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Original Article

Cytotoxicity of iodine-131 radiopharmaceutical in tumor and non-tumor human cells and radioprotection by integral juices of *Vitis labrusca* L.

Citotoxicidade do radiofarmacêutico iodo-131 em células humanas tumorais e não tumorais e radioprotecção por sucos integrais de *Vitis labrusca* L.

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Abstract

Iodine-131 (I-131) radioisotope it causes the formation of free radicals, which lead to the formation of cell lesions and the reduction of cell viability. Thus, the use of radioprotectors, especially those from natural sources, which reduce the effects of radiation to healthy tissues, while maintaining the sensitivity of tumor cells, stands out. The objective of the present study was to evaluate the cytoprotective/radioprotective effects of whole grape juices manufactured from the conventional or organic production systems, whether or not exposed to ultraviolet (UV-C) light irradiation. The results showed that I-131 presented a cytotoxic effect on human hepatocellular cells (HepG2/C3A) at concentrations above 1.85 MBq/mL, after 24 and 48 hours of treatment, though all concentrations (0.0037 to 7.40 MBq/mL) were cytotoxic to non-tumor human lung fibroblast (MCR-5) cells, after 48 hours. However, grape juices (10 and 20 µL/mL) did not interfere with the cytotoxic effect of the therapeutic dose of I-131 on tumor cells within 48 hours of treatment, while protecting the non-tumor cells, probably due to its high antioxidant activity. In accordance with their nutraceutical potential, antioxidant and radioprotective activity, these data stimulate in vivo studies on the use of natural products as radioprotectants, such as grape juice, in order to confirm the positive beneficial potential in living organisms.

Keywords: human lung fibroblast cells (MRC-5), human liver tumor cells (HepG2/C3A), radioprotection, MTT test, whole grape juice.

Resumo

O radioisótopo iodo-131 (I-131) causa a formação de radicais livres, que levam à formação de lesões celulares e redução da viabilidade celular. Assim, destaca-se a utilização de radioprotetores, principalmente de origem natural, que reduzem os efeitos da radiação nos tecidos saudáveis, mantendo a sensibilidade das células tumorais. O objetivo do presente estudo foi avaliar os efeitos citoprotetores/radioprotetores de sucos de uva integral fabricados em sistemas de produção convencional ou orgânico, expostos ou não à radiação ultravioleta (UV-C). Os resultados mostraram que o I-131 apresentou efeito citotóxico nas células hepatocelulares humanas (HepG2/C3A) em concentrações acima de 1,85 MBq/mL, após 24 e 48 horas de tratamento, embora todas as concentrações (0,0037 a 7,40 MBq/mL) fossem citotóxicas para células de fibroblasto de pulmão humano não tumoral (MCR-5), após 48 horas. No entanto, os sucos de uva (10 e 20 µL/mL) não interferiram no efeito citotóxico da dose terapêutica de I-131 nas células tumorais em 48 horas de tratamento, protegendo as células não tumorais, provavelmente devido ao seu alto poder antioxidante. atividade. De acordo com seu potencial nutracêutico, atividade antioxidante e radioprotetora, esses dados estimulam estudos *in vivo* sobre o uso de produtos naturais como radioprotetores, como o suco de uva, a fim de confirmar o potencial benéfico positivo em organismos vivos.

Palavras-chave: células de fibroblastos de pulmão humano (MRC-5), células tumorais de fígado humano (HepG2/C3A), radioproteção, teste MTT, suco de uva integral.

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1. Introduction

Iodine-131 (I-131) is a radioisotope used mainly in the diagnosis and treatment of the thyroid gland (Béron and Wémeau, 2020; Hailan and Yassin, 2021), but also has diagnostic applications in the labeling of antibodies and the adrenal gland, in the treatment of ganglion, lung or bone metastases (Sapienza et al., 2005) and the treatment of glioblastoma tumors (Koosha et al., 2021) and hepatocellular carcinoma (Boucher et al., 2007; Trall and Ziessman, 2003; Costa and Sapienza, 2012).

Hepatocellular carcinoma or primary liver cancer is a malignant epithelial tumor that arises from parenchymal liver cells and is one of the malignant diseases that affect a large number of individuals worldwide (Ahmadzadehfar et al., 2011). Its treatment is performed with I-131-lipiodol, which is prepared by replacing the iodine atom of the lipiodol molecule with its radioactive isotope I-131, which has a half-life of 8.06 days and releases beta particles and gamma radiation during its decay (Trall and Ziessman, 2003; Ahmadzadehfar et al., 2011; Costa and Sapienza, 2012).

The use of radiopharmaceuticals in carcinoma treatments is based on the fact that when they are present, in sufficient quantity in the tissue or organ, they can cause cell destruction and prevent the formation of new tissues, mainly by excitation and ionization, which may alter cell molecules chemically and result in their death (Fischer et al., 2018; Asadian et al., 2020; Nouailhetas et al., 2021). Besides, ionizing radiation may cause swelling or damage of the nucleus and cell structure as a whole, increased cell media viscosity, increased permeability, cell membrane damage, or cell cycle arrest (Fischer et al., 2018), induction of apoptosis, necrosis and autophagy, as well as organ fibrosis, atrophy and inflammation (Yun and Wang, 2017). However, these effects may also affect healthy cells (Murayama et al., 2021) and pulmonary complications have been evidenced in patients undergoing I-131-lipiodol due to tumoral arteriovenous permeation resulting in migration of the molecule to the pulmonary circulation (Ahmadzadehfar et al., 2011; Costa and Sapienza, 2012).

Consequently, the use of radioprotectants stands out. These compounds are administered together with irradiation and reduce their effects on healthy tissues, maintaining the sensitivity to radiation damage to tumor cells (Painuli and Kumar, 2016; Kuruba and Gollapalli, 2018). Radioprotectors, due to their antioxidant activities and the reduction of oxidation, prevent radiation-generated free radicals from attacking cell biomolecules, preventing injury formation and loss of cell integrity (Kunwar et al., 2012; Kumar et al., 2021; Rahgoshai et al., 2021) and promoting DNA repair (Kitabatake et al., 2020).

The evaluation of natural compounds as radioprotective agents is relevant due to their non-toxic essence, their easy availability (Yun and Wang, 2017; Kuruba and Gollapalli, 2018; Dowlath et al., 2021; Kumar et al., 2021), their antioxidant activity and the possibility that they may act effectively in reducing cancer incidence without adverse side effects (Fischer et al., 2018; Kuruba and Gollapalli, 2018; Dowlath et al., 2021). Plants possess a renewable source of metabolites with enormous chemical structural diversity, which may have potential therapeutic relevance (Bomfim et al., 2022).

In this sense, grapes (*Vitis labrusca* L.) and their derivatives stand out, which have numerous nutritional, pharmacological and therapeutic benefits (Apostolou et al., 2013; Kim et al., 2013; Huang et al., 2015; Devi and Singh, 2017; Lopes et al., 2019), mainly due to the presence of phenolic compounds and their antioxidant activity (Fernandes et al., 2013; Maestre et al., 2013; Pinto et al., 2022).

Therefore, the present study aimed to verify the cytotoxic activities of diagnostic and therapeutic doses of I-131 in human hepatocellular carcinoma (HepG2/C3A) and non-tumoral human lung fibroblast (MCR-5) cells. Also, to evaluate the radioprotective effects of whole grape juices produced from the conventional or organic production system, whether or not exposed to type C ultraviolet light irradiation (UV-C), which has been shown to increase the synthesis of flavonoids, anthocyanins and aromatic compounds (Cantos et al., 2001; Maurer et al., 2017; Pinto et al., 2022), without producing cytotoxic or mutagenic substances (Düsman et al., 2014).

2. Materials and Methods

2.1. Cell cultures

Considering that I-131 can be used for the treatment of hepatocellular carcinoma (Boucher et al., 2007; Trall and Ziessman, 2003; Costa and Sapienza, 2012) and that due to pulmonary circulation of I-131, pulmonary complications of patients have already been identified (Ahmadzadehfar et al., 2011; Costa and Sapienza, 2012), we used in the present study, as a test system, the human hepatocarcinoma cells, HepG2/C3A (Cat. 0291) and non-tumor human lung fibroblast, MRC-5 (Cat. 0180), obtained from the Rio de Janeiro Cell Bank. The cells were cultivated in 25cm^2 culture flasks containing complete culture medium (DMEM supplemented with 10% of fetal bovine serum and 1 mL/L antibiotic/antimycotic solution) and kept in a CO_2 incubator (5%, 95% humidity) at 37º C. Under these conditions, the cell cycle of both cells is approximately 24 hours. Cells in the logarithmic growth phase were used in these experiments.

2.2. Treatment solution

2.2.1. Grape juice

Vitis labrusca L. variety Concord grapes harvested in 2012 were cultivated in the municipality of Verê, belonging to the Francisco Beltrão microregion, in southwestern Paraná, Brazil. The grapes were obtained from two nearby properties (5.3km), one using conventional cultivation practice (altitude 564 m, latitude 25º 54 '01' 'S and longitude 52º 53' 51 " W) and one with organic cultivation practice (altitude 492 m, latitude 25º 51 '21' 'S and longitude 52º 55' 06 " W) under similar climatic and soil conditions.

The selection and treatment of grapes with UV-C irradiation was described by Cantos et al. (2001) and Düsman et al. (2014). The following parameters were

used: radiation fluence rate of 65.6 J/m² at a distance of 30cm from the light source. The grapes were arranged in a single layer in trays that were irradiated in a cabin equipped with three UV-C lamps (Philips® 90 W) for 5 minutes. The bunches of grapes were rotated 180° and the light source remained for another 5 minutes, totaling 10 minutes of irradiation time. The irradiated material was stored for three days at 25 ± 5º C in the absence of light to promote the biosynthesis of bioactive compounds.

Organic and conventional grapes, with or without UV-C postharvest treatment, were used to obtain whole juices. The whole juice was obtained in a steam puller at 90º C and bottled in glass. Sterile samples of the four conventional, UV-C-treated and conventional UV-C-treated organic grape juices were stored in a -4° C freezer and thawed at the time of experimentation.

2.2.2. I-131 radioisotope

The I-131 radioisotope was obtained from the Institute of Nuclear Energy Research, São Paulo, Brazil. On the day of the experiment, doses of I-131 were prepared using a CRC®-25R Dose Calibrator (Capintec Inc., USA) in order to determine the activity of the radiopharmaceutical. As I-131 was diluted in saline solution, the calculation was made of the amount of µL needed to obtain each concentration that was evaluated. The cells were directly exposed to radioactive iodine, which was diluted in the culture medium, and remained so throughout the experimental period.

2.2.3. Chemical agents

Methyl methane sulfonate (MMS - 99%, CAS 66-27-3), Dimethyl sulfoxide (DMSO - 99.7%, CAS 67-68-5) and 2,2-diphenyl-1picrylhydrazil (DPPH, CAS 1898 -66-4) were purchased from Sigma. Dulbecco's Modified Eagle Medium (DMEM), Fetal Bovine Serum, Antibiotic/Antimycotic and MTT [3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide] (98%, CAS 298 -93-1) were purchased from Gibco® Life Technologies (Carlsbad, CA, USA).

2.3. MTT cytotoxicity test

For the cytotoxicity test, the MTT assay was performed as described by Lopes et al. (2019). The 96-well cell culture plates were used, wherein each well 10^4 HepG2/C3A or MRC-5 cells were seeded. After stabilization, the cultures were treated as follows: control group (CO), cytotoxic agent methyl methane sulfonate (MMS 50µM) and I-131 concentrations, defined after pilot tests and spanning a wide range of equivalent diagnostic and therapeutic doses, with activities based clinical extrapolations (0.0037; 0.0185; 0.037; 0.185; 0.37; 0.74; 1.85; 3.70; 7.40 MBq/mL). The cytoprotection/radioprotection experiment was performed with the following treatments: control group (CO); organic, conventional, organic UV-C post-treated and conventional UV-C post-treated grape juices (10 and 20 µL/mL); I-131 (1.85 MBq/mL); and cytoprotective treatments with organic grape juice (10 and 20 µL/mL) + I-131 (1.85 MBq/mL), conventional grape juice (10 and 20 µL/mL) + I-131 (1.85 MBq/mL), organic UV-C post-treated grape juice (10 and 20 µL/mL) + I-131 (1.85 MBq/mL), and conventional UV-C post-treated grape juice (10 and 20 µL/mL)) + I-131 (1.85 MBq/mL).

After 24 or 48 hours of incubation, the culture medium was replaced with MTT (0.2 mg/mL) and the absorbance readings were performed in a 550nm microplate reader (Lopes et al., 2019).

Cell viability was estimated based on the mean absorbance (A) of the control by the following formula: $A_{treatment}/A_{control}$ x 100. The experiments were performed in three independent repetitions.

All experiments involving the use of radioisotope I-131 were developed at the Mutagenesis Laboratory of the Department of Biotechnology, Genetics and Cell Biology, adequately registered by the Brazilian National Nuclear Energy Commission (CNEN).

2.4. Antioxidant activity

Antioxidant activity was determined by the DPPH (2,2-diphenyl-1picrylhydrazil) method, as described by Brand-Wiliams et al. (1995) with modifications by Rufino et al. (2007). Four dilutions of each sample (grape juice) were performed. An aliquot of 0.1 mL of each sample dilution was transferred to test tubes with 3.9 mL of DPPH radical and 0.1 mL of the control solution of methyl alcohol, acetone and water. Then, homogenization was performed in a tube shaker. Spectrophotometer absorbance readings at 515 nm were taken after 30 minutes of reaction. The reaction time was established by previous tests, ranging from 30 minutes to 24 hours. Quantitation was based on the establishment of a standard curve at concentrations 10, 20, 30, 40, 50 and 60 µM from an initial DPPH solution (60 µM). Results were expressed as EC50 (mL juice/g DPPH), e.g., the values obtained correspond to the amount of sample needed to reduce the initial concentration of the DPPH radical by 50%. The experiments were performed in three independent repetitions.

2.5. Statistical analysis

The results were presented as mean and standard deviation, and submitted to one-way analysis of variance (ANOVA), followed by Dunnett's test (juice and radioisotope cytotoxicity experiments) and Tukey's (cytoprotection experiment and antioxidant activity) by the GrafPad® Prism 5 software. Differences were considered to be statistically significant when the p-value was less than 0.05.

3. Results and Discussion

Treatment with the radioisotope I-131 was cytotoxic to HepG2/C3A tumor cells at concentrations above 1.85 MBq/mL after 24 and 48 hours of exposure (Figure 1A). In addition, concentrations above 1.85 MBq/mL showed cell viability below 60% (24 hours) and 70% (48 hours) (Table 1). These data corroborate the commonly used in nuclear medicine, where low doses of I-131 presented a diagnostic effect, while high doses have a therapeutic effect, inducing tissue injury (Trall and Ziessman, 2003).

However, due to radiation migration to the pulmonary circulation, lung cells are affected by the use of

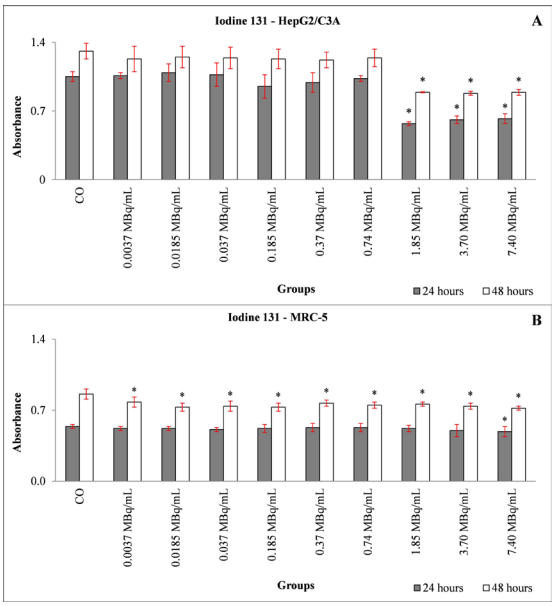

Figure 1. Mean absorbance and standard deviation of HepG2/C3A (A) and MRC-5 (B) cells treated with I-131. CO: Control; $1x10^4$ cells per well, incubated for 24 and 48 hours, n = 3. *Statistically significant result compared to Control ($p < 0.05$, Dunnett's test).

Table 1. Percentage of viability of HepG2/C3A and MRC-5 cells treated with Iodine-131 for 24 and 48 hours, by the MTT assay.

Groups	HepG2/C3A		MRC-5	
	CV %		CV %	
	24 h	48 h	24 h	48 h
CO	100.0	100.0	100.0	100.0
0.0037 MBq/mL	100.9	93.8	96.2	90.6
0.0185 MBq/mL	103.8	95.4	96.2	84.8
0.037 MBq/mL	101.9	94.6	94.4	86.0
0.185 MBq/mL	90.4	93.8	96.2	84.8
0.37 MBq/mL	94.2	93.1	98.1	89.5
0.74 MBq/mL	98.0	94.6	98.1	87.2
1.85 MBq/mL	54.3	67.9	96.3	88.3
3.70 MBq/mL	58.0	67.1	92.5	86.0
7.40 MBq/mL	59.0	67.9	90.7	83.7

CV = Cell viability. Groups: CO = Control; Treatment with Iodine-131; $1x10^4$ cells per well, incubated for 24 and 48 hours, n=3.

I-131 (Ahmadzadehfar et al., 2011; Costa and Sapienza, 2012). Data from the present study confirm that for MRC-5 non-tumor lung cells (Figure 1B), all I-131 concentrations (from 0.0037 to 7.40 MBq/mL) were cytotoxic within 48 hours presenting a significant decrease in the absorbances compared to the control. Within 24 hours, only the highest I-131 concentration (7.40 MBq/mL) was cytotoxic. Nevertheless, the minimum cell viability observed was 90.7% for 24 hours and 83.7% for 48 hours, showing a higher resistance of healthy cells to the harmful effects of radiation when compared to the tumor cells (Table 1), which is essential in terms of diagnostic or therapeutic applications with I-131.

The cytotoxic effect (cell death) induced by the I-131 on HepG2/C3A and MRC-5 cells were possibly due to the induction of oxidative stress generated mainly by beta particles resulting from the radioactive decay of I-131 (Hosseinimehr, 2010; Asadian et al., 2020; Nouailhetas et al., 2021). Reactive oxygen species can interact with cell macromolecules, inducing damage and even cell mortality, and this is one of the main mechanisms of the therapeutic and side effects of I-131 (Hosseinimehr et al., 2013; Nouailhetas et al., 2021).

The simultaneous administration of the radioisotope I-131 and the whole grape juices to HepG2/C3A cells for 24 hours (Figure 2) showed that these treatments, except for the concentration of 20 µL/mL of UV-C organic grape juice statistically increased the absorbance compared to the treatment with I-131, which was cytotoxic at 24 and 48 hours. Also, simultaneous administration of I-131 and juices presented mean absorbances similar to the treatment with only juice, except for treatment with conventional juice (20 µL/mL), indicating that even with the administration of I-131, cell divisions were not inhibited, nor cell viability decreased (Table 2).

Kim et al. (2013) have shown the protection of HepG2/C3A cellular oxidation pretreated with procyanidins, a grape juice derivative. The authors justify this protection by reducing reactive oxygen species levels and modulating the activity of glutathione, malondialdehyde and antioxidant enzymes, events that may have occurred in the present experiment when HepG2/C3A cells were treated with grape juices and the radioisotope.

However, within 48 hours (Figure 2), this cell division-inducing effect was reversed, as all treatments simultaneously performed with I-131 and juices, except 10 µL/mL of UV-C organic and conventional grape juices, showed a cytotoxic effect compared to control, reducing cell viability by more than 15% compared to the 24-hour treatment (Table 2). Also, all treatments performed with I-131 and juices presented mean absorbances similar to those obtained with the treatment performed with I-131 alone.

According to Costa and Sapienza (2012), the combined administration of chemotherapies can potentiate the effect of I-131-lipiodol in hepatocellular carcinoma treatments, increasing from 40% to 90% the cases with tumor reduction or stabilization. This effect was not observed in the present study considering the simultaneous administration of grape juices and I-131, which resulted in cytotoxic activity similar to the radioisotope alone within 48 hours.

Nevertheless, it is noteworthy that within 48 hours, UV-C conventional grape juice (20 µL/mL), organic grape juice (20 µL/mL) and UV-C organic grape juice (10 and 20 µL/mL) showed a cytotoxic effect on HepG2/C3A cells. These data corroborate the work of Lopes et al. (2019), in which the same juices evaluated in the present study, at concentrations of 10 to 100 µL/mL, presented antitumor activity for HepG2/C3A cells, especially for the highest concentrations, in the longest exposure time and with UV-C postharvest treatment. According to El-Din et al. (2019),

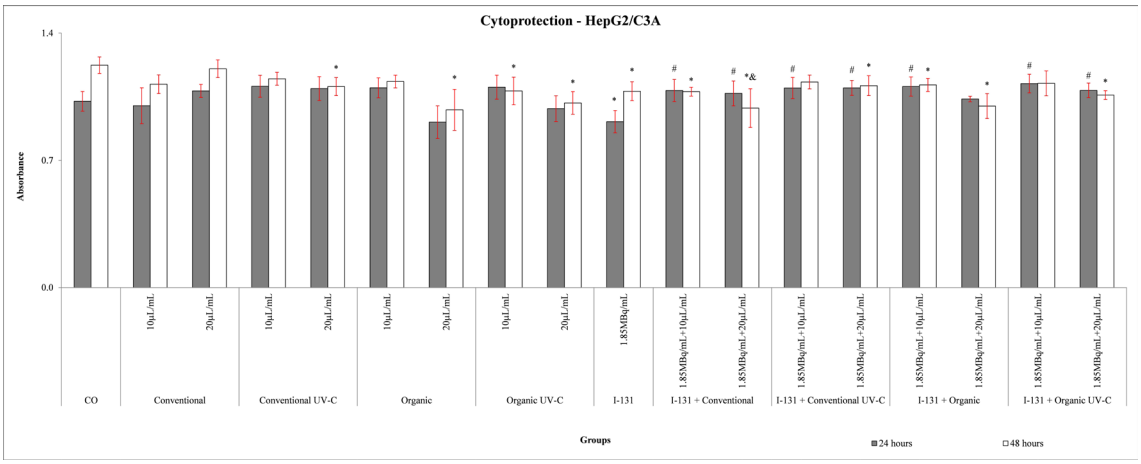

Figure 2. Mean absorbance and standard deviation of HepG2/C3A cells treated with conventional and organic grape juices, exposed or not to UV-C irradiation (10 and 20 µL/mL), treated alone or in cytoprotective tests with I-131 (1.85 MBq/mL). CO: Control; $1x10^4$ cells per well, incubated for 24 and 48 hours, n = 3, Tukey test. *Statistically significant result compared to Control; #Statistically significant result compared to treatment with I-131; &Statistically significant result compared to treatment with the same juice concentration, without I-131.

Table 2. Percentage of viability of HepG2/C3A and MRC-5 cells treated with conventional and organic grape juices, exposed or not to UV-C irradiation and Iodine-131, separately and simultaneously, for 24 and 48 hours, by the MTT assay.

Groups			HepG2/C3A		MRC-5	
			CV %		CV %	
			24 h	48 h	24 h	48 h
CO			100.0	100.0	100.0	100.0
Grape Juice	Conventional	10 µL/mL	97.6	91.4	101.1	97.2
		20 µL/mL	105.6	98.4	97.7	99.7
	UV-C Conventional	10 µL/mL	108.0	93.8	103.4	116.6
		20 µL/mL	106.8	90.3	101.5	108.6
	Organic	10 µL/mL	107.2	92.6	100.5	105.0
		20 µL/mL	88.8	80.0	97.3	104.4
	UV-C Organic	10 µL/mL	107.6	88.4	100.2	114.3
		20 µL/mL	96.0	83.0	97.3	109.9
I-131		1.85 MBq/mL	89.0	88.2	71.8	82.9
Cytoprotection: I-131 1.85 MBq/mL + Grape Juice	Conventional	10 µL/mL	105.8	88.0	96.4	94.4
		20 µL/mL	104.2	80.7	87.5	90.9
	UV-C Conventional	10 µL/mL	107.1	92.4	104.4	101.5
		20 µL/mL	107.2	90.7	97.2	94.2
	Organic	10 µL/mL	107.9	91.1	94.6	91.1
		20 µL/mL	101.2	81.6	93.6	99.4
	UV-C Organic	10 µL/mL	109.5	91.8	95.4	93.1
		20 µL/mL	105.8	86.5	90.6	91.7

CV = Cell viability. Groups: CO = Control; conventional and organic grape juices, exposed or not to UV-C irradiation (10 and 20 µL/mL), treated alone or in the cytoprotective tests together with Iodine-131 (1.85 MBq/mL); $1x10^4$ cells per well, incubated for 24 and 48 hours, n=3.

grape seed and peel extracts showed antitumor effect by cell cycle arrest, apoptosis induction and inhibition of cell proliferation. Thus, it is suggested the consumption of grape juices before or after the therapeutic use of I-131, allowing the stimulation of tumor cell elimination by the therapeutic action of these juices. According to Painuli and

Kumar (2016), free radical scavenging is the most common mechanism of antitumor activity, and this antioxidant effect also plays an important role in protecting healthy tissues from the lethal effect of radiation exposure.

Studies have shown that the use of antioxidants during radiotherapy does not interfere with the beneficial effects of treatment, and their careful use can improve and reverse the adverse effects of radiotherapy, acting as radioprotective agents and reducing levels of oxidizing agents (Moss, 2007). These effects were observed in the present study since all simultaneous treatments with I-131 and grape juices (organic or conventional, exposed or not to UV-C), were not cytotoxic to MRC- 5 cells (Figure 3). Besides, all treatments performed with I-131 and juices together within 24 hours and the treatment with 10 µL/mL of UV-C conventional grape juice and I-131, within 48 hours, presented statistically higher mean absorbances compared to the treatment with the radioisotope I-131 alone. Furthermore, considering that I-131 was cytotoxic to MRC-5 cells at 24 and 48 hours, with cell viability less than 72% (24 hours) and 83% (48 hours) (Table 2), the data of the present study have shown the radioprotective effect of grape juices.

This effect may have happened because of grape juices are rich in antioxidant substances (Table 3), as shown in other studies with these fruits, their extracts or compounds isolated from them (Trindade et al., 2016), may have reduced the cytotoxicity caused by the radioisotope to MRC-5 cells, preventing free radical formation by I-131 (Alam et al., 2002; Amri et al., 2014) and induced-damage to cells (Andrade et al., 2011). This may have favored the DNA repair and the reconstitution of damaged cell membranes (Picada et al., 2003), and even protecting from radiation-induced apoptosis (Kim et al., 2018).

A similar result was found by Hosseinimehr and Hosseini (2014), who showed that resveratrol, an antioxidant present in grapes, increased I-131-induced cell death in thyroid cancer cells and protected human non-malignant fibroblast cells (HFFF2) from the toxicity of this radioisotope, also by the MTT test.

Chaturvedi et al. (2021) showed that two fractions of *Cryptosporidium parvum* lysate showed radioprotective activity by normal cells irradiated with 10 Gy, improving cell viability and preventing radiation-induced DNA damage, which could be a candidate for the development of radioprotectant.

In the present study, the radioprotective activities were similar between the different types of grape juices tested (grapes from the organic or conventional production system, post-treated or not with UV-C), and the antioxidant contents of these juices did not differ from each other (Table 3). Data from Kalinova and Vrchotova (2011) and Margraf et al. (2016) corroborate those of the present study since they also did not observe an increase in bioactive compounds in organic foods when compared to conventional ones. Moreover, Cava and Sgroppo (2015) also did not identify the increased antioxidant activity of grape juices treated with UV-C.

So, these data stimulate in vivo studies about the consumption of grape juice as adjuvant in I-131 therapy for human hepatocarcinoma, considering the beneficial

Table 3. Mean and standard deviation of antioxidant activity (EC_{50}) of conventional and organic grape juices, with and without UV-C postharvest treatment.

Grape Juice	EC_{50} (mL juice/g DPPH)
Conventional	2,736.63 ± 510.42 a
UV-C Conventional	3,365.13 ± 241.81 a
Organic	2,802.94 ± 175.92 a
UV-C Organic	2,850.53 ± 207.55 a

Means followed by the same letter (a) do not differ statistically from each other at the 5% probability level by Tukey's test, n=3. EC_{50} = Amount of sample required to reduce initial DPPH concentration by 50%.

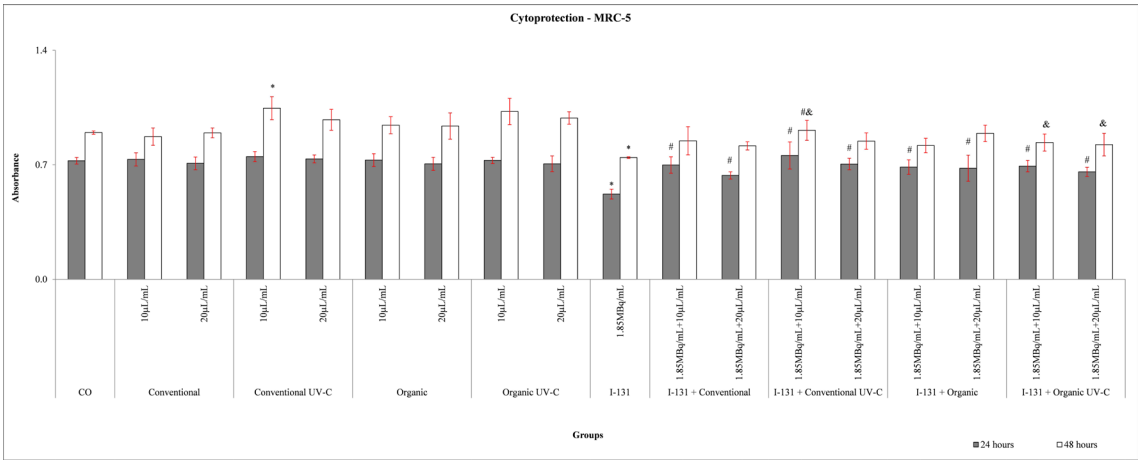

Figure 3. Mean absorbance and standard deviation of MRC-5 cells treated with conventional and organic grape juices, exposed or not to UV-C irradiation (10 and 20 µL/mL), treated alone or in cytoprotective tests with I-131 (1.85 MBq/mL). CO: Control; $1x10^4$ cells per well, incubated for 24 and 48 hours, n = 3, Tukey test. *Statistically significant result compared to Control; #Statistically significant result compared to treatment with I-131; &Statistically significant result compared to treatment with the same juice concentration, without I-131.

potential in vitro of this juice as antitumor, nutraceutical potential, antioxidant activity and radioprotective of stimulated metabolic cells against ionizing damage from radiation.

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