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Original article

Anti-bacterial, anti-scavenging and cytotoxic activity of garden cress polysaccharides

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ABSTRACT

Plants polysaccharides are an infinite stock of drug composites with varying pharmacological and biological activities. The present investigation aimed to examine the antibacterial, anti-scavenging and cytotoxic potential of garden cress (GC) polysaccharides. The antibacterial effects *vs Escherichia coli* and as well as *Staphylococcus aureus* of GC polysaccharides were examined by means of agar diffusion assay, minimum inhibitory concentration (MIC), outer and inner cell membrane permeability. Antioxidant potential of the GC polysaccharides were performed by free radical DPPH scavenging, superoxide anion scavenging, hydroxyl radical scavenging, reducing power potential assay, and hydrogen peroxide method. Cytotoxicity potential of GC polysaccharides were evaluated by MTT assay in human cervical (HeLa) and liver carcinoma (HepG2) cell lines. The findings showed that GC polysaccharides MIC were 1.06 and 0.56 mg mL^{-1} against E. coli and S. aureus, respectively. Compared to the standard inhibitor, the GC polysaccharides showed essential inhibitor assays in a very dose dependent approach, and notable actions to scavenge reactive oxygen species (ROS) are also due to the large quantities of hydrophilic polyphenols. The IC_{50} values of all tested parameters were measured against standard ascorbic acid antioxidant agent. The GC polysaccharides diminish the cell viability percentage of HeLa and HepG2 in a concentration dependent manner. GC polysaccharides at a dose of 500 μg ml^{-1} exhibited higher anti-tumor activity in both HeLa (65.33 ± 3.75%) and HepG2 (60.33 ± 3.48%). The findings obtained in this study indicate that GC polysaccharides has antibacterial and has a possible source of natural antioxidant and also has cytotoxic effect on different carcinoma cell lines.

1. Introduction

From prehistoric times medicinal plants are used for their therapeutic qualities. Plants are the paramount sources for phytochemical and chemical ingredients for treatment of different ailments (Petrovska, 2012). Medicinal herbs are an infinite home of drug compounds with diverse biological and pharmacological activities. At present, raw extracts from these medicinal plants have been shown to pay attention to the production and preparation of alternative conventional medicines. (Pan et al., 2013; Yuan et al., 2016). Garden cress (GC) biological name *Lepidium sativum* belonging to the *Cruciferae* family, is a fast growing annual herb that is native to western Asia and Egypt but is extensively used for various medicinal and culinary uses worldwide (Doke, 2014; Sharma SK). GC has been shown to have remarkable preventive functions in various ailments such as high blood pressure, bronchitis, inflammation, arthritis, cancer, liver disorder and hyperglycemia (KM, 2005; Sharma SK). The GC mucilage, also recognized as polysaccharides, is a blend of cellulose and uronic acid, (L)-rhamnose, arabinose, (D)-glucose, galactose, and galacturonic acid are the acid hydrolysis products (Doke, 2014). Polysaccharides are classified as carbohydrate polymeric molecules consisting long monosaccha-

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ride strings. Polysaccharides are classified as polymeric molecules of carbohydrate consisting long monosaccharides strings. Because of their unique properties they serve as key components in the growth and progress of living things (Samavati and Manoochehrizade, 2013). Polysaccharides are also reported to have anti-cancer properties (Gamal-Eldeen et al., 2009; Gao et al., 2005; Thulasi et al., 2010). Several reports have suggested that polysaccharides have ability to inhibit the tumor growth via following mechanisms: the inhibition of tumorigenesis by oral active preparations, apoptosis of tumor cells, inhibition of tumor metastasis, and immunopotentiation activity in combination with anticancer therapy (Zong et al., 2012). Polysaccharides are heterogeneous, and depending on the composition they may have different features from their monosaccharide key components. Polysaccharides may be amorphous in liquids, or even insoluble. Their structural spectrum ranges from linear to strongly branched, including starch, cellulose, glycogen and, chitin (Cumpstey, 2013; Varki et al., 2009).

Antibiotics are widely used as an important means of control and prevention of diseases in people's everyday lives, however, thorough or long term use of antibiotics can help strengthen tolerance against pathogenic organisms (Fonberg, 1971; Palanisamy et al., 2019). Repeated usage of antibiotic medications can induce resistance to the microbes have sparked the search for new antimicrobials or antibiotics (Fair and Tor, 2014). Hence, natural compound with antibacterial properties have a strong potential for production as antimicrobials (Angiolella et al., 2018). Free radical disruption to the human body primarily has three components: damaging the cell membrane; inactivating the serum antiprotease; and allowing the cell mutations to develop and accumulate (Kurutas, 2016). Antioxidant is a chemical capable of slowing certain molecules down or preventing their oxidation (Kurutas, 2016; Phaniendra et al., 2015). Within natural circumstances the antioxidants detoxify the body's reactive oxygen species (ROS) stream. The ROS can quickly target and inflict oxidative harm to various biomolecules like proteins, lipids, lipoproteins, and DNA (Birben et al., 2012; Shahidi and Wanasundara, 1992). Oxidative disruption is a key etiological element of importance in a number of serious medical diseases such as atherosclerosis, high blood sugar, autoimmune, arthritis, and in the ageing cycle (Uttara et al., 2009). Considering the budding attention in oxygen radical science, and the absence of proper treatment for most chronic conditions, antioxidants are necessary to protect against such illnesses. Several research studies have shown that antioxidant intake such as vitamin C diminishes the incidence of cancer and cardiovascular complications (Phaniendra et al., 2015; Tan et al., 2018). The occurrence of chronic diseases may be minimized and the disease stopped either by strengthening the body's natural antioxidant defenses or by incorporating existing nutritional antioxidants (Ardelean et al., 2017; Lobo et al., 2010). The purpose of this analysis was to investigate the yield of GC polysaccharides and to evaluate possible beneficial effects antibacterial, antioxidant and cytotoxic activity.

2. Methods

2.1. Plant materials and bacterial strains

Seeds of GC were bought from a nearby herbal store in Srinagar, India. The seeds were evaluated by Taxonomist Dr. Hilal A. Lone, Cluster University, Srinagar. Using a pestle and mortar before extraction, the seeds were washed with water (distilled), deshelled. Dried and crushed to powder form. The seed powder has been stored hidden from sunshine and clear of moisture in dark containers before further study. *E. coli* strain ATCC-25992 and *S aureus* ATCC-25923 (ATCC, Rockville, USA), were procured from ATCC, USA. All bacteria were incubated in biological oxygen demand (BOD) incubator 5% CO_2 at 37 °C. Bacterial cell cultures were extracted and resuspended with the sterilized 0.9% NaCl to allow the bacterial cell suspension for the antibacterial behavior test. Until usage, the turbidity has been set to a concentration of 1×10^6 CFU mL^{-1}. All other reagents and chemicals used by Panreac Chemicals (Barcelona, Spain) were of analytical grade and were procured.

2.2. Extraction and physicochemical properties of polysaccharides

The extraction of polysaccharides from GC was performed by means of UAE as per the scheme defined by Ahmad et al. (2018). In order to test the main polysaccharides/sugars as previously stated by Karazhiyan et al. (2011), FT-IR assessed the characterizations of molecular and chemical compositions of GC seeds (Karazhiyan et al., 2011)

2.3. Antibacterial activity of GC polysaccharides

2.3.1. Agar diffusion assay

GC polysaccharides anti-bacterial activity *vs S. aureus* and *E. coli* were examined using the process of agar diffusion previously mentioned by (Hassanpour et al., 2018; Wang et al., 2019).

2.3.2. Minimum inhibitory concentration (MIC)

The GC polysaccharide MIC *vs S. aureus* and *E. coli* were analyzed by using double dilution method as previously described by (Wang et al., 2019).

2.3.3. Membrane permeability and cell wall determination

The test organisms (*S. aureus* and *E. coli*) were cultured in 500 mL flask in 100 mL of culture medium. The measurement membrane permeability and cell wall determination by alkaline phosphatase (ALP) and beta-galactosidase activity (β-Gal) detection kit (Bio-Vision, USA) were used as per the manufacturer protocols.

2.4. Antioxidant activity

2.4.1. Assay of reducing power

The reducing power of GC Polysaccharides were checked as per the previously reported procedure (Amir et al., 2011). Improved absorption of the mixture of samples shows increased power reduction. In comparison, ascorbic acid as a standard.

2.4.2. Superoxide anion scavenging activity assay

The scavenging potential of GC polysaccharides against superoxide anion radicals was assayed using previous report of (Amir et al., 2011). The activity of superoxide anions scavenging was measured as per the following Eq. (1):

$$\%\boldsymbol{Inhibition} = \frac{\boldsymbol{Ao} - \boldsymbol{A1}}{\boldsymbol{A0}} \times 100 \quad (1)$$

2.4.3. Hydroxyl radical scavenging activity assay

Fenton reaction was used to evaluate the hydroxyl radical scavenging activity (Amir et al., 2011; Yu et al., 2004). Using a (Shimadzu, UV–Vis) spectrophotometer, the absorbance of the sample mixtures was recorded at 560 nm after 5 m of incubation at room temperature. The scavenging behavior of hydroxyl radicals was measured using Eq. (1).

2.4.4. Scavenging potential of 2, 2-diphenyl-1-picryl hydrazyl radical (DPPH)

The GC polysaccharide extracts free radical scavenging potential was calculated using DPPH (Amir et al., 2011). The absorbance was measured at 515 nm by (Shimadzu UV–Vis) spectrophotometer after 10 m of room temperature incubation. The DPPH activity was recorded using Eq. (1) and ascorbic acid was used as a reference standard.

2.4.5. Hydrogen peroxide scavenging

GC polysaccharide capacity to scavenge H_2O_2 was calculated as per the reported procedure of (Amir et al., 2011; Ruch and Klaunig, 1989). The percentage of GC polysaccharide H_2O_2 scavenging and standard compounds was determined using the above Eq. #1.

2.5. Cytotoxicity assay

2.5.1. MTT assay

Cancer cell lines of human cervical (HeLa, ATCC, Rockville, USA) were maintained in a culture flask in Dulbecco's Modified Eagle Medium (DMEM, Gibco) supplemented with 10% *v/v* of fetal bovine serum (FBS). Cells were incubated at 37 °C in biological oxygen demand (BOD) incubator with 95% of O_2 and 5% of CO_2. The cells were washed with phosphate buffer saline- Ethylene diamine tetra acetic acid (PBS-EDTA) (1x), trypsinised with (trypsin 0.25%), neutralization was carried out with DMEM. Cells were centrifuged at 1500 rpm for 15 min and the supernatant was taken out and remaining pellet was suspended in DMEM for cell culturing. Viability of cells was measured by MTT (3-(4, 5-dimethylthiazol-2-yl)-2, 5-diphenyl tetrazolium bromide) dye. In 96-well plates (1×10^5) cells/ml in each well were transferred and were incubated overnight. The cells were treated with different concentrations (250 and 500 μg mL^{-1}) of GC polysaccharides for 48 h, and MTT (5 mg mL^{-1}) was added, and incubated for 4 h. The medium was aspirated and Dimethyl sulfoxide (DMSO) were added to solubilize the crystals. The optical density (OD) of purple formazan was measured after 5 min of color development by Bio-Rad 680 model colorimeter at 570 nm (Pillai et al., 2017). The untreated samples were also processed under same conditions to serve as assay control. Hepatoprotective activity (in vitro) of GC polysaccharides was evaluated against the liver damage induced by D-GalN in HepG2 cells (ATCC, Rockville, USA). The cells were placed at a density of (1×10^5) cells/well in a 96 well plate supplemented with 100 μl of MEM medium with 10% of FBS in each well. HepG2 cells along with (D-GalN (50 mmol) were treated with different GC polysaccharide extract concentration (250 and 500 μg mL^{-1}) for 8 h and the cell viability was assessed by MTT assay (Yang et al., 2019). Silymarin (100 μg mL^{-1}) was used as the reference standard. The percentage protection was measured to quantify the cell viability assay.

2.6. Statistical analysis

The data analysis of result responses was performed by applying one-way analysis of variance (ANOVA) test ($p \leq 0.05$) using Graph pad software (Version 6, USA).

3. Results and discussion

The objective of the study was to determine the antibacterial, antioxidant and cytotoxic effects of garden cress polysaccharide. Molecular characteristics and chemical compositions using FT-IR specify the main sugars such a as fructose, arabinose, glucose, rhamnose, uronic acid and galactose, which admits the polyelectrolyte existence to the GC seed extract (Karazhiyan et al., 2011).

GC polysaccharide exhibit antibacterial potential *vs* S. aureus and E. coli were studied and described in terms of zone of inhibition (mm). As depicted in Fig. 1, GC polysaccharide antibacterial function *vs S. aureus* and *E. coli* was concentration-dependent (0.25–3.0 mg mL^{-1}), and polysaccharides from GC exhibited enriched antibacterial potential *vs S. aureus* than *E. coli*. The GC polysaccharide inhibition zones toward *S. aureus* and *E. coli* at a dose of 0.25 mg mL^{-1} were found to be 10.39 ± 0.42 and 6.89 ± 0.65 mm, respectively. Once the dose of GC polysaccharide has risen to 3 mg mL^{-1}, the zone inhibitions were 19.65 ± 1.06 mm and 13.91 ± 0.76 mm of *S. aureus* and *E. coli*, respectively. In line with previous reports (Wang et al., 2019) revealed that polysaccharides isolated from *Chaetomium globosum* demonstrated stronger antimicrobial potential *vs S. aureus* than *E. coli*. Another research showed that polysaccharide antimicrobial production from fruits of B. papyrifera toward S. aureus was greater than that of *E. coli* (Han et al., 2016). The GC polysaccharides display stronger antimicrobial potential against *S. aureus* than *E. coli* in present research. GC polysaccharides' MIC against S. aureus and E. coli were 0.56 mg mL^{-1} and 1.06 mg mL^{-1} respectively. The GC polysaccharide is indicated to be a significant and active constituent for food and pharmaceutical businesses (Fig. 1).

A mechanism for antimicrobial activity of a GC polysaccharide that interact with outer and inner cell membrane permeability that diminishes the production of protein synthesis and inhibition of nucleic acids. ALP exists between the cell membrane and the cell nucleus, with β-Gal in the cell membrane. Under normal conditions, their activities cannot be identified in the culture medium. As is evident from Fig. 2A and B, ALP behaviors in the control group culture medium and experiment group were essentially invariable amongst 1 and 6 h, suggesting cell wall stability. While experimental group ALP behaviors grown exponentially between 8 and 10 h, this may be attributed to GC polysaccharides induced cell lysis. Fig. 2C reported that the development of β-galactosidase in the experimental population culture medium displayed a strong growing pattern and that the level of β-glucosidase in E. coli (Fig. 2D) was lower than that in S. aureus. These findings hypothesize that GC polysaccharides may damage inner membrane permeability of *E. coli* and *S. aureus*. A few reports propose that the antibacterial potential of polysaccharides may be identified with rupture the

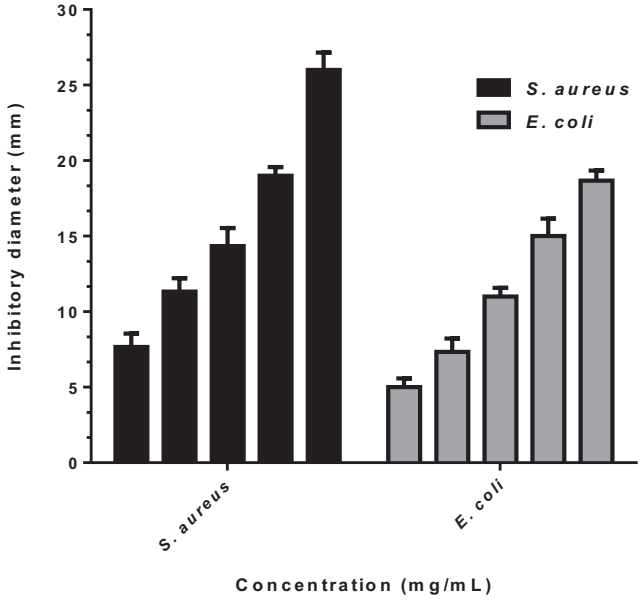

Fig. 1. The inhibitory action of GCP on *E. coli* and *S. aureus* with different concentrations.

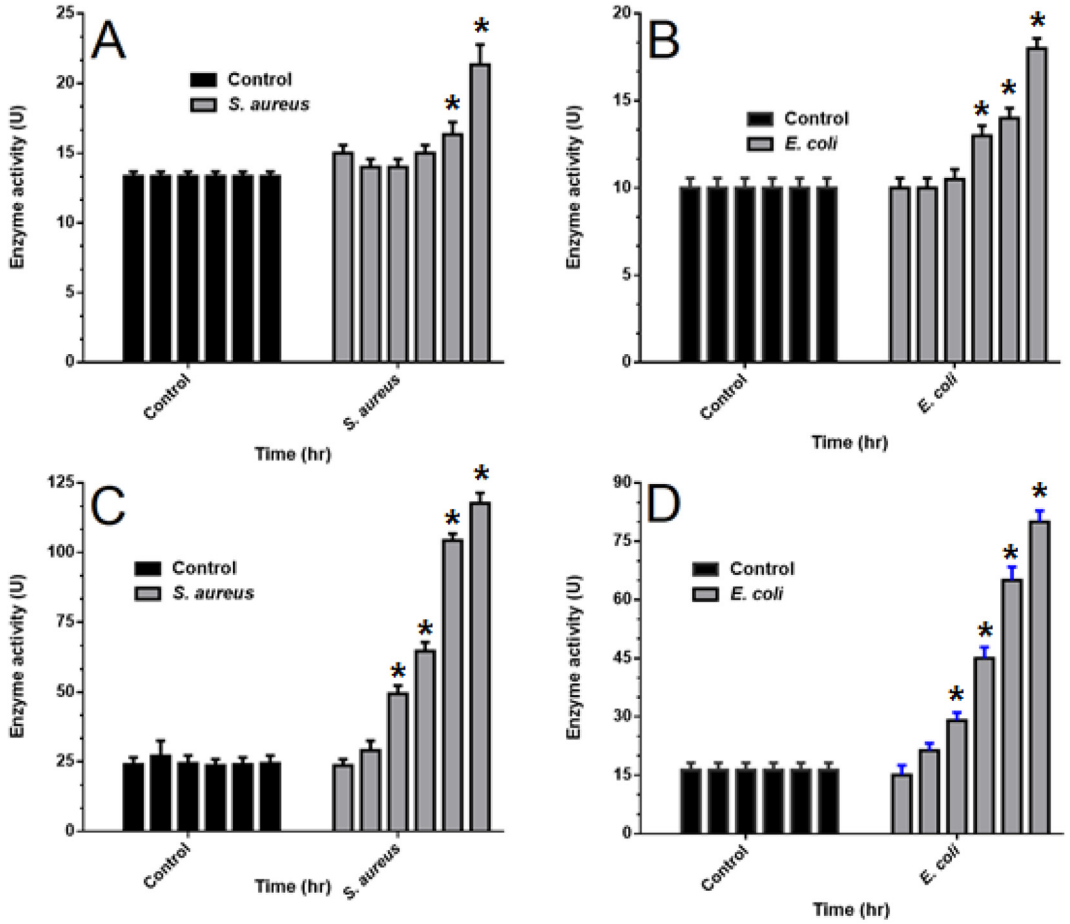

Fig. 2. Alkaline phosphatase activity and β-glucosidase activity in the culture medium of *S. aureus* (A, C) and *E. coli* (B, D) treated with GCP. *In comparison to control group ($p < 0.05$).

outer and inner cell membrane of pathogens (Wang et al., 2019; Zhang et al., 2017).

3.1. Antioxidant potential

Free radicals are continuously produced and can exert severe tissue injury in biological molecules that lead to several diseases (Phaniendra et al., 2015). Various synthetic compounds protect against oxidative damage, consuming natural substances like food supplements could be an alternative solution due to their minima side effects (Lobo et al., 2010; Neha et al., 2019). Several plant extracts have proved effective free radical scavengers. Natural polyphenols exert their useful health effects mainly via their antioxidant potential by reducing the concentration of oxygen, interrupting singlet oxygen, averting beginning of the first chain by scavenging early hydroxyl radicals, attaching metal ion catalysts, and decomposing primary oxidation products to non-radical organisms, and breaking chains to avoid continued extraction of hydrogen from substances (Lobo et al., 2010; Pham-Huy et al., 2008). Natural antioxidants found in plants scavenge our body's toxic, free radicals. Free radicals play a major role in human health and are helpful in the battle against various diseases such as inflammation, cardiovascular disorders, and lung damage (Krishnaiah, 2007). Such free radicals are very uneven, and when the quantity of certain free radicals in the body exceeds it can destroy the tissues and cells may involve multiple diseases. (Neha et al., 2019; Pham-Huy et al., 2008; Shahidi & Wanasundara, 1992). Therefore, naturally occurring antioxidants are required because they can defend the human body from various ailments exerted by free radicals (Krishnaiah D., 2007).

GC polysaccharide reduction power was very powerful, and the extract's power was increased with sample quality. The plant extract was capable of reducing the most Fe^{3+} ions, but to a lesser extent than the regular/standard ascorbic acid. Increase in reaction mixture absorbance suggesting reducing power augmentation (Table 1). It is well known that, during any pathological event and/or aging, such as ischemic reperfusion injury, Superoxide anions cause direct or indirect damage to biomolecules by the formation of H2O2, OH and peroxide nitrite or singlet oxygen. Also superoxide was observed to initiate lipid peroxidation directly (Amir et al., 2011; Pham-Huy et al., 2008). Table 2

Table 1
Reducing power ability of ascorbic acid and GCP extract.

S. No.	Conc. (μg/ml)	Absorbance at 700 nm	Mean ± SEM
Ascorbic Acid			
1	5	0.559, 0.568, 0.574	0.567 ± 0.004
2	10	0.596, 0.607, 0.615	0.606 ± 0.005
3	15	0.648, 0.660, 0.671	0.659 ± 0.006
4	20	0.711, 0.701, 0.719	0.710 ± 0.005
5	25	0.750, 0.761, 0.765	0.758 ± 0.004
GCP Extract			
1	10	0.377, 0.386, 0.398	0.387 ± 0.006
2	20	0.403, 0.414, 0.421	0.412 ± 0.005
3	30	0.448, 0.457, 0.462	0.455 ± 0.004
4	40	0.493, 0.498, 0.515	0.502 ± 0.006
5	50	0.539, 0.548, 0.559	0.548 ± 0.005

Table 2
Superoxide anion scavenging activity of ascorbic acid and GCP extract.

Ascorbic Acid				*GCP Extract*		
S. No.	Conc. (μg/ml)	% Inhibition (Mean ± SEM)	IC_{50}(μg/ml)	Conc. (μg/ml)	% Inhibition (Mean ± SEM)	IC_{50} (μg/ml)
1	5	47.42 ± 1.22	6.76	10	40.46 ± 1.22	31.22
2	10	54.64 ± 1.95		20	45.40 ± 1.01	
3	15	60.92 ± 1.09		30	49.39 ± 1.29	
4	20	67.16 ± 1.07		40	53.79 ± 1.48	
5	25	74.06 ± 1.60		50	58.41 ± 1.58	

presents the superoxide anion radical scavenging potential of GC polysaccharides assessed by PMS-NADH scheme. GC polysaccharides' superoxide scavenging potential has been found to be dependent upon concentration. Additionally, the greater inhibition effect of the extracts of polysaccharides on the formation of the superoxide anions described in this can facilitate them as successful antioxidants. The GC polysaccharide extract half-inhibition concentration (IC_{50}) was 31.22 μg mL^{-1} though the ascorbic acid IC50 value was 6.76 μg mL^{-1}. These results indicate that GC polysaccharides possess strong radical scavenging effect on superoxide (Table 3). Hydroxyl radical is a highly reactive, radical oxygen from the transitional reaction of various hydroperoxide with metal ions. This reaches most biological molecules along with DNA, proteins and polyunsaturated fatty acid (Lobo et al., 2010; Pham-Huy et al., 2008). In a Fenton reaction method, GCP extract showed dose dependent scavenging behavior alongside hydroxyl radical. The ascorbic acid and GCP IC_{50} values were observed to be 29.05 μg/ml and 4.17 μg mL^{-1}, respectively. Table 4 data shows the GCP extract's DPPH radical scavenging activity, compared to the standard ascorbic acid. The IC_{50} values of ascorbic acid and GCP extract were 29.98 μg mL^{-1} and 6.71 μg mL^{-1}, respectively. DPPH exert antioxidant effects thought hydrogen donating ability (Amir et al., 2012; Khan et al., 2013). It was found that the GC polysaccharide extract radical scavenging potential to DPPH was lower than that of ascorbic acid, the results indicated that the polysaccharides had proton-giving properties and may possibly function as free radical scavengers or inhibitors, probably act as key antioxidant (Amir et al., 2011; Khan et al., 2013). GC polysaccharides showed decomposition behavior of H_2O_2 in a dose dependent manner in present findings with an IC_{50} of 24.46 μg mL^{-1}, and for ascorbic acid was 4.77 μg mL^{-1} (Table 5)·H2O2 can directly inactivate a few enzymes and is a slow oxidizing agent, typically through the oxidation of thiol (-SH) groups·H_2O_2 is capable of rapidly crossing cell membranes, interacting with Fe2 + and likely Cu^{2+} ions to express its noxious effects when entering cell (Kurutas, 2016; Lennicke et al., 2015). Therefore, regulation of the amount of H_2O_2 that can be produced is biologically beneficial for cells·H_2O_2 decay by GC polysaccharides can result its antioxidant and free radical scavenging potential (Lennicke et al., 2015).

Table 3
Hydroxyl radical scavenging activity of ascorbic acid and GCP extract.

Ascorbic Acid				*GCP Extract*		
S. No.	Conc. (μg/ml)	% Inhibition (Mean ± SEM)	IC_{50} (μg/ml)	Conc. (μg/ml)	% Inhibition (Mean ± SEM)	IC_{50} (μg/ml)
1	5	50.95 ± 1.06	4.17	10	40.07 ± 1.56	29.05
2	10	59.97 ± 1.79		20	45.24 ± 1.23	
3	15	67.86 ± 1.39		30	50.78 ± 1.06	
4	20	75.85 ± 1.02		40	55.73 ± 1.80	
5	25	83.90 ± 1.17		50	60.63 ± 1.36	

Table 4
Scavenging effect on 2, 2-diphenyl-1-picryl hydrazyl radical of ascorbic acid and GCP extract.

Ascorbic Acid				*GCP Extract*		
S. No.	Conc. (μg/ml)	% Inhibition (Mean ± SEM)	IC_{50} (μg/ml)	Conc. (μg/ml)	% Inhibition (Mean ± SEM)	IC_{50} (μg/ml)
1	5	46.21 ± 1.27	6.71	10	37.76 ± 1.90	29.98
2	10	56.64 ± 2.61		20	44.51 ± 1.78	
3	15	66.12 ± 1.24		30	49.82 ± 3.63	
4	20	75.11 ± 1.45		40	56.03 ± 1.47	
5	25	84.11 ± 1.28		50	61.96 ± 1.74	

Table 5
Scavenging of Hydrogen Peroxide of ascorbic acid and GCP extract.

Ascorbic Acid				*GCP Extract*		
S. No.	Conc. (μg/ml)	% Inhibition (Mean ± SEM)	IC_{50} (μg/ml)	Conc. (μg/ml)	% Inhibition (Mean ± SEM)	IC_{50} (μg/ml)
1	5	50.04 ± 1.32	4.77	10	38.36 ± 1.21	24.46
2	10	55.41 ± 1.35		20	46.10 ± 2.31	
3	15	60.54 ± 2.06		30	54.22 ± 1.49	
4	20	66.42 ± 1.66		40	62.82 ± 2.61	
5	25	71.55 ± 2.31		50	70.60 ± 1.65	

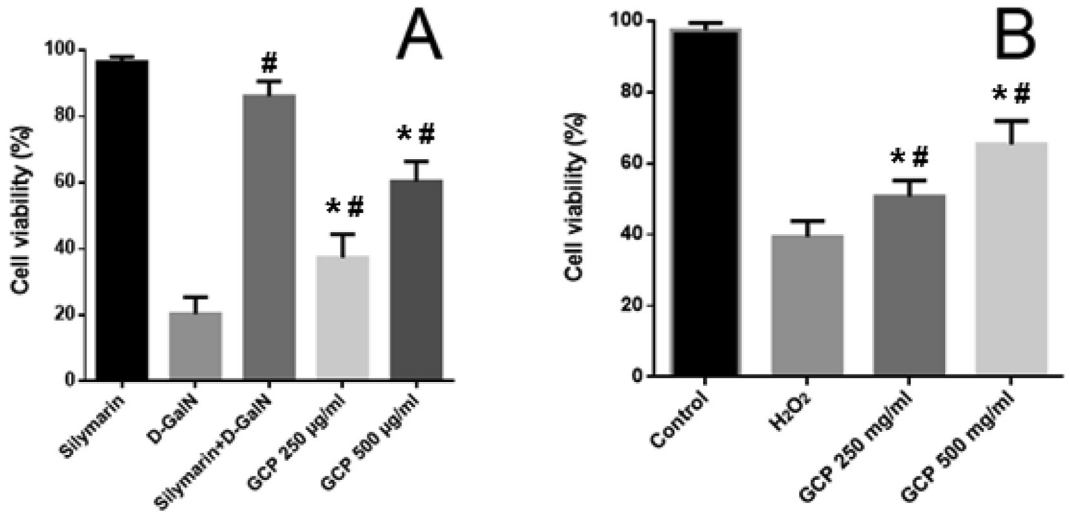

Fig. 3. Cytotoxicity assessment by MTT assay in (A) HeLa and (B) HepG2 cell lines following the exposure of D-GalN and H_2O_2, respectively. Values are expressed in mean ± SEM. *In comparison to control group ($p < 0.05$) and #In comparison to D-GalN and H_2O_2 and $p < 0.05$).

3.2. Cytotoxicity

The anti-tumor property of polysaccharides is shown in Fig. 3A. The GC polysaccharide exhibited growth inhibition as compared to the control samples in HeLa cell lines. The percent inhibition potential was found to be concentration dependent. GC polysaccharides at a dose of 500 μg ml^{-1} displayed higher anti-tumor activity (65.33 ± 3.75%; $p < 0.05$). At two different doses of GC polysaccharide extracts were tested for their hepatoprotective effect on D-GalN induced liver damage in terms of oxidative stress in HepG2 hepatic cell lines. The results exhibited that hepatoprotective activity of GC polysaccharide extract was significant at different dose concentrations ($p < 0.05$). The GC polysaccharide produced maximum hepatoprotective effect at a dose of 500 μg mL^{-1} against D-GalN induced hepatic damage (60.33 ± 3. 48%) Fig. 3B. The potential anti-mutagenic effect of GC polysaccharides can be described by its competence to reduce free radicals. The potential of GC polysaccharides to restrain the in vitro growth of cancerous cell lines (HeLa and HepG2) was measured by MTT assay. The polysaccharides exhibited concentration dependent cytotoxic activity. The potential of polysaccharides and attached protein moieties to modify large number of immune cells may be due to their structural diversity and variation of these biological macromolecules. Polysaccharides hold repetitive structural features unlike proteins and nucleic acids, joined to each other by glycosidic linkages (Zong et al., 2012).

4. Conclusion

GC polysaccharides displayed excellent antimicrobial activity on *S. aureus* relative to *E. coli*, with a minimum inhibitory concentration of 1.06 and 0.56 mg mL^{-1}, respectively. GC polysaccharides reportedly disrupted the inner membrane of the pathogens tested. The GC polysaccharides also displayed strong dose-dependent behavior in all antioxidant evaluates relative to the normal antioxidant, and exceptional performance for scavenging of ROS could be attributable to the large volume of hydrophilic phytochemicals. Moreover, higher anti-mutagenic effects of GC polysaccharides of both cervical and hepatic carcinoma cell lines were observes. The present findings suggest that GC polysaccharides may be investigated as a likely antibacterial, antioxidant and anti-mutagenic agent in the drug and food industries. Additional studies are desirable to investigate the clinical effects of such findings. Consequently, the present results provide a scientific basis for the use of plant extracts in home-made remedies and their possible use in microbial-induced disease care. Further studies may result in their use as natural alternatives to synthetic antimicrobial and cytotoxic. Over the last decade, there has been incredible attention in developing the anti-cancer drugs from polysaccharides. Some in vitro studies seem to be helpful, but *in vivo* studies need to be performed to validate these outcomes. More effective and economical methods to the modification and preparation of polysaccharides remain major challenges and therefore an important field of research is yet to come.

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