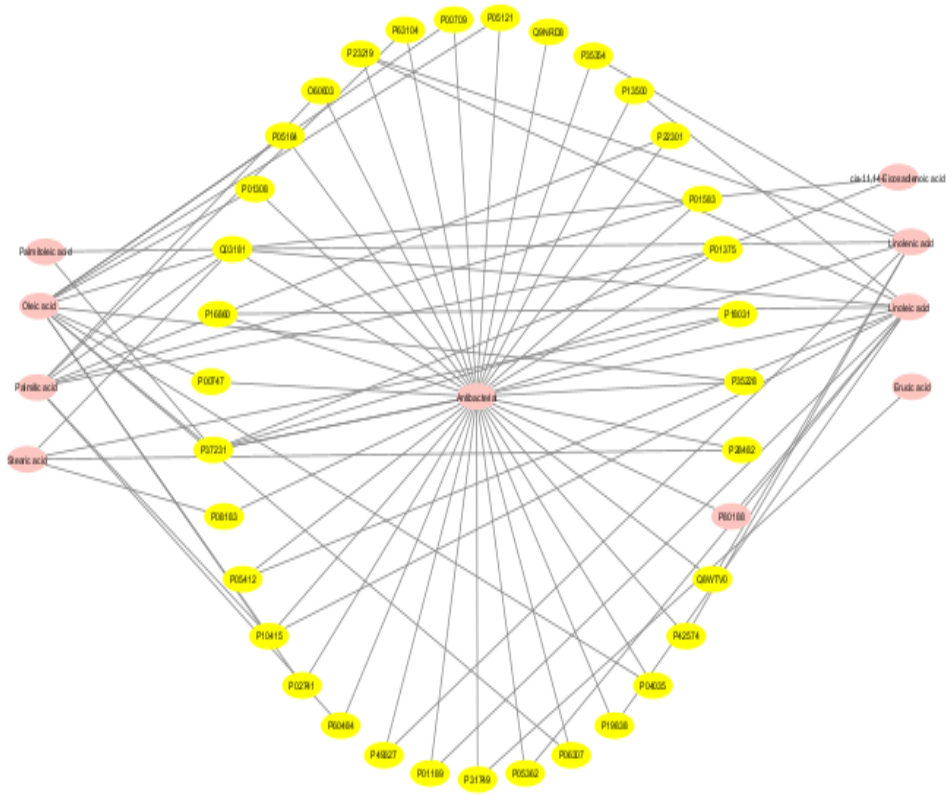

Fig. (5). Compound–target network. pink circles represent the compounds of canola; yellow circles represent targets of antibacterial and edges represent interaction between ingredients and targets.

Table (9). Degree of connectivity of compounds and targets of antibacterial via Cytoscape.

Name	Degree	Closeness centrality	Betweenness centrality
Antibacterial	35	0.843137	0.771231
Oleic acid	12	0.434343	0.055250
Linoleic acid	11	0.425743	0.046620
Q03181	8	0.544304	0.094554
Palmitic acid	8	0.401869	0.024347
Linolenic acid	7	0.394495	0.019630
P37231	6	0.518072	0.052555
P10415	4	0.494253	0.013354
Stearic acid	4	0.373913	0.006854
P23219	3	0.483146	0.007611
P06307	2	0.472527	0.001998
Q8WTV0	2	0.472527	0.003846
P18031	2	0.472527	0.007332
P13500	2	0.472527	0.002219
P05121	2	0.472527	0.001998
P01308	2	0.472527	0.001998
P00747	2	0.472527	0.001998
P05412	2	0.472527	0.002219
P60484	2	0.472527	0.003361
P31749	2	0.472527	0.046512
P04035	2	0.472527	0.001998
P28482	2	0.472527	0.007332
P01583	2	0.472527	0.003361
P35354	2	0.472527	0.003846
P00709	2	0.472527	0.001998
O60603	2	0.472527	0.003361
P49327	2	0.472527	0.003846
P05362	2	0.472527	0.002219
P42574	2	0.472527	0.003846
P35228	2	0.472527	0.001998
P22301	2	0.472527	0.003361
P63104	2	0.472527	0.003361
P05164	2	0.472527	0.001998
P16860	2	0.472527	0.002219
P08183	2	0.472527	0.007332
P02741	2	0.472527	0.001998
P01189	2	0.472527	0.002219
P19838	2	0.472527	0.002219
P80188	2	0.472527	0.002219
P01375	2	0.472527	0.003361
Palmitoleic acid	2	0.361345	3.81E-04
cis-11,14-Eicosadienoic acid	2	0.361345	3.81E-04
Q9NRD8	1	0.462366	0.000000
Erucic acid	1	0.323308	0.000000

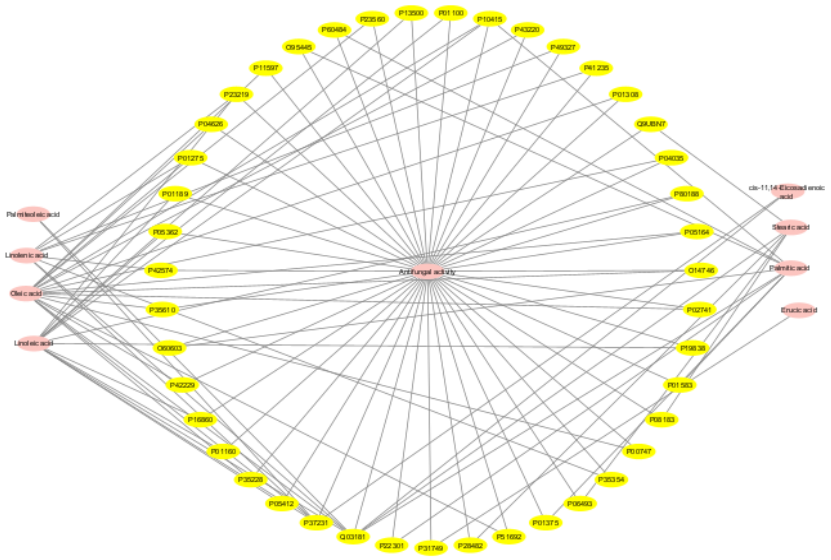

Fig. (6). Compound–target network. pink circles represent the compounds of canola; yellow circles represent targets of antifungal activity and edges represent interaction between ingredients and targets.

Table (10). Degree of connectivity of compounds and targets of antifungal activity via Cytoscape.

Name	Degree	Closeness centrality	Betweenness centrality
Antifungal activity	43	0.864407	0.772415
Oleic acid	18	0.459459	0.080998
Linoleic acid	13	0.421488	0.044081
Q03181	8	0.536842	0.075167
Palmitic acid	8	0.389313	0.016701
Linolenic acid	8	0.389313	0.017006
P37231	6	0.515152	0.043338
Stearic acid	5	0.372263	0.007099
P10415	4	0.495146	0.010424
P01275	3	0.485714	0.005401
P23219	3	0.485714	0.005693
P19838	2	0.476636	0.001642
P80188	2	0.476636	0.001642
P41235	2	0.476636	0.002858
P01100	2	0.476636	0.001642
O95445	2	0.476636	0.002990
P35610	2	0.476636	0.001049
P01160	2	0.476636	0.001642
P51692	2	0.476636	0.001049
P00747	2	0.476636	0.001049
O14746	2	0.476636	0.001049
Q9UBN7	2	0.476636	0.005185
P43220	2	0.476636	0.001049

Table (10). Cont.

P23560	2	0.476636	0.001049
P05362	2	0.476636	0.001642
P42229	2	0.476636	0.001049
P05412	2	0.476636	0.001642
P31749	2	0.476636	0.039216
P06493	2	0.476636	0.005185
P01583	2	0.476636	0.002990
P05164	2	0.476636	0.001049
P01308	2	0.476636	0.001049
P60484	2	0.476636	0.002990
P04626	2	0.476636	0.001049
P42574	2	0.476636	0.002858
P16860	2	0.476636	0.001642
P28482	2	0.476636	0.005185
P35354	2	0.476636	0.002858
P02741	2	0.476636	0.001049
P04035	2	0.476636	0.001049
P49327	2	0.476636	0.002858
P13500	2	0.476636	0.001642
P11597	2	0.476636	0.001049
P01189	2	0.476636	0.001642
O60603	2	0.476636	0.002990
P35228	2	0.476636	0.001049
P22301	2	0.476636	0.002990
P01375	2	0.476636	0.002990
P08183	2	0.476636	0.005185
Palmiteoleic acid	2	0.356643	2.61E-04
cis-11,14-Eicosadienoic acid	2	0.356643	2.61E-04
Erucic acid	1	0.324841	0.000000

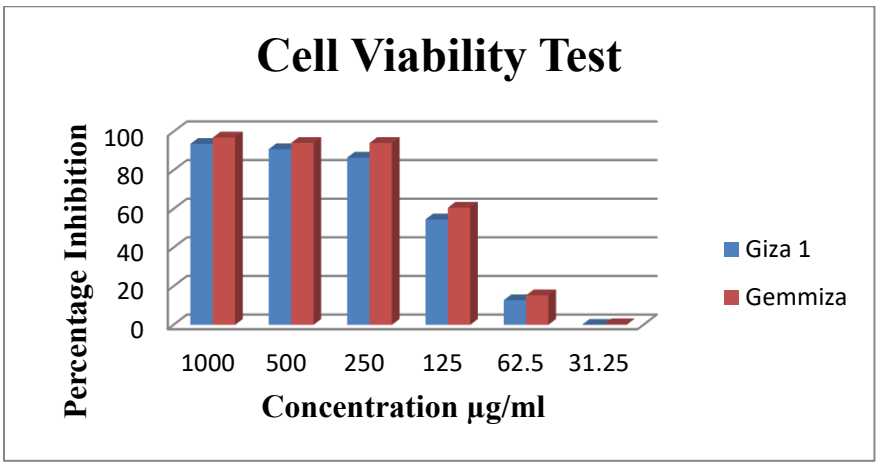

Fig. (7). Percentage inhibition of oil produced from two canola cultivars (Giza 1 and Gemmiza 1) grown under Central North Sinai conditions on colon carcinoma cell line (Caco2).

Table (11). IC_{50} of oil produced from two canola cultivars (Giza 1 and Gemmiza 1) grown under North Sinai conditions on colon carcinoma cell line (Caco2).

	Giza 1	Gemmiza 1	Doxorubicin
IC_{50} (µg/ml)	118.77 ± 2.96	110.16 ± 1.34	15.22 ± 0.07

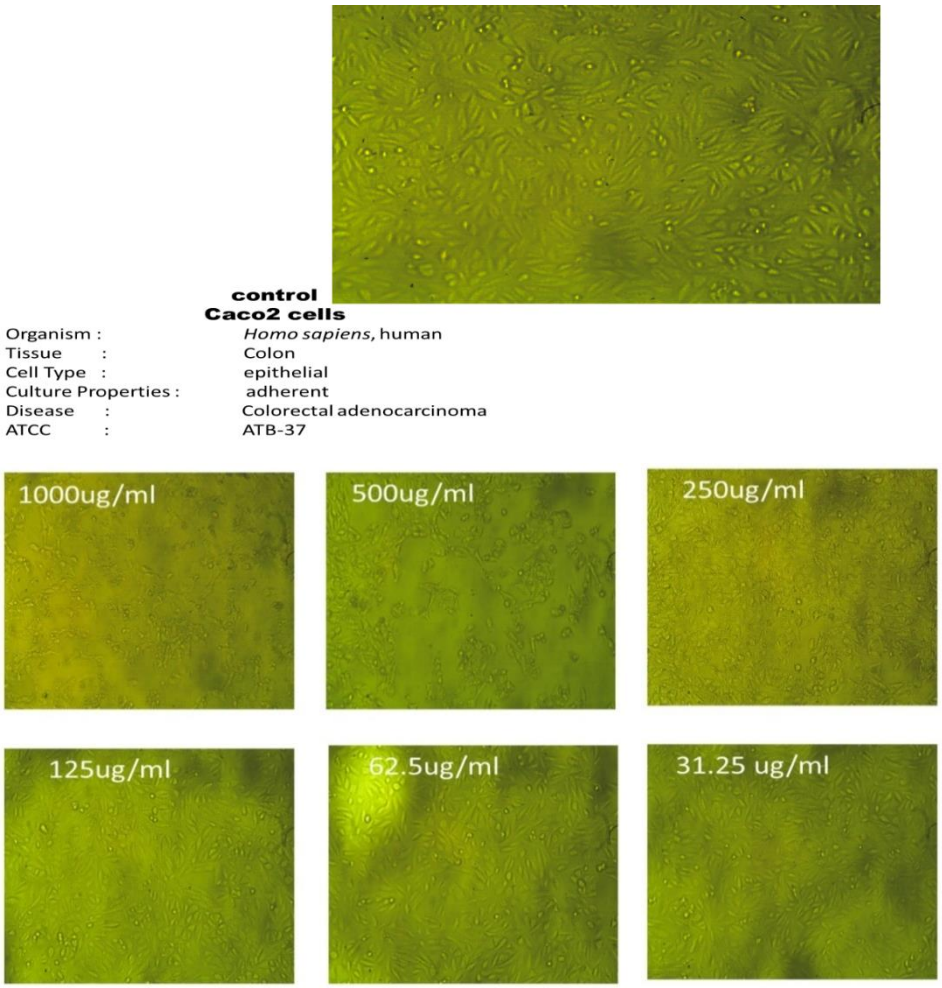

Fig. (8). Effect of oil produced from (Gemmiza 1) canola cultivar grown under Central North Sinai conditions on colon carcinoma cell line (Caco2).

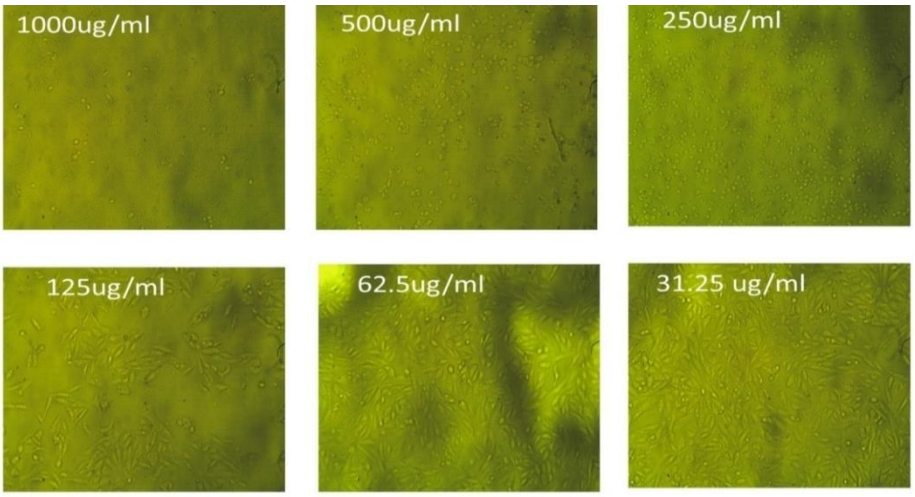

Fig. (9). Effect of oil produced from (Giza 1) canola cultivar grown under Central North Sinai conditions on colon carcinoma cell line (Caco2).

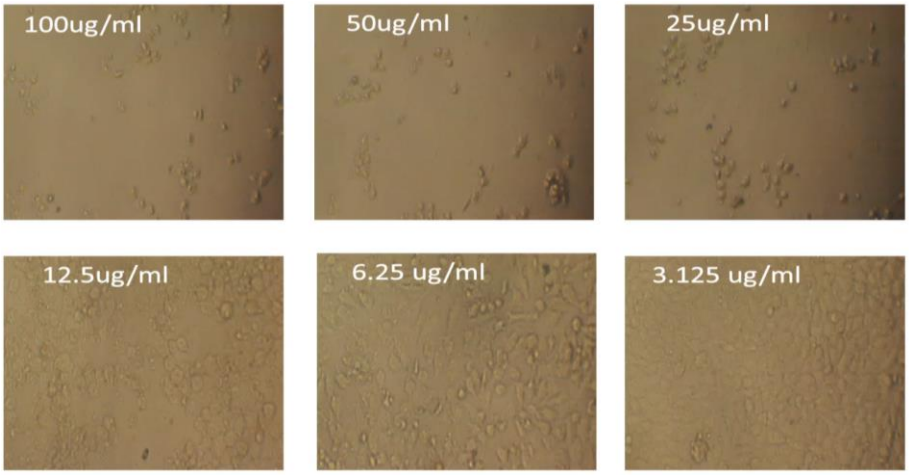

Fig. (10). Effect of doxorubicin on colon carcinoma cell line (Caco2).

Concerning the cytotoxic activity against colon carcinoma cell line both canola oil (Giza 1 and Gemmiza 1) caused inhibition of cell viability which was correlated to the concentration of the oil (IC_{50} 118.77 ± 2.96 and 110.16 ± 1.34 µg/ml, respectively). The main SFAs in both oils produced from canola cultivars (Giza 1 and Gemmiza 1) were palmitic acid (5.13% and 7.21% respectively) and stearic acid (2.42% and 2.23%, respectively). From pharmacology networking the degree of connection of both palmitic acid and

stearic acid were (4 and 3, respectively). Palmitic acid was reported to be active against breast cancer due to its apoptotic potential (Zafaryab et al., 2019). SFAs palmitic acid and stearic acid are more anticancer against liver cells than the USFAs oleic acid and linoleic acid (Jung Min et al., 2012). Stearic and oleic acids inhibited the viability of five human cancer cell lines (two bladder, two testicular and one colon carcinoma cell line). The dilution of oleic acid needed to diminish colony development capability by half was 2.5-6.0-fold greater than that of stearic acid (Beverley et al., 1992).

The main USFAs in both oils produced from canola cultivars (Giza 1 and Gemmiza 1) were oleic acid (43.99% and 44.55% respectively) and linoleic acid (19.22% and 18.72% respectively). Pharmacology networking showed that, the degree of connection of both oleic acid and linoleic acid was (4 and 3, respectively). Oleic acid showed cytotoxic activity on human lung adenocarcinoma (A549) and human prostate cancer (PC-3) cell lines with the IC_{50} of 20 nM and 15 nM, respectively. While no cytotoxic activity was determined on human breast adenocarcinoma (MCF-7), human cervix adenocarcinoma (HeLa), human glioblastoma–astrocytoma (U-87-MG) and human colon colorectal adenocarcinoma (Caco2) cells. It has been proved that oleic acid is characterized by a high cytotoxic activity towards cancer cells due to anti-proliferative activities (Sadi et al., 2022). Conjugated linoleic acids are distinctive PUFAs. They are found in nutrition have anti-carcinogenic activity due to their multiple effects on tumor expansion such as anti-tumor efficiency, anti-mutagenic and anti-oxidant activity (Marko et al., 2021).

3.3. Antimicrobial activity

The results shown in Table (12) indicates that both oil samples inhibit the microbial growth of all tested organisms except of Gemmiza 1 oil against *Pseudomonas aeruginosa*, that is not active. The highest inhibition zone was for Giza 1 oil against *Candida albicans* (27 mm) and *Escherichia coli* (25 mm). Table (13) shows the MIC for both Giza 1 and Gemmiza 1 oil against the tested organisms. The lowest MIC was exerted by Giza 1 oil against both *Candida albicans* (100 µg/ml) and *Escherichia coli* (75 µg/ml).

Table (12). Antimicrobial activity of oil produced from two canola cultivars (Giza 1 and Gemmiza 1) grown under Central North Sinai conditions on different pathogenic microorganisms.

Pathogenic microorganisms	Inhibition zone (mm)	
	Giza 1	Gemmiza 1
Bacillus subtilis	16	15
Staphylococcus aureu	17	12
Escherichia coli	25	16
Pseudomonas aeruginosa	20	-
Candida albicans	27	12

Table (13). Minimum inhibitory concentration MIC (µg/ml) of oil produced from two canola cultivars (Giza 1 and Gemmiza 1) grown under Central North Sinai conditions on different pathogenic microorganisms.

Pathogenic microorganisms	MIC (µg/ml)			
	Giza 1	Gemmiza 1	Imipenem	Fluconazole
Bacillus subtilis	175	150	36	-
Staphylococcus aureu	125	200	34	-
Escherichia coli	75	200	35	-
Pseudomonas aeruginosa	150	-	38	-
Candida albicans	100	200	-	38

Palmitic acid and stearic acid are two SFAs that exhibit antibacterial effect against Gram-positive and Gram-negative bacteria. From pharmacology networking the degree of connection of both palmitic acid and stearic acid was (8 and 4, respectively) for antibacterial activity. The pharmacology networking showed that, the degree of both palmitic acid and stearic acid were (8 and 5, respectively) for antifungal activity. These SFAs were enclosed in liposome vehicle display bactericidal effect against multidrug-resistant *Staphylococcus epidermidis* and Vancomycin-resistant *Enterococcus faecalis* (Giancarlo et al., 2021). Lauric, palmitic, linolenic, linoleic, oleic, stearic and myristic acids are proven to possess proper antibmicrobial activity against bacteria and fungi (Agoramoorthy et al., 2007).

An important semblance of antimicrobial activity is progress suppression or quick destruction of bacteria. Many researches explored the antimicrobial activity of fatty acid derivatives. Fatty acids, such as lauric, palmitic, linolenic, linoleic, oleic, stearic, and myristic acids are active against both bacteria and fungi (Seidel and Taylor, 2004). The most potential SFAs stated as gram-positive antibacterial agent was lauric acid, however linoleic acid is the most possible gram-positive antibacterial agent among the USFAs (Galbraith et al., 1971). Oleic acid exhibits antibacterial action against *Staphylococcus aureus* via destruction of the cell membrane (Yoon et al., 2018). Long-chain USFAs; oleic acid, linoleic acid and linolenic acid, are more effective as antibacterial agents than long chain SFAs; palmitic acid and stearic acid (Sun et al., 2003).

CONCLUSION

The evaluation of canola (*Brassica napus* L.) productivity under salt stress conditions and its impact on oil quality, both chemically and biologically. The experiment was conducted during the 2022/2023 season in the salinity-affected North Sinai region, where two canola cultivars (Giza 1 and Gemmiza 1) were grown to assess yield and oil quality. The results showed significant differences between both the seed and oil yield of the two tested cultivars where the Gemmiza 1 cultivar outperformed the Giza 1 cultivar (1869.84, 1683.60 and 753.55, 670.07 kg/ha, respectively). Chemical

investigation of canola seeds of two cultivars (Giza 1 and Gemmiza 1) indicated that, Gemmiza 1 cultivar outperforms Giza 1 cultivar in raising the acid and peroxide values by 3.28 and 1.19 times, respectively. Also, the percentage of iodine value reduces by 3.69% in Gemmiza 1 cultivar relative to the Giza 1 cultivar. Chemical investigation canola seeds of two cultivars (Giza 1 and Gemmiza1) showed that, the Gemmiza 1 cultivar outperforms the Giza 1 cultivar in raising the acid and peroxide values by 3.28 and 1.19 times, respectively. Under growth conditions in Central North Sinai, the percentage of iodine value reduces by 3.69% in the Gemmiza 1 cultivar relative to the Giza 1 cultivar. The quality and quantity of fatty acids using GC-MS analysis was performed lead to thirteen fatty acids in the oil, and four of them (palmitic, stearic, arachidic, and lignoceric acids) are SFAs. Compared to the Giza 1 cultivar, the Gemmiza 1 cultivar has a 28% higher amount of SFAs. The data demonstrated that the oil from the Giza 1 cultivar had 3.21% more PUFAs than that from the Gemmiza 1 cultivar. Under salinity conditions in Central North Sinai, there were no appreciable differences between the Giza 1 and Gemmiza 1 cultivars regarding the proportion of MUFAs to PUFAs.

Network pharmacology study determined significant natural compounds and their connection to colon cancer antimicrobial and antifungal targets. The cytotoxic activity against colon carcinoma cell line (Caco2), canola oil of both two cultivars (Giza 1 and Gemmiza 1) caused inhibition of cell viability which was correlated to the concentration of the oil (IC_{50} 118.77 ± 2.96 and 110.16 ± 1.34 µg/ml, respectively). Both oil samples inhibit the microbial growth of all tested organisms except of Gemmiza 1 oil against *Pseudomonas aeruginosa*, which is not active. The lowest MIC was exerted by Giza 1 oil against both *Candida albicans* (100 µg/ml) and *Escherichia coli* (75 µg/ml). Concerning the cytotoxic and antimicrobial activity, there were no noticeable differences between the Giza 1 and Gemmiza 1 cultivars under salinity conditions in Central North Sinai.

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تقييم المحصول وجودته كيميائيًا وبيولوجيًا في الكانولا تحت ظروف الإجهاد الملحي

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تهدف هذه الدراسة إلي تقييم إنتاجية الكانولا (*Brassica napus* L.) تحت ظروف الإجهاد الملحي وتأثيرها على جودة الزيت، كيميائيًا وبيولوجيًا. أجريت التجربة خلال موسم ٢٠٢٣/٢٠٢٢ في منطقة شمال سيناء المتضررة من الملوحة، حيث تمت زراعة صنفين من الكانولا (الجيزة ١ والجميزة ١) وتم تقييم المحصول وجودة الزيت. وأظهرت النتائج وجود فروق ذات دلالة إحصائية بين كل من إنتاج البذور والزيت للصنفين المختبرين حيث تفوق صنف الجميزة ١ على صنف الجيزة ١ (١٦٨٣.٦، ١٨٦٩.٨٤، ٧٥٣.٥٥، ٦٧٠.٠٧ كجم/هكتار على التوالي). انخفضت نسبة قيمة اليود بنسبة 3.69٪ في صنف الجميزة ١ مقارنة بصنف الجيزة ١. تبين من نتائج تحاليل الأحماض الدهنية بتقنية GC-Mass إلى وجود ثلاثة عشر حمضًا دهنيًا في الزيت منها أربعة أحماض دهنية مشبعة. وقد أوضحت الدراسة أن زيت الكانولا (الجيزة ١ والجميزة ١) له نشاط المضاد للخلايا السرطانية ضد (سرطان القولون) والجرعة النصف المميتة لكل منهما (١٨٨.٧٧± ٢.٩٦ و ١١٠.١٦ ± ١.٣٤ ميكروجرام/مل، على التوالي). قد تم الاستعانة بنموذج تحليل شبكي فارماكولوجي لبعض المكونات النباتية المهمة مثل حمض اللينولينيك وحمض البالمتيك وحمض الأوليك وحمض الستياريك، والتي كشفت ارتباطها بمستقبلات سرطان القولون. وأن حمض الأوليك وحمض اللينوليك وحمض البالمتيك وحمض اللينولينيك لها ارتباط فعال كضد مضادات الفطريات. توفر هذه الدراسة معلومات قيمة لتقييم إنتاجية الزيت ومراقبة طبيعة الأصناف في ظل الظروف الحيوية والإتجاه المستقبلي لإعتماد الكانولا وجودة الزيت المستخرج كيميائيًا وبيولوجيًا.