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In-vitro antioxidant and selective cytotoxicity of *Garcinia cambogia* and *Garcinia indica* leaf extracts on human kidney cancer cell line

Varsha Jayakar, Vinayak Lokapur, Manjula Shantaram*

Department of Studies and Research in Biochemistry Mangalore University, Jnana Kaveri PG Centre, Chikka Aluvara, Kodagu-571 232, Karnataka, India

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ABSTRACT

Kidney cancer is one among the top 10 cancers. Renal cell carcinoma (RCC) is commonly known kidney cancer. Almost all clinically targeted drugs used in treating RCC have many aftereffects. To overcome this problem with herbal products, the present study is aimed to investigate the preliminary phytochemicals, antioxidants and selective cytotoxicity of aqueous leaves extract of *Garcinia cambogia* (GC) and *Garcinia indica* (GI) on (HEK-293) human embryonic kidney cells and (A498) human renal carcinoma cells. The phytochemical analyses were done using standard protocols. *In-vitro* antioxidant activity was carried out using DPPH, FRAP, and Phosphomolybdenum assay. Anticancer activity on A498 kidney cancer cell line was evaluated by MTT assay and the selectivity index was calculated. Preliminary phytochemical analysis of GC and GI leaf extract divulged the presence of various phyto-constituents. GI extract revealed higher phenolic content while flavonoid content was more in GC extract compared to alkaloids and saponins. Both plant extracts exhibited higher antioxidant capacities based on the test performed.GC and GI leaf extract was selectively cytotoxic *in-vitro* to (A498) human renal carcinoma cells and can be safely used against kidney cancer at 500 μM (GC extract)and 300 μM (GI extract). Selective index for GC and GI extract was15.9 and 2.4respectively. The results indicate that GC and GI extracts are a favourable antioxidant and anti-cancer agents for A498 human renal carcinoma cells. However, further studies to isolate the bioactive compounds responsible for these activities are underway and to explore their molecular mechanism.

*Corresponding Author

Name: Manjula Shantaram
Phone: 9845225882
Email: manjula59@gmail.com

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INTRODUCTION

Cancer holds the top second rank when it comes to deadly disease of today's human population in entire Europe (Liu *et al.*, 2019; Rutz *et al.*, 2019). Among the 10 common death-causing cancers, kidney cancer is one of the debilitating health problems world wide bannering RCC as the most commonly known renal malignancy (Xu *et al.*, 2020; Swiatek *et al.*, 2020). Couple of years ago, 1.8 crore (18 million) new cancer cases were diagnosed globally. RCC constituted around 2% of them; although it is relatively rare compared to other cancers, both incidence and mortality are raising in an alarming rate of 3% per decade. Recent studies report about 3% of adult cancer patients suffer from malignant

kidney tumour (Rutz *et al.*, 2019). The diagnosing techniques for renal cancer have advanced in different methods over the past decades. Surgery is the best therapy for early-stage kidney cancer, but survival rate was hardly 1 year. Specific therapies prevailed for advanced kidney cancer in the clinic since the disease is unsusceptible to chemotherapy (Wang *et al.*, 2019). Researchers all over the world are effectively working on exploring a cost-effective anticancer drug with little or no side effects. In the last fifteen years, more than 1,100 anticancer drugs were developed. Out of which, only few drugs are in favourable state to get the approval from Food and Drug Administration, however, most of them are in clinical trials (Liu *et al.*, 2019; Millimouno *et al.*, 2014) reported that anticancer drugs target growth factors, tumour suppressor proteins, apoptotic proteins, and transcription factors. These drugs available in the market inhibits the cancer development effectively; However, the chances of second cancer or the side effects caused by these drugs are unavoidable. This affects the life style of the cancer-surviving patients. Therefore, it is essential to discover a plant-based drug, which is a potent anticancer drug with low cost, eco-friendly and less after effect.

Since ancient times, natural compounds are used as a source in developing new drugs (Caparica *et al.*, 2020). Cancer, diabetes, rheumatoid arthritis, stroke and coronary heart diseases are some of the fatal diseases, which are generally caused by ROS such as hydroxyl radical, super oxide anion and other exogenous factors. However, medicinal plants possessing antioxidant activity could be a potential lead for curing the above mentioned diseases (Geetha *et al.*, 2020). Research into medicinal plants also provides essential knowledge about nutraceuticals and herbal medicines due to enriched amount of secondary metabolites produced by them. Among many nutraceutical plants, Garcinia species are known to possess nutraceutical properties and proved to be rich sources of compounds with relevant therapeutical properties (Santo *et al.*, 2020). Garcinia species are evergreen polygamous trees and shrubs covering a total of 400 species which covers both the hemispheres of tropical forests. Seventeen out of thirty-five garcinia species are reported from Western ghats, India (Seethapathy *et al.*, 2018). Among which, GC and GI are widely seen in the southern parts of Western Ghats, and are natively marketed as Kodampulior Gummi Gutta and Punarpuli or Kokum, respectively. The bioactive compounds present in GC and GI leaves were reported in our previous report (Jayakar *et al.*, 2020). Based on the phytochemicals present, the current study was carried out in order to evaluate the cyto-toxicity using MTT assay. Literature reveals that anti-cancer activity of G. indica against various types of cancers such as gall bladder (Duan *et al.*, 2018), human breast cancer (Ahmad *et al.*, 2010), prostate, colon, pancreatic, and leukemia (Saadat and Gupta, 2012), whereas anti-cancer activity of G. cambogia against various types of cancer like colon, adenocarcinoma (Banu and Ramakrishnaiah, 2018) colorectal, cervical cancer (Hart and Cock, 2016) have been reported. Despite the claims and the use of GC and GI for treating various cancers, little is known and documented. Hence, the present study is aimed to explore the in vitro antioxidant activity and to analyse preliminary phytochemicals along with their cytotoxic effects on Kidney cell lines. This is the first report, as per our knowledge, in evaluating the possible beneficial interaction of aqueous extracts on viability of the human renal cancer cell line (A498) and human embryonic renal cell line (HEK 293) by using MTT assay as an *in-vitro* technique as well as determining its selective cytotoxicity.

MATERIALS AND METHODS

Authentication of selected plants

The leaves of G. cambogia (GC) and G. indica (GI)were collected from the Central Horticultural Experiment Station, Chettalli, Kodagu district of Karnataka, India in the month of September 2020. Both species of Garcinia were identified and authenticated by Principal Scientist at PND Herbarium, Mangalore, Karnataka, India vide letter no. SKPND: CR: 113: Herbarium Collection/19-20. The herbarium is kept at PND Herbarium, Mangalore as G. indica (accession no. 2286) and G. cambogia (accession no. 9743) for further reference.

Preparation of plant extracts

Fresh plant leaves of GC and GI were cleansed in running tap water followed by deionised water and shade dried. The air-dried leaf samples were crushed into a coarse powder using mixer grinder. The powdered sample was stored in airtight brown bottle at 4°C till further use.50 grams of shade-dried leaves were crudely powdered before mixing with 500ml of double distilled water and kept in a shaker incubator for 24 hrs.

Temperature was set to 37°C and the incubator was set to 150 rpm. Muslin cloth was used to filter the extract and then by Whatman no. 1 filter paper. The filtrate was evaporated in hot air oven at 50°C till dryness and residue was scrapped and stored at 4°C until further use.

Phytochemical screening

The aqueous crude extracts of GC and GIleaves were separated using water to ensure obtaining bioactive constituents, which were qualitatively, screened for secondary metabolites like phenols, alkaloids, saponins, tannins, flavonoids and glycosides using standard procedures (Lokapur *et al.*, 2020).

Total alkaloids content assay

The total alkaloid content present in GC and GI leaves extract was determined using standard protocol (Lokapur *et al.*, 2020). The alkaloid content was expressed as mg /100 g.

Total saponin content assay

Saponins present in the extracts were quantitatively determined using (Nahapetian and Bassiri, 1975). The saponin content was calculated in percentage.

Total phenolic content assay

The amount of total phenol content (TPC) present inboth plant extracts was quantitatively determined using Folin-Ciocalteu's colorimetric method (Noreen *et al.*, 2017). Gallic acid was used as a standard while TPC was expressed as mg/g Gallic acid equivalent (GAE).

Total flavonoids content assay

The quantity of flavonoids present in the extracts were quantified using the aluminium chloride assay (Iqbal *et al.*, 2015).

The total flavonoid content is expressed as mg/g quercetin equivalents of the extract.

In vitro methods to determine antioxidant activity

Ferric ion reducing antioxidant power (FRAP) assay

FRAP assay was used to determine the total antioxidant power of the extracts. The FRAP assay was performed according to Benzie and Strain (1996) with slight medications. Aqueous plant extract of GC and GI in varied concentrations ranging from 100μg to 500μg /ml were mixed with 2.5 mL of 0.2 mM phosphate buffer (pH 7.4) and 2.5 mL of potassium ferricyanide [1% weight/volume (W/V)].

Temperature was set to 50°C and the resulting solution was incubated for 20 minutes. Later 2.5 mL of TCA (10% W/V) was added and centrifuged for 10 minutes (3000 rpm). Then, 2.5 mL of deionised water was added followed by 0.5 mL of ferrous chloride (0.1% W/V).

Finally, the optical density was measured at 700 nm. A positive reference standard, ascorbic acid, was used to compare the antioxidant property of GC and GI extracts.

Phosphomolybdenum (PM) assay

PM assay was used to estimate the total antioxidant activity using the standard procedure (Ghagane *et al.*, 2017). Aqueous leaf extract of GC and GIin different concentrations ranging from 100μg to 500μg/ml were added to each test tube individually containing 3 mL of distilled water and 1 mL of molybdate reagent solution. These tubes were kept incubated at 95°C for 90 minutes. After incubation, they were maintained at room temperature for 20-30 minutes and the optical density was measured at 695 nm. Ascorbic acid was used as the positive standard reference.

2, 2-Diphenyl-1-picrylhydrazyl radical scavenging ability (DPPH) assay

Free radical scavenging effect of aqueous plant extract was determined using the 2-diphenyl-1picrylhydrazyl (DPPH) with slight modifications put forward by (Brand-Williams *et al.*, 1995). In brief, the concentrations (100- 500ug/ml) of extracts were prepared. 1 mL of DPPH solution (0.004% prepared in ethanol) was treated with 1 mL of aqueous leaf extracts and standard ascorbic acid solution separately. The mixture was left for incubation in the dark under room temperature for 30 minutes and the optical density was measured at 517 nm. The extent of DPPH-purple decolourization to DPPH yellow confirmed the scavenging efficiency of the extract. Higher antioxidant activity was observed as the optical density of the reaction mixture was decreased. Scavenging activity was calculated using the following formula:

$$DPPH\ scavenging\ activity\ (\%) = A_C - A_T/A_C \times 100$$

A_C - the absorbance of the control reaction (1 ml of ethanol with 1 ml of DPPH solution);A_T- the absorbance of the test sample.

The results were analyzed in triplicates. The IC_{50} value indicates the required sample concentration to inhibit 50% of the DPPH free radical.

In-vitro cytotoxicity assay

Culturing of cell lines

The human renal carcinoma cells (A498) and Human embryonic kidney cells (HEK 293) were acquired from the NCCS, Pune, India. Cell lines were maintained using Dulbecco's Modified Eagle Media (DMEM, Invitrogen, USA) with low glucose and supplemented with 10% Foetal Bovine Serum (FBS, Invitrogen, USA). Antimycotic 100X solution were added to the medium to prevent bacterial contamination. The medium with cell lines was maintained in a humidified environment with 5% CO_2 at 37

°C. Cells were detached by treatment with trypsin-EDTA after reaching 80% confluency, and reseeded in fresh media.

Treatment groups

The cytotoxicity activity of the aqueous extracts of GC and GI was evaluated using 3-(4, 5-dimethylthiazol-2-yl)-2, 5-diphenyltetrazoliumbromide (MTT) assay by following Tangjitjaroenkun *et al.* (2021) with slight modifications. Briefly, A498 and HEK293 cells were seeded at a density of 1 $\times 10^4$ cells per well in 96-well flat-bottom micro plate and controlled at 37ºC in 95% humidity and 5% CO_2 humidified atmosphere for overnight before the treatment. These cells were treated with a varied concentration ranges of GC and GI extracts (100-500μg/mL) followed by incubation for 48 hours respectively, which was the standard treatment time of the extracts in each of the cell lines. Cisplatin was used as a standard drug to compare the effects induced for HEK293 human embryonic kidney cell and A498 human kidney cancer cell lines. The current study was carried out with the treatment groups in the following set up. Negative control: only cells. Positive control: cells + cisplatin. Test groups: cells+ aqueous GC extract; and cells+ aqueous GI extract; same procedure was followed for human embryonic kidney cell (HEK-293) line.

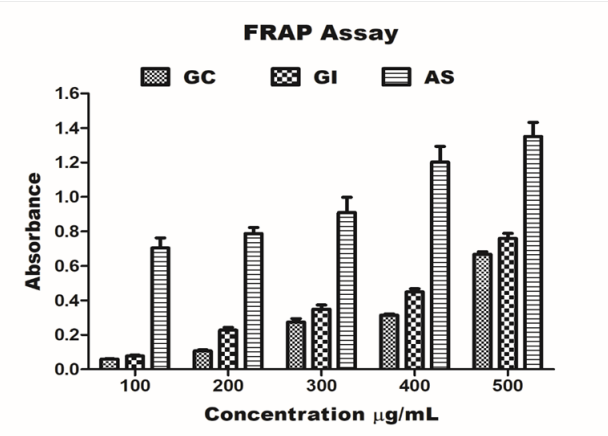


Figure 1: FRAP activity of aqueous extract of GC and GI. AS: Ascorbic acid.

MTT cell viability assays

After 48h incubation, the images were captured using phase contrast microscopy. During successive follow up, cells which were attached to the well were removed by trypsinization and the wells were washed two times with phosphate buffered saline (pH 7.4) without serum and 20 μL of the MTT staining solution was added to each well and further incubated at 37ºC for next 4 hrs. Formazan crystals

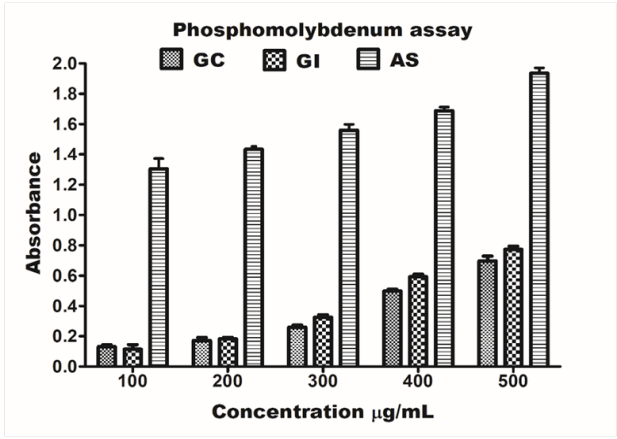


Figure 2: PM assay of aqueous extract of GC and GI. AS: Ascorbic acid.

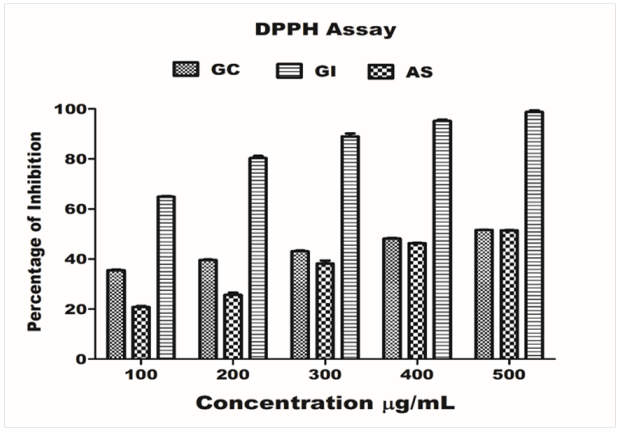


Figure 3: DPPH scavenging activity of aqueous extract of GC and GI. AS: Ascorbic acid.

were dissolved by adding DMSO (100 μL) to each well, and optical density was quantified spectrophotometrically at 570 nm against the control using micro plate reader (Bio-Rad, California, USA). Experiment was carried out in triplicates and the percentage of the average was taken into final consideration. The survival percentage was plotted against control (untreated) group using the following equation,

$$Viable\ cells\ (\%) = (Mean\ OD\ of\ test\ compound / Mean\ OD\ of\ Negative\ control) \times 100$$

Selectivity index (SI)

The A498 and HEK293 cells were used to measure the SI. The SI, which represents the cytotoxic selectivity (i.e. drug safety) for both plant extracts was calculated using the following formula (Ogbole *et al.*, 2017).

SI = IC_{50} calculated for normal/ IC_{50} calculated for cancer

The SI values > 2 is considered as high selectivity (Machana *et al.*, 2011; Prayong *et al.*, 2008).

Table 1: Preliminary phytochemical analysis of aqueous extracts of GC and GI

Phytochemical tests	GC	GI
Tannins	-	-
Saponins	+	+
Flavonoids	+	+
Alkaloids	+	+
Glycosides	+	+
Cardiac glycosides	+	+
Terpenoids	+	+
Steroids	+	-
Phenols	+	+
Resin	-	-

+:positive;- : negative

Table 2: Quantitative analysis of aqueous extracts of GC and GI

Secondary Metabolites	GC	GI
Total alkaloid (mg/g)	0.243±0.002	0.059±0.003
Total saponin (mg/g)	1.323±0.025	1.423±0.030
Total Phenol (mg GAE/g extract)	1.05±0.49	3.0±0.47
Total Flavonoid (mg QE/g extract)	2.35±0.49	2.21±0.33

Values are mean of triplicate determination (n =3) ± standard deviation;
GAE–Gallic acid equivalents; QE-Quercetin equivalents.

Table 3: Percentage inhibition of DPPH radical scavenging activity of GC and GI

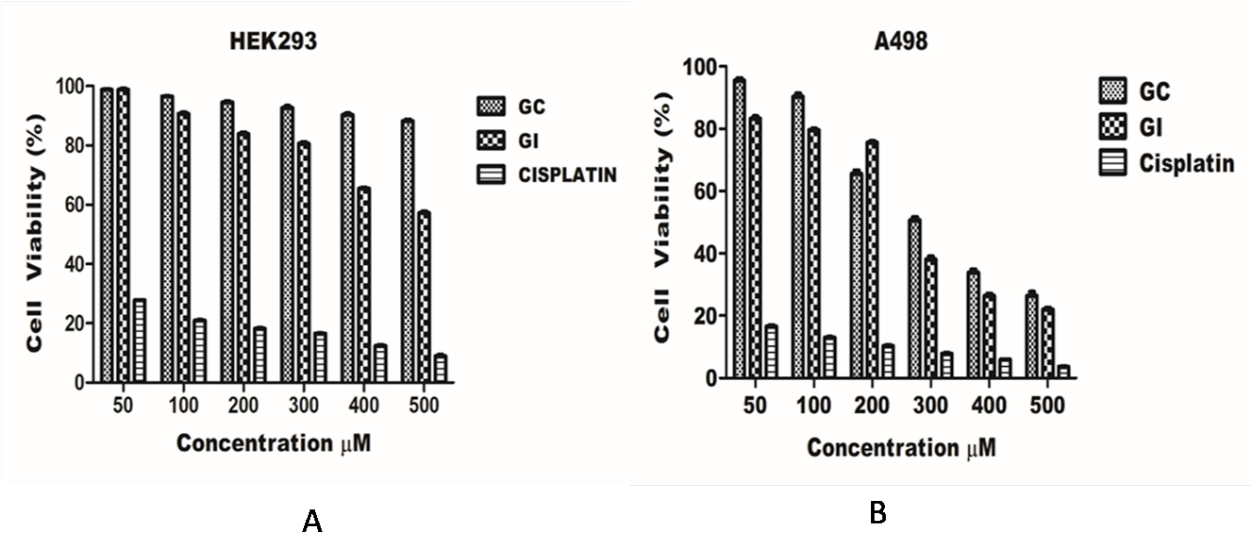
Concentration (mg)	Percentage of Inhibition		
	G. cambogia extract	G. indica extract	Standard Ascorbic acid
100	35.52±0.43	20.78±0.78	64.90±0.50
200	39.61±0.35	25.68±1.57	80.31±1.52
300	43.08±0.57	38.18±2.07	89.03±2
400	48.16±0.50	46.31±0.29	95.15±1
500	51.60±0.21	51.49±0.14	98.78±1
IC_{50} value	493.7	485.1	68.68

The values are expressed as mean ±SD, n = 3. Results were analyzed using descriptive statistics.

Table 4: IC_{50} and R^2 values of aqueousleaf extracts of GC, GI and Cisplatin on two renal cell lines by MTT

Compound	Concentration	HEK293	A498	Selectivity index (SI)
Aqueous G. cambogia extract	IC_{50} (mg/mL)	ND	291.2	15.93
	R^2	0.9661	0.9938	
Aqueous G. indica extract	IC_{50} (mg/mL)	628.2	255.9	2.45
	R^2	0.9618	0.8939	
Cisplatin	IC_{50} (mg/mL)	6.676	2.832	2.36
	R^2	0.9272	0.9322	

HEK293- Human embryonic kidney cells
A498- Human kidney cancer cell
SI values > 2 was considered as highselectivity.
ND - IC_{50}> 1000 μg/mL is consideredas not- determined (ND).



A B

Figure 4: Cytotoxicity of aqueous extracts of GC and GI compared with standard drug on HEK-293 and A498 kidney cancer cell line.

Statistical analysis

The study was carried out in triplicates and results were expressed as mean ± SD. Mean, standard deviation, variation and level of statistical significance between groups was analyzed using descriptive statistics. Percent inhibition of cell growth was analyzed and $P < 0.05$ and $P < 0.01$ was considered statistically significant.

RESULTS AND DISCUSSION

Qualitative and quantitative phytochemical analysis

Results of preliminary phytochemical analysis obtained from aqueous extracts of GC and GI revealed the presence of diversity of secondary metabolites. Steroids were present only in aqueous GC extract while tannin and resin were absent in both extracts. Contrary to this, alkaloids, flavonoids, saponins, glycosides,cardiac glycosides, terpenoids and polyphenols were present in both extracts (Tables 1 and 2). Existence or lack of bioactive compoundsmay be due to the solvent medium used for extraction. These bioactive components are responsible for biological activities (Hemshekhar *et al.*, 2011). Both GC and GI showed highest quantity of flavonoid and phenolic compounds compared to saponins, and alkaloids. However, GC extract contains highest flavonoids (2.35±0.49) and saponin as second highest level followed by phenolic compounds and alkaloids whereas GI contain highest phenolic compounds (3.0±0.47) followed by flavonoids, saponins and alkaloids. Alkaloids were found to be the least in both plant extracts (Table 2).

Garcinia species are widely used for their phenolic compounds such as flavonoids, phenolic acids, xanthones, biflavonoids and benzophenones (Han *et al.*, 2007). Flavonoids were known to possess many biological properties like anti-inflammatory, antimicrobial, enzyme inhibition, anti-allergic, antioxidant and anti-tumour activity (Harborne and Williams, 2000). Even though both the Garcinia species showed least alkaloid content, its presence in plants can be used in medicine as aesthetic agents (Hong and Wrolstad, 1990). Saponins are traditionally used as detergents, and pesticides as well as advantageous health effects apart from their industrial applications as foaming and surface-active agents (Shi *et al.*, 2004).

Antioxidant activity of plants

As health-related risks are increasing at an alarming rate, evaluation of plant-derived antioxidants is of immense importance in today's context. Many bioactive components like phenols and flavonoids acts as sources of antioxidants and perform scavenging activity (Diplock, 1997). In order to obtain the complete potential of antioxidants from any source, it is wise to utilise different assays while estimating the total antioxidant activity (Sethi *et al.*, 2020). In the present study, FRAP, PM and DPPH were used to evaluate in vitro antioxidant capacity of aqueous extracts of GC and GI.

FRAP assay

In the current study, the antioxidants present in the extracts would result in the reduction of ferri cyanide Fe^{3+} to ferro cyanide Fe^{2+} by contributing an electron, which was measured spectrophotometrically at 700 nm. Standard ascorbic acid and aque-

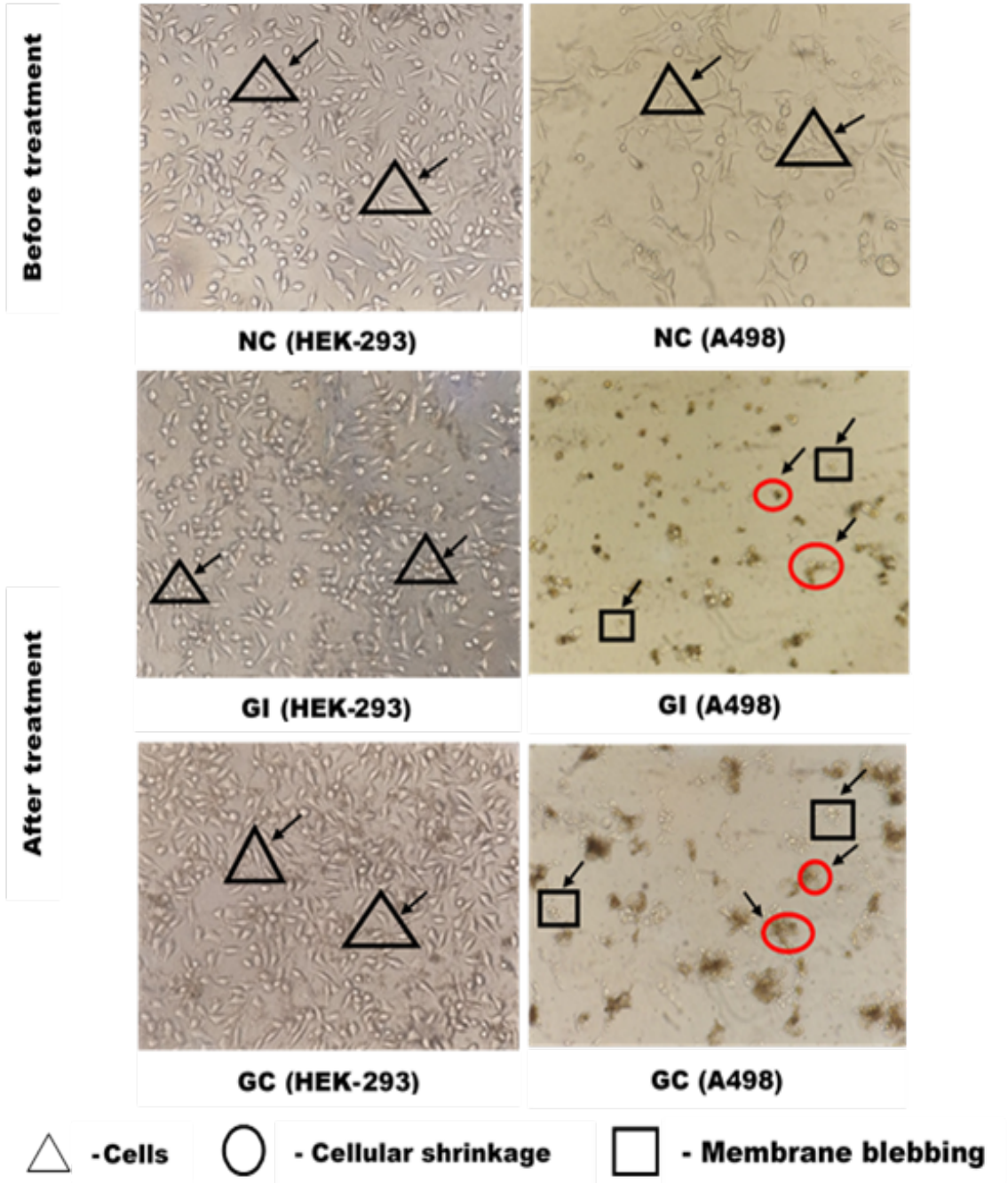


Figure 5: Morphological changes showing inhibition of A498 and HEK-293 cell line for 48 h. CS: Cellular shrinkage; BL: Membrane blebbing (Magnification for A498 was 40X and HEK-293 was 20X).

ous leaf extracts of GC and GI were subjected to FRAP assay. The experimental data revealed that the aqueous crude extract of GI showed stronger FRAP activity compared to GC (Figure 1). In this Data is expressed as mean ± SEM (n=3). Statistical significance was assessed using one way ANOVA (* p <0.05) as compared to standard group.

PM assay

Phosphomolybdenum method was selected to analyse the total antioxidant activity of the sample. It is a colorimetric method, whichhelps in measuring the reduction of Phosphate-Mo (VI) to Phosphate-Mo (V) by the sample and eventually development of a bluish green coloured Phosphate-Mo (V) complex (Prieto *et al.*, 1999). In the current study, Phosphomolybdenum assay showed better result in aqueous crude extract of GI extract compared to GC (Figure 2). In this Data is expressed as mean ± SEM(n=3). Statistical significance was assessed using one way ANOVA (* p <0.05) as compared to standard group. For FRAP and PM assay, both the

plants exhibited higher activity with increasing concentration compared to standard ascorbic acid.

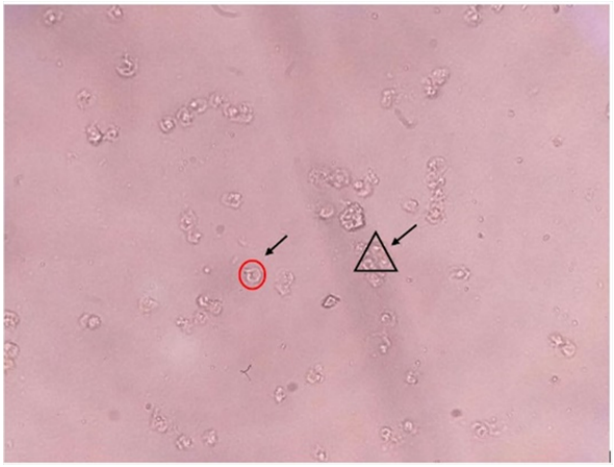


Figure 6: Morphological changes of Standard drug cisplatin on A498 kidney cancer cell lines.

DPPH assay

In the current study, the varied concentrations of GC and GI leaf extracts were subjected to 2-diphenyl- 1picrylhydrazyl free radical scavenging assay. Ascorbic acid was used as a standard drug to compare the antioxidant capacity of the extracts. Aqueous crude extract of GC showed stronger antioxidant activity than GI (Figure 3). In this Each value is expressed as means ± standard deviation. Concentration (μg/ml) take non x-axis and percentage inhibition taken on y-axis. When the antioxidant activities of both the plants were compared, GC showed the IC_{50} value of 493.7 μg/mL and GI showed IC_{50} value of 485.1 μg/mL with R^2 value of 0.9430 and 0.9536 respectively (Table 3). Even though both the crude extracts of Garcinia species showed good antioxidant activity when compared to standard, GC showed slightly better result with consistency at different concentrations while GI gradually increased along with the concentration. The difference in the IC_{50} value and potential DPPH radical scavenging activity observed in this study may be due to the phytochemical components present in the extracts. Our observation is in agreement with the studies of (Izuegbuna *et al.*, 2019).

Cytotoxicity

Plant extracts are the best sources to evaluate the anticancer activity with least or no side effects for safe diagnosis. It helps in identifying the deep-rooted toxicity of the plant and the effects of critical overdose. It could also help in screening possible cytotoxic properties of GC and GI leaf extract (Ghagane *et al.*, 2017). In the current study, the MTT assay was used to evaluate the influence of aqueous extract (0–500 μM; 48 h) treatment on the cell viability of two kidney cell lines, HEK293 human embryonic renal cells and A498 human renal carcinoma cells. It is one of the most commonly used in vitro model system to evaluate the cytotoxic effects of many toxic substances and plant extracts against cancer cell lines (Sharif *et al.*, 2017).

The viability of A498 cells decreases with increase in concentration of the aqueous extracts. Only viable cells have the ability to reduce MTT tetrazolium into a coloured formazan product. The cytotoxic activity was expressed as percentage of cell viability in Hek-293 and A-498 cell lines when compared with the control and both the plant extracts revealed more cytotoxicity towards cancer cell line A498. Concentration in the range of 100 -500 μg/ml and 100 – 500 μM for aqueous extracts of GC and GI extract and Cisplatin (control) respectively were used for the study. Both plants showed no cytotoxic effect towards non-cancerous HEK-293cell line. To be precise, in GC extract, 500 μM did not show significant cytotoxic effects in HEK-293 cells but in A498 cells, a drastic decrease in cell viability was observed. Similarly, GI extract (upto 300 μM) did not show cytotoxicity in HEK-29. However, in A498 cells, a remarkable downfall in cell viability was observed (Figure 4 a,Figure 4b). In this Data is expressed as mean ± SEM (n = 3). Statistical significance was determined using one-way ANOVA (* $p < 0.05$, ** $p < 0.01$)as compared to standard and control group.

The effect produced by the extracts is comparable to that of the standard drug cisplatin, which is commonly, used in the treatment of renal carcinoma. The results exposed morphological changes and cellular shrinkage resulting to cell death caused by the extracts in the renal cancer cell lines (Figures 5 and 6). In Figure 6, CS: Cellular shrinkage; BL: Membrane blebbing (Magnification for A498 was 20X).

The survivability of cells to the leaf extract of GC, GI and Cisplatin was characterized by IC_{50} and R^2 values (Table 4). In vitro growth inhibition effects was observed in the kidney cancer cell line (A498), while there was no effect on the growth of normal cells (HEK-293). Such selective effects were incubation time and concentration dependent. All the extracts were evaluated in triplicates with respect to concentration (100, 200, 300, 400, 500μg/ml) by serial dilution. Higher concentrations, 500 μg/ml of both plant extracts were the most effective in producing growth inhibition. However, the pure standard Cisplatin drug showed significant inhibition on the cancer cell lines. The results confirmed the differential effect induced by the extracts and cisplatin in A498 and HEK-293 cell lines.

When the concentration of the extract was

increased, it was observed that there was rapid decrease in cell-cell contact and cell proliferation. It indicates that the cytotoxic effect gradually increases with increase in the concentration. SI values were also calculated for both the extracts on renal cell lines and compared to those calculated for cisplatin (Table 4). The highest SI values calculated for aqueous extract of GC was 15.93 and GI extract was 2.45. The SI values calculated for Cisplatin for renal cell line was low (2.36), indicating the superiority of GC and GI extract on the cancer cell line compared to Cisplatin. Based on the low IC_{50} value and high SI values for both extract in these cells suggest GC and GI extract as a promising therapeutic candidate in patients with renal cancers. Higher the SI value, the more selective it is and SI values less than 2 indicate general toxicity (Badisa *et al.*, 2009). Gleaned from the results, it can be inferred that compared to Cisplatin, a common chemotherapy drug, GC and GI extracts are better candidates for growth suppression of renal cell lines with SI values > 2.

CONCLUSION

It was observed that the aqueous extract of GC and GI contains a wide variety of bio active components that possess strong antioxidant capacity based on the experiments performed which gives a scientific evidence to conduct further studies. The present *in-vitro* anti-cancer activity exposed the abilities of GC and GI extract as a curative agent for cancer treatment as it not only has a highly potent activity at lower concentrations but also exhibits a high degree of selectivity in kidney cancer cells. This activity may be due to the presence of bio active compounds and antioxidants. Our results reveal that aqueous leaf extract of GC and GI displays cytotoxic effects on A498 human renal carcinoma cells at 500 μM and 300 μM respectively, it may be safely used against kidney cancer since, at this concentration; no significant effect was observed in normal renal cells. Our results suggest GC and GI extracts are attractive option for pharmaceutical companies as a potential agent for the management of human cancer. However, further studies to isolate the secondary metabolites responsible for these activities are underway and to explore their molecular mechanism.

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Conflict of Interest

The authors declare that they have no conflict of interest for this study.

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