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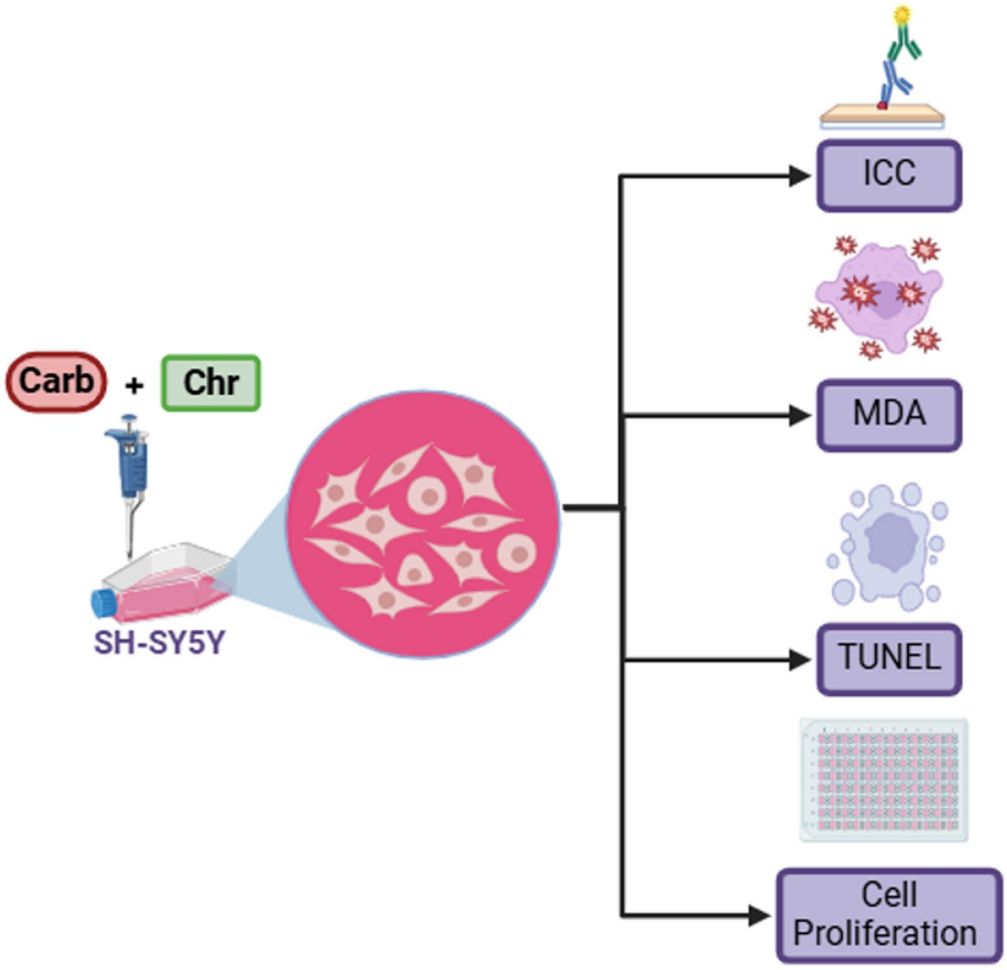

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AIM AND SCOPES
Journal of Cellular Neuroscience and Oxidative Stress is an online journal that publishes original research articles, reviews and short reviews on the molecular basis of biophysical, physiological and pharmacological processes that regulate cellular function, and the control or alteration of these processes by the action of receptors, neurotransmitters, second messengers, cation, anions, drugs or disease.

Areas of particular interest are four topics. They are;

A-Ion Channels (Na^+- K^+ Channels, Cl^- channels, Ca^{2+} channels, ADP-Ribose and metabolism of NAD^+, Patch-Clamp applications)

B-Oxidative Stress (Antioxidant vitamins, antioxidant enzymes, metabolism of nitric oxide, oxidative stress, biophysics, biochemistry and physiology of free oxygen radicals)

C-Interaction Between Oxidative Stress and Ion Channels in Neuroscience
(Effects of the oxidative stress on the activation of the voltage sensitive cation channels, effect of ADP-Ribose and NAD^+ on activation of the cation channels which are sensitive to voltage, effect of the oxidative stress on activation of the TRP channels in neurodegenerative diseases such Parkinson's and Alzheimer's diseases)

D-Gene and Oxidative Stress
(Gene abnormalities. Interaction between gene and free radicals. Gene anomalies and iron. Role of radiation and cancer on gene polymorphism)

READERSHIP

Biophysics	Biochemistry
Biology	Biomedical Engineering
Pharmacology	PhysiologyGenetics
Cardiology	Neurology
Oncology	Psychiatry
Neuroscience	Neuropharmacology

Keywords
Ion channels, cell biochemistry, biophysics, calcium signaling, cellular function, cellular physiology, metabolism, apoptosis, lipid peroxidation, nitric oxide, ageing, antioxidants, neuropathy, traumatic brain injury, pain, spinal cord injury, Alzheimer's Disease, Parkinson's Disease.

RESEARCH ARTICLE

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Chrysin protects neuronal cells against carboplatin exposure-induced apoptosis and oxidative damage

Adnan AYNA[1*], Sevda SAĞ[2], İbrahim BAYAV[2], Ekrem DARENDELIOGLU[3]

[1]*Department of Chemistry, Faculty of Arts and Sciences, Bingol University, Bingol/Türkiye*
[2]*Department of Medical Biology, Faculty of Medicine, Pamukkale Universty, Denizli/ Türkiye*
[3]*Department of Molecular Biology and Genetics, Faculty of Arts and Sciences, Bingol/ Türkiye*

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***Address for correspondence:**
Adnan AYNA,
Address: Department of Chemistry, Faculty of Arts and Sciences, 12000-Bingol University, Bingol, Turkey.
Email: *aayna@bingol.edu.tr*
Phone: *00904262160012 – 2546*

List of Abbreviations;

Carb, *Carboplatin;* ***Chr,*** *Chrysin;* ***LPO,*** *Lipit peroxidations;* ***TUNEL,*** *Terminal deoxynucleotidyl transferase dUTP nick end labelling;* ***Cyt c,*** *Cytocrome C;* ***ROS,*** *Reactive oxygen species;* ***DMEM,*** *Dulbecco's modified eagle medium;* ***FBS,*** *Fetal Bovine Serum;* ***WST-1,*** *Water Soluble Tetrazolium-1;* ***MDA,*** *Malondialdehyde;* ***TBA,*** *Thiobarbituric acid;* ***SDS-PAGE,*** *Sodium dodecyl sulfate-polyacrylamide gel;* ***TBS-T,*** *TBS with 0.05% Tween-20*

Abstract

Chemotherapy drugs such as carboplatin (Carb) are widely used to treat various cancers, including testicular, lung, and ovarian cancer. Although Carb primarily targets cancer cells, it can also damage healthy cells, including neuron, leading to potential adverse effects. Notably, some side effects of Carb therapy are associated with nerve cells and the nervous system. This study aimed to investigate the potential protective effects of chrysin (Chr) against Carb-induced oxidative toxicity in SH-SY5Y neuronal cells. The ameliorative effects of Chr on Carb-induced cellular toxicity were evaluated through cell viability assay, lipid peroxidation (LPO) analysis, TUNEL assay and immunocytochemistry staining. The results indicated that Chr mitigates Carb toxicity in SH-SY5Y cells through alleviating cell viability and reducing LPO levels. The study also demonstrated that Carb treatment caused apoptosis by causing DNA strand breaks while Chr treatment alleviated these effects. These findings suggest that Chr, may diminish the apoptotic effects of Carb in SH-SY5Y cells and could provide insights into potential therapeutic strategies for mitigating cell damage caused by Carb.

Keywords: Apoptosis, carboplatin, chrysin, SH-SY5Y cells, neurotoxicity

Introduction

Treating cancer disease still mainly relies on the ability of chemotherapeutics to eradicate cancer cells, diminish tumour growth and relieve pain. Chemotherapeutic drugs are widely employed to combat different forms of cancer by inhibiting the proliferation of cancer cells (Glazer, 2019). Chemotherapeutic agents are known to have multiple mechanisms of action and may fall into more than one category. A key category of chemotherapeutic drugs is alkylating agents, which directly damage DNA. This group includes platinum-based drugs such as cisplatin, oxaliplatin, and carboplatin (Carb) (Yousef et al., 2018). A common chemotherapy treatment for cervical cancer, ovarian cancer, malignant lymphoma and non-small-cell lung cancer is Carb, a platinum-based

anticancer medication (Miyano et al., 2019). The occurrence and intensity of side effects at therapeutic doses are one of the factors that differentiate anticancer treatments from other medications. Chronic or acute, self-limited, long-lasting, mild, or possibly fatal adverse effects are all possible. The most frequent adverse effects include anemia, alopecia, mucositis, bleeding (thrombocytopenia), nausea and vomiting, hyperuricemia, bone marrow depression, and hair loss. Therefore, in order to avoid, minimize, and overcome these adverse effects, a variety of factors need to be taken into account. It's critical to concentrate on this area of research to identify the appropriate strategies for mitigating these negative effects. In recent decades, a variety of therapy approaches have been used to reduce adverse effects (Luo etal., 2018; Kikuchi et al., 2019; Ayna et al, 2020; Cavalier et al., 2020; Çelik et al., 2020)

Antioxidants, especially, can be effective in mitigating the toxic effects of Carb and in reducing the harm that cytotoxic drugs inflict on non-tumor tissue (Bailly, 2019; Ma et al., 2020). The molecular mechanisms responsible for the antitumor actions rely on binding to nucleus DNA, preventing DNA synthesis and resulting in necrosis or apoptosis (Ho et al., 2016). Additionally, Carb may cause oxidative stress, which would result in reactive oxygen species (ROS) and encourage apoptosis (Cheng et al., 2008). Direct or indirect ROS activity appears to play a crucial role in facilitating the release of Cyt c in cytoplasm from mitochondria, which subsequently leads to the activation of caspases (Simon et al., 2000). Endovascular and neurological disorders are closely associated with oxidative stress brought on by ROS build up (Li et al., 2011). Through apoptotic and autophagic pathways, ROS destroys neurons and promotes cell death in other endothelial cells (Focaccetti et al., 2015). LPO induced by ROS is critical for cell death, specifically apoptosis. This essential mechanism involves an excess of ROS that propagates LPO chain reactions, infiltrates biological membranes, and ultimately initiates cellular death (Su et al., 2019).

Chrysin (Chr; 5,7-dihydroxyflavone) is a phytochemical widely found in various fruits and vegetables, including honey, bee propolis, mushrooms, blue passion flowers (such as *Passiflora incarnata*, *Oroxylum indicum*, and *Passiflora caerulea*) and *Scutellaria baicalensis* (Mani et al., 2018). With potent antioxidant, anticancer, and anti-inflammatory properties, Chr is a promising naturally occurring bioactive flavonoid. Chr has the potential to be used in clinical and therapeutic settings to counteract the physiological and metabolic impacts of ageing. It possesses cytotoxic and anti-inflammatory properties and is frequently used to treat a number of degenerative disorders (Özbolat and Ayna 2020).

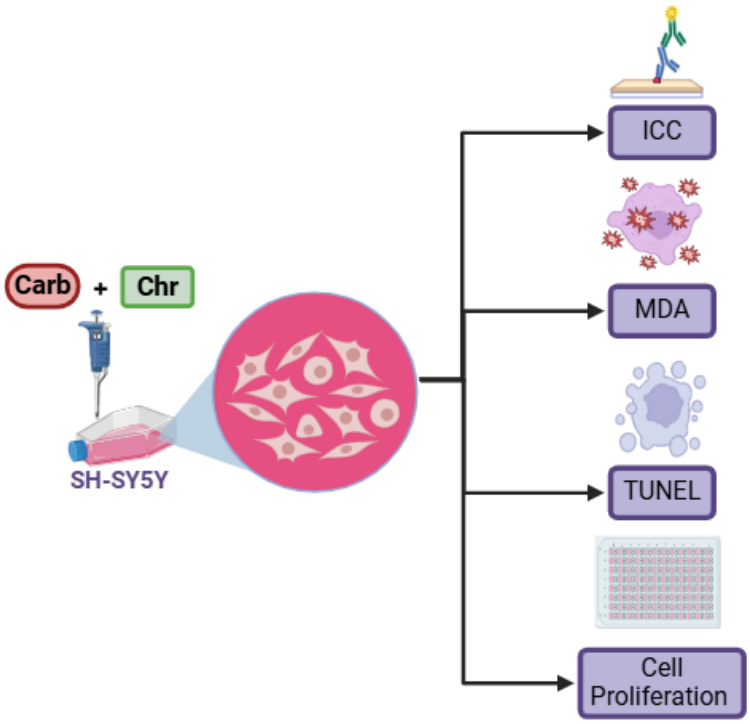

Figure 1. *Neuroprotective effects of chrysin against carboplatin-induced neurotoxicity. ICC: immunocytochemistry; Carb: Carboplatin; Chr: Chrysin; MDA: Malondialdehyde; TUNEL; Terminal deoxynucleotidyl transferase dUTP nick end labelling.*

While the molecular effects of platinum-based chemotherapeutic medicines have been extensively explored in cancer cells, there has been less research and clarity regarding the spectrum of these agents' potential effects in non-dividing cells. Our study's objective was to assess Chr's potential neuroprotective effects using an *in vitro* model of SH-SY5Y cells through analysis of cell viability, lipid peroxidation and apoptotic marker

caspase-3 (**Figure 1**).

Materials and Methods

Materials

The human neuroblastoma cell line (SH-SY5Y) obtained from ATCC (American Type Culture Collection) is available at Bingöl University and these cell lines were used in the studies. Chrysin (97% purity) was purchased from Sigma-Aldrich (St Louis, MO, USA) Chrysin was dissolved in DMSO (Sigma-Aldrich, Cat no: 67-68-5). Carboplatin was purchased from Koçak Farma (Üsküdar, İstanbul, Türkiye). Malondialdehyde bis (PubChem CID:67147) was bought from Merck (Germany). Dulbecco's Modified Eagle Medium (DMEM), Fetal Bovine Serum (FBS) and penicillin/streptomycin antibiotics were supplied from GIBCO (Gibco, USA).

Cell culture and treatment

Cells were grown in cell culture medium containing 89% DMEM, 1% penicillin/streptomycin antibiotics and 10% FBS in 75 cm^2 flasks at 37 ^oC in a CO_2 incubator. The chemotherapeutic drug Carb and the experimental agent Chr were prepared in cell medium and applied at different doses. The therapeutic dose of Chr at different concentrations was determined against the effective dose of Carb.

The cells were exposed to varying concentrations of Carb (7.8 μM to 1 mM) for a duration of 24 hours. They were divided into three distinct groups;

i) Control group: Only medium was given to the cells.

ii) Carb group: The cells treated with 1 mM Carb for 24 hours.

iii) Chr group: The cells treated with 500 μM Chr for 24 hours. (Ayna and Varan, 2023)

iv) Chr+Carb group: The cells were pre-treated with 500 μM Chr for 4 hours, followed by treatment with 1 mM Carb in the presence of the same Chr concentration for an additional 24 hours.

Cell proliferation assay

Once the SH-SY5Y cells cultured in a flask reached adequate growth, they were harvested using Trypsin-EDTA, stained with trypan blue on a Thoma slide, and counted under an inverted microscope. The counted cells were then seeded into 96-well plates, with 10,000 cells per well. Various concentrations of Carb (7.8 μM to 1 mM) were applied to the cells to identify the most effective cytotoxic concentration, as detailed in section cell culture and treatment. After 24 hours of incubation under the specified experimental conditions, cell viability was evaluated using the Water Soluble Tetrazolium-1 (WST-1) cell proliferation assay kit. This colorimetric assay is based on the enzymatic cleavage of the tetrazolium salt (WST-1) by mitochondrial dehydrogenases in metabolically active cells, producing a water-soluble formazan dye. The intensity of the formazan dye, which directly correlates with the number of viable cells, was measured spectrophotometrically at 450 nm using a microplate reader. The procedure followed the manufacturer's protocol and was consistent with the methodology established in our previous studies (Aykutoglu et al., 2020).

Analysis of intracellular malondialdehyde (MDA) level

LPO analysis was performed using this method based on the measurement of thiobarbituric acid (TBA) reactive species composed of MDA. Cells were treated as indicated in the experimental groups. Then, these cells were taken and 0.25 mL trichloroacetic acid (70% w/v) was reacted with 1 mL TBA (0.8% w/v) and kept in a 95 ^oC boiling water bath for 30 minutes. Afterwards, it was placed on ice and waited for 5 minutes. After centrifugation at 10,000 rpm, MDA levels of each sample were calculated by taking 532 nm absorbance measurements using an ELISA reader, and the results were expressed in nmol/mg protein.

TUNEL experiments

In situ apoptosis detection was conducted following the protocol provided with the ApopTag® Plus Peroxidase In Situ Apoptosis Detection Kit. Cells were plated in a 6-well plate and treated according to the experimental conditions. These cells were then fixed to the surface with 1% paraformaldehyde. After the cells were treated according to the kit protocol, at least five photographic images were taken for each sample under a light microscope. The apoptotic index was determined by dividing the number of apoptotic cells in each image by the total number of cells.

Immunocytochemical staining

UltraVision™ LP Detection System, HRP Polymer/DAB Plus Chromogen kit was used to histologically identify cells that specifically express the target proteins. Cells were stained according to the kit

protocol and cells positive for the targeted specific protein were visualized.

Statistics

The data obtained from our experimental studies, each conducted in at least three replicates, were statistically analyzed using one-way ANOVA followed by Tukey's Multiple Comparison test in GraphPad Prism 5.01 software, with a significance level considered at.

Results

Cell proliferation assay

The effect of Chr treatment on Carb -induced cell death in SH-SY5Y cells was assessed using the WST-1 assay. To induce cytotoxicity, SH-SY5Y cells were exposed to various concentrations of Carb (7.8 µM, 15.62 µM, 31.25 µM, 62.5 µM, 125 µM, 250 µM, 500 µM, 1 mM) for 24 hours. Carb treatment for 24 hours reduced cell proliferation in a concentration-dependent manner. Cell proliferation was markedly attenuated ($p < 0.01$) under 1 mM Carb treatment, relative to the untreated control group (**Figure 2a**). Consequently, a concentration of 1 mM Carb was selected for inducing SH-SY5Y cell death in subsequent experiments. To evaluate the protective effects of Chr, cells were pretreated with 500 µM Chr for 4 h, followed by treatment to 1 mM Carb in the presence of the same antioxidant concentrations for another 24 h, and the concentration of Chr used was shown to have a protective effect against Carb toxicity ($p < 0.01$) (**Figure 2b**).

Analysis of intracellular MDA level

LPO experiment was performed to assess the extent of cell damage following the application of 1 mM Carb to SH-SY5Y and to evaluate the effectiveness of 500 µM Chr treatment in mitigating this damage, as well as to determine the MDA levels. As shown in **Figure 3**, the MDA level in Carb-treated SH-SY5Y cells significantly increased ($p < 0.01$), whereas Chr pre-treatment markedly reduced MDA levels ($p < 0.05$) compared to Carb-treated SH-SY5Y cells.

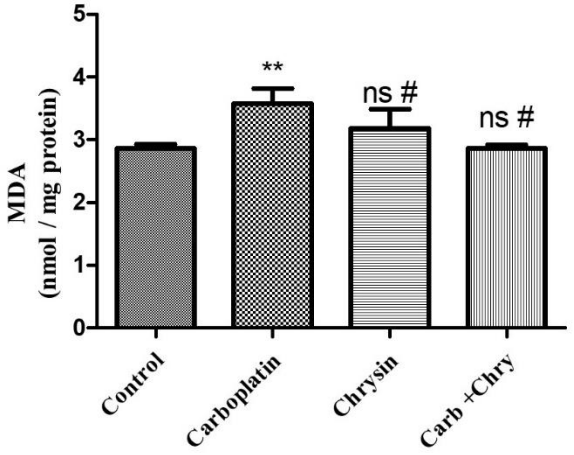

Figure 3: *Results of LPO obtained in the SH-SY5Y cell line at 24 hours (**p<0.01 control vs others, #p<0.05 carboplatin vs others ns; not significant, n=3)*

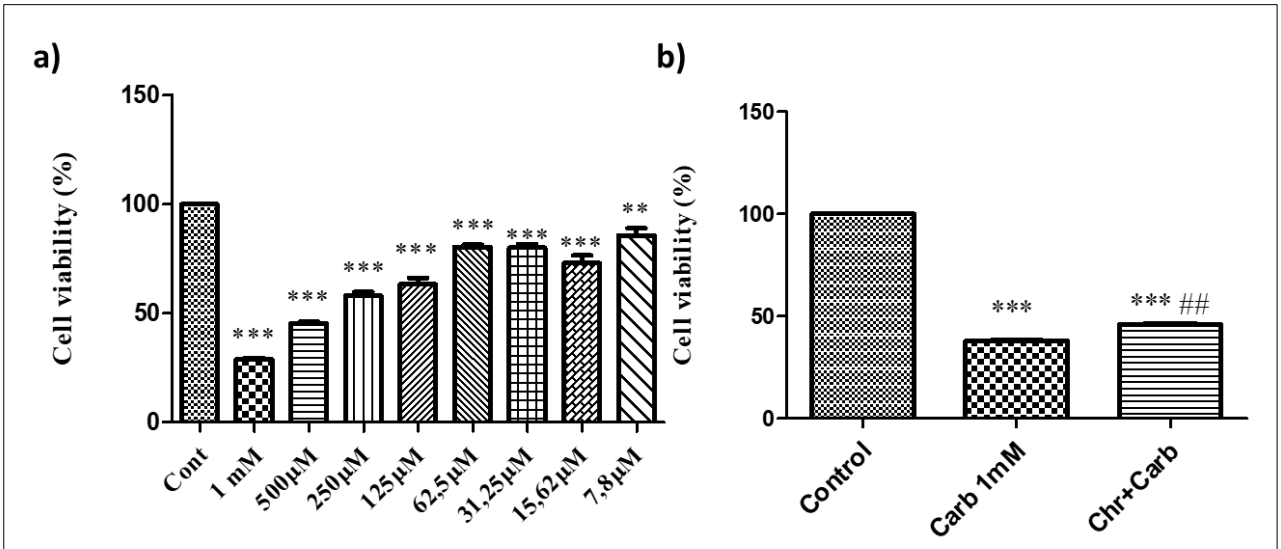

Figure 2. *Wst-1 results in SH-SY5Y cell line. a) Effect of Carb on cell viability. b) Determination of cell viability using Carb+Chr combine. Data were expressed as mean ± SEM, n = 3. (**p < 0.01 Control vs Others, ***p < 0.001 Control vs Others, ## p< 0.01 Carb vs Chr+Carb, ns: not significant, n=3)*

TUNEL assay

It was also assessed how well Chr pre-treatment prevented the apoptosis that Carb caused in SH-SY5Y cells. According to the TUNEL assay, the results showed that Carb caused apoptosis by causing DNA strand breaks, a significant increase in the Carb group ($p<0.001$). Chr pre-treatment greatly reduced the apoptosis caused by Carb ($p<0.001$) (**Figure 4**).

cell death was caused by apoptosis. Incubation with Carb-induced SH-SY5Y cells strongly stimulated caspase-3 expression, as **Figure 5** illustrates. Chr significantly reduced the activation of caspase-3.

Discussion

Platinum drugs, including oxaliplatin, carboplatin, and cisplatin, are essential components of combination

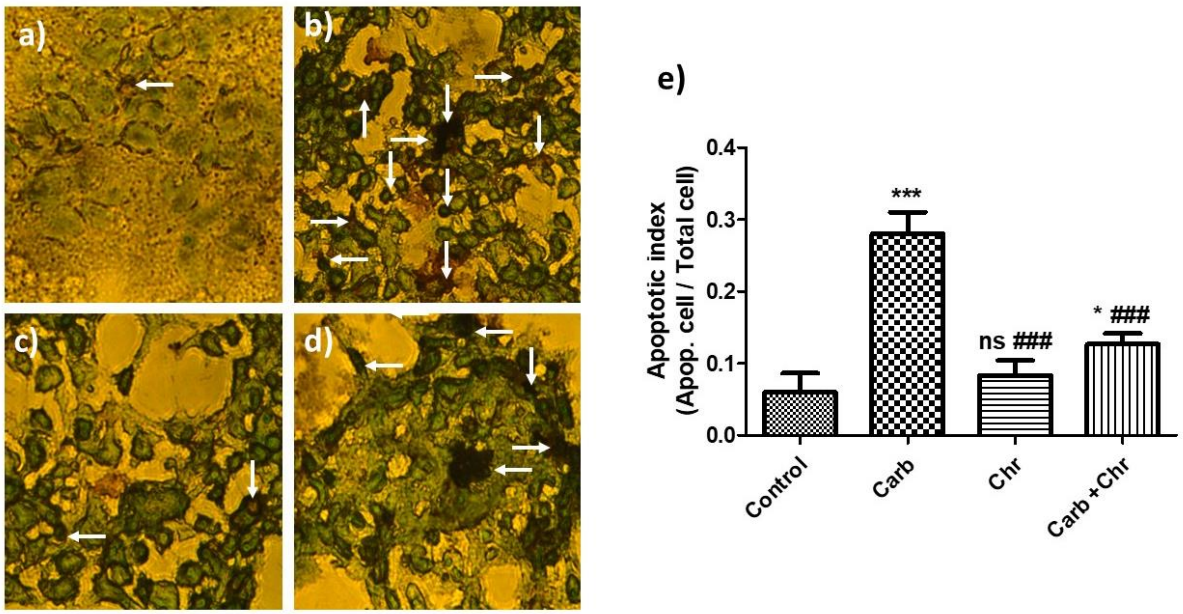

Figure 4: *The 24-hour apoptotic effects of Carb and Chr on the SH-SY5Y cell line were evaluated using the TUNEL assay method. Representative photographs of a) Control, b) Carb, c) Chr, d) Carb+Chr. Normal cells (green nuclei) and apoptotic cells (black and brown nuclei) were examined under an inverted light microscope e) Quantification analysis of TUNEL-positive cells. Data are shown as mean ± SD. (*p < 0.05 and ***p < 0.001 Control vs. Others, ###p < 0.001 Carb vs. Others, ns: not significant, n=3).*

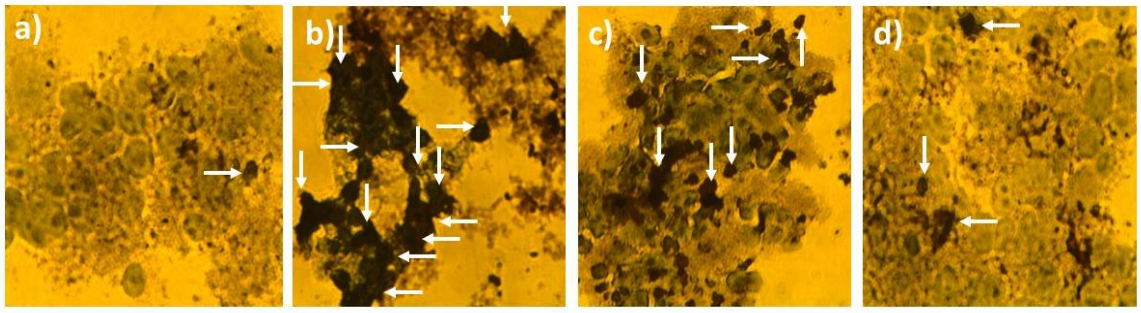

Figure 5: *The immunocytochemical staining of caspase 3 results were examined under an inverted light microscope. The caspase 3 positive cells (black) are indicated by arrows. a) Control, b) Carb, c) Chr, d) Carb+Chr.*

Immunocytochemical staining

Studies were conducted on the activation of caspase-3, a crucial mediator of apoptosis, to verify if Carb-induced

chemotherapy for various solid tumors. While these agents have potent anti-tumor properties, they are also associated with side effects such as neurotoxicity (Avan et al., 2015;

Carozzi et al., 2015). Although Carb is generally less neurotoxic than cisplatin and oxaliplatin, high doses can still induce neurotoxicity (Amptoulach and Tsavaris, 2011). *In vitro* studies have shown that exposure to cisplatin, oxaliplatin, or Carb increases cell death and apoptosis in rat sensory neurons in a concentration-dependent manner (Kanat et al., 2017). A Cochrane review analyzed data from 29 randomized controlled trials involving 2,906 participants receiving platinum-based chemotherapy with or without potential chemo-protectants such as oxcarbazepine, calcium/magnesium infusion, and glutathione (Albers et al., 2014). The review found insufficient evidence to recommend any specific agent for preventing or reducing platinum-induced neurotoxicity. While the molecular effects of platinum drugs in cancer cells are well-studied, their impact on non-dividing cells remains less understood. Our study's objective was to assess Chr's potential neuroprotective qualities using SH-SY5Y cells. We were able to assess Chr's potential to lessen the generation of ROS and neurotoxicity caused by Carb using this model system. The present investigation assessed the protective effects of Chr against Carb-induced oxidative toxicity in human SH-SY5Y cells by a battery of assays, including cell proliferation, LPO, western blotting, immunocytochemical labeling, and Tunel assay.

The most significant phytochemicals found in food are dietary flavonoids, which have numerous positive effects on human health (Caglayan et al. 2018; Taslimi et al. 2019). Several independent investigations have concentrated on the biological and pharmacological characteristics of naturally occurring flavonoids, such as their anti-inflammatory, antibacterial, antidiabetic, antioxidant, antiallergic, and antiapoptotic effects, as summarized in Gulçin, 2020 (Gulcin, 2020). Natural flavonoid Chr was employed in herbal medicine and was frequently present in propolis and honey. An increasing amount of research has demonstrated that Chr protects animal tissues, such as the brain, liver, lung, kidney, and heart, against harmful substances (Shooshtari et al., 2020; Kucukler et al., 2020; Temel et al., 2020; Temel et al., 2021). By reducing oxidative stress, Chr efficiently suppresses cisplatin-induced renal, intestinal, and ototoxicities (Kelleş et al., 2014). The protective role of Chr in Carb induced cytotoxities remained unclear. In our study, Chr effectively suppress Carb-induced cytotoxicity in SH-SY5Y cells.

MDA is a toxic and reactive byproduct of LPO. It is commonly used as a biomarker to assess LPO levels in various studies (Bayav et al., 2024). MDA can alter membrane permeability and disrupt membrane fluidity (Emra et al., 2023). Carb has been shown to significantly increase MDA levels in rat cochlea (Husain et al., 2001). In a study involving rats, thalidomide and Carb were found to induce brain damage through elevated LPO levels (Yousef et al.,2018). Additionally, another study demonstrated that aluminium chloride-induced LPO in mouse brains significantly decreased MDA levels in the cerebral cortex and hippocampus of Chr-treated mice (Campos et al., 2022). In our study, MDA levels were measured as an indicator of LPO. The data obtained were consistent with the literatüre. While MDA levels significantly increased in the Carb group compared to the control group, this increase was mitigated in the Chr pretreated group.

For most multicellular organisms, the maintenance and development of homeostasis depend heavily on apoptosis. Recent research has confirmed that mitochondria play a critical role in mediating the apoptotic process, as stated (Abotaleb, 2019). It is commonly recognized that oxidative stress can trigger apoptosis. The activation of p53 in response to oxidative stress results in mitochondrial damage and upsets the balance between anti-apoptogenic Bcl-2 and Bcl-xl and pro-apoptogenic Bax proteins (Baiyun et al., 2018). Some significant proteins, such as Cyt c or other molecules that induce apoptosis, are released into the cytoplasm as a result of this process. Caspases 3 and 4 are activated by the release of Cyt c into the cytoplasm, resulting in cleaved caspase-3 (Adelusi et al., 2024). Apoptosis results from a sequence of enzymatic events set off by caspase-3 activation (Baiyun et al., 2018; Adelusi et al., 2024). In apoptotic cells, many morphological changes occur, such as chromatin condensation, apoptotic body formation, cell membrane shrinkage, and DNA fragmentation. In our study, the TUNEL method was used to evaluate the DNA fragmentation characteristics of apoptosis, while immunocytochemical staining was performed to detect the expression of cleaved caspase-3 (Zhivotosky and Orrenius, 2001). As a result of these analyses, the Carb group showed an increase in both DNA fragmentation and cleaved caspase-3 expression compared to the control group. However, it was determined that these increases were significantly reduced in the Carb+Chr group compared to the Carb group. Platinum-based chemotherapeutics have

increased DNA fragmentation, and the obtained results are in accordance with the literature (Donzelli et al., 2004, Balaraman, 2005).

Conclusion

The purpose of this work was to evaluate Chr's potential to mitigate the cytotoxicity of Carb-induced SH-SY5Y. In this investigation, the application of Carb to the SH-SY5Y cells resulted in a reduction in cell division, elevation of LPO levels, and elevation of the apoptotic index. These results revealed the protective effects of Chr against Carb-induced apoptosis and oxidative stress in SH-SY5Y cells.

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Conflict of interest

The authors declare that there are no conflicts of interest.

Author contributions

AA: Methodology, statistical analysis, writing – review and editing, supervision. SS: Investigation, methodology, conceptualization. IB: investigation, methodology, validation, writing – review and editing, statistical analysis. ED: writing – review and editing, supervision.

Availability of data and materials

The data produced in this study is available from the corresponding author upon reasonable request.

Compliance with ethical standards

The authors are committed to compliance with ethical standards.

ORCID

A AYNA	0000-0001-6801-6242
S SAĞ	0000-0001-8258-7324
İ BAYAV	0000-0002-3533-3935
E DARENDELİOĞLU	0000-0002-0630-4086

References

Abotaleb M. (2019). Flavonoids in cancer and apoptosis. Cancers. 11:28. doi: 10.3390/cancers11010028.

Adelusi OB, Etemadi Y, Akakpo JY, Ramachandran A, Jaeschke H. (2024). Effect of ferroptosis inhibitors in a murine model of acetaminophen-induced liver injury. J Biochem Mol Toxicol. 38(8), e23791. doi:10.1002/jbt.23791.

Albers JW, Chaudhry V, Cavaletti G, Donehower RC. (2014) Interventions for preventing neuropathy caused by cisplatin and related compounds. Cochrane Database Syst Rev. (3). doi: 10.1002/14651858.CD005228.pub4.

Amptoulach S, Tsavaris N. (2011). Neurotoxicity caused by the treatment with platinum analogues. Chemother Res Pract. 2011. doi:10.1155/2011/843019.

Avan A, Postma TJ, Ceresa C, Avan A, Cavaletti G, Giovannetti E, Peters GJ. (2015). Platinum-induced neurotoxicity and preventive strategies: past, present, and future. Oncologist. 20(4), 411. doi:10.1634/theoncologist.2014-0044.

Aykutoglu G, Tartik M, Darendelioglu E, Ayna A, Baydas G. (2020). Melatonin and vitamin E alleviate homocysteine-induced oxidative injury and apoptosis in endothelial cells. Mol Biol Rep. 47, 5285–5293. doi:10.1007/s11033-020-05607-z.

Ayna A, Özbolat SN, Darendelioglu E. (2020). Quercetin, chrysin, caffeic acid and ferulic acid ameliorate cyclophosphamide-induced toxicities in SH-SY5Y cells. Mol Biol Rep. 47(11), 8535-8543. doi:10.1007/s11033-020-05896-4.

Ayna A, Varan SN. (2023) Investigation of the Protective Effects of Chrysin Against Paclitaxel-Induced Oxidative Stress and Apoptosis in Human Neuronal SH-SY5Y Cells. Turkish Journal of Nature and Science. 12(4), 107-113. doi: 10.46810/tdfd.1375041.

Bailly C. (2019). Potential use of edaravone to reduce specific side effects of chemo-, radio-and immuno-therapy of cancers. Int Immunopharmacol. 77, 105967. doi:10.1016/j.intimp.2019.105967.

Baiyun R, Li S, Liu B, Lu J, Lv Y, Xu J, Zhang Z. (2018). Luteolin-mediated PI3K/AKT/Nrf2 signaling pathway ameliorates inorganic mercury-induced cardiac injury. Ecotoxicol Environ Saf. 161, 655-661. doi:10.1016/j.ecoenv.2018.06.046.

Balaraman, P. (2005). An investigation of the mechanism of cisplatinum-induced apoptosis in SH-SY5Y neuroblastoma cells. University of London, University College London (United Kingdom).

Bayav I, Darendelioğlu E, Caglayan C. (2024). 18β-Glycyrrhetinic acid exerts cardioprotective effects against BPA-induced cardiotoxicity through antiapoptotic and antioxidant mechanisms. J Biochem Mol Toxicol. 38(2), e23655. doi:10.1002/jbt.23655.

Caglayan C, Temel Y, Kandemir FM, Yildirim S, Kucukler S. (2018). Naringin protects against cyclophosphamide-induced hepatotoxicity and nephrotoxicity through modulation of oxidative stress, inflammation, apoptosis, autophagy, and DNA damage. Environ Sci Pollut Res. 25, 20968–20984. doi:10.1007/s11356-018-2242-5.

Campos H M, da Costa M, da Silva Moreira LK, da Silva Neri HF, da Silva C R.B, Pruccoli L, Ghedini PC. (2022). Protective effects of chrysin against the neurotoxicity induced by aluminium: In vitro and in vivo studies. Toxicology. 465, 153033. doi:10.1016/j.tox.2021.153033.

Carozzi VA, Canta A, Chiorazzi A. (2015). Chemotherapy-induced peripheral neuropathy: what do we know about mechanisms?. Neurosci Lett. 596, 90-107. doi:10.1016/j.neulet.2014.10.014.

Cavalier AN, Clayton Z, Wahl D, Seals D, LaRocca T. (2020). The Effects of Chemotherapeutic Agents and a Mitochondrial Antioxidant on the Brain Transcriptome and Cognitive Function. FASEB J. 34(S1), 1-1. doi:10.1096/fasebj.2020.34.s1.09524.

Çelik H, Kandemir F M, Caglayan C, Özdemir S, Çomaklı S, Kucukler S, Yardım A. (2020). Neuroprotective effect of rutin against colistin-induced oxidative stress, inflammation and apoptosis in rat brain associated with the CREB/BDNF expressions. Mol Biol Rep. 47(3), 2023-2034. doi:10.1007/s11033-020-05302-z.

Cheng CF, Juan SH, Chen JJ, Chao Y C, Chen HH, Lian WS, Lin H. (2008). Pravastatin attenuates carboplatin-induced cardiotoxicity via inhibition of oxidative stress associated apoptosis. Apoptosis. 13(7), 883. doi:10.1007/s10495-008-0214-9.

Donzelli, E., Carfì, M., Miloso, M., Strada, A., Galbiati, S., Bayssas, M., ... & Tredici, G. (2004). Neurotoxicity of platinum compounds: comparison of the effects of cisplatin and oxaliplatin on the human neuroblastoma cell line SH-SY5Y. J Neurooncol, 67, 65-73.

Emre Kızıl H, Gür C, Ayna A, Darendelioğlu E, Küçükler S, Sağ S. (2023). Contribution of oxidative stress, apoptosis, endoplasmic reticulum stress and autophagy pathways to the ameliorative effects of hesperidin in NaF-induced testicular toxicity. Chem Biodivers. 20(3), e202200982. doi:10.1002/cbdv.202200982.

Focaccetti C, Bruno A, Magnani E, Bartolini D, Principi E, Dallaglio K, Albini A. (2015). Effects of 5-fluorouracil on morphology, cell cycle, proliferation, apoptosis, autophagy and ROS production in endothelial cells and cardiomyocytes. PloS one. 10(2), e0115686. doi:10.1371/journal.pone.0115686.

Glazer RI. (2019). Developments In Cancer Chemotherapy: Vol. 2. CRC Press.

Gulcin İ. (2020). Antioxidants and antioxidant methods: an updated overview. Arch Toxicol. 1-65. doi:10.1007/s00204-020-02689-3.

Ho G Y, Woodward N, Coward J I. (2016). Cisplatin versus carboplatin: comparative review of therapeutic management in solid malignancies. Crit Rev Oncol Hematol. 102,37-46. doi:10.1016/j.critrevonc.2016.03.014.

Husain K, Whitworth C, Somani SM, Rybak L P. (2001). Carboplatin-induced oxidative stress in rat cochlea. Hear Res. 159(1-2), 14-22. doi:10.1016/S0378-5955(01)00306-9.

Kanat O, Ertas H, Caner B. (2017). Platinum-induced neurotoxicity: a review of possible mechanisms. World J Clin Oncol. 8(4), 329. doi: 10.5306/wjco.v8.i4.329.

Kelles M, Tan M, Kalcioglu MT, Toplu Y, Bulam N. (2014). The protective effect of Chrysin against cisplatin induced ototoxicity in rats. Indian J Otolaryngol Head Neck Surg. 66(4), 369-374. doi:10.1007/s12070-013-0695-x.

Kikuchi H, Yuan B, Hu X, Okazaki M. (2019). Chemopreventive and anticancer activity of flavonoids and its possibility for clinical use by combining with conventional chemotherapeutic agents. Am J Clin Cancer Res. 9(8), 1517.

Kucukler S, Benzer F, Yildirim S, Gur C, Kandemir FM, Bengu AS, Dortbudak, MB. (2020). Protective effects of chrysin against oxidative stress and inflammation induced by lead acetate in rat kidneys: A biochemical and histopathological approach. Biol Trace Elem Res. 1-14. doi:10.1007/s12011-020-02268-8.

Li X, Jankovic J, Le W. (2011). Iron chelation and neuroprotection in neurodegenerative diseases. J Neural Transm. 118:473–7. doi:10.1007/s00702-010-0518-0.

Luo S, Zhou L, Dai Y, Lu Y, Liu Q. (2018). Analysis of Cytotoxic Effects of Chemotherapeutic Agents for Gastrointestinal Cancer with Cell-based Impedance Biosensor. Sens. Mater. 30(9), 1977-1987.

Ma Z, Xu L, Liu D, Zhang X, Di S, Li W, Yan X. (2020). Utilizing Melatonin to Alleviate Side Effects of Chemotherapy: A Potentially Good Partner for Treating Cancer with Ageing. Oxid Med Cell Longev. doi:10.1155/2020/6841581.

Mani R, Natesan V. (2018). Chrysin: Sources, beneficial pharmacological activities, and molecular mechanism of action. Phytochemistry. 145, 187-196. doi:10.1016/j.phytochem.2017.09.016.

Miyano K, Shiraishi S, Minami K, Sudo Y, Suzuki M, Yokoyama T, Uezono Y. (2019). Carboplatin enhances the activity of human transient receptor potential ankyrin 1 through the Cyclic AMP-Protein Kinase AA-Kinase Anchoring Protein (AKAP) Pathways. Int J Mol Mol Sci. 20(13), 3271. doi:10.3390/ijms20133271.

Özbolat SN, Ayna A. (2020). Chrysin suppresses HT-29 cell death induced by diclofenac through apoptosis and oxidative damage. Nutr Cancer. 1-10. doi:10.1080/01635581.2020.1801775.

Shooshtari M K, Sarkaki A, Mansouri SMT, Badavi M, Khorsandi L, Dehcheshmeh MG, Farbood Y. (2020). Protective effects of Chrysin against memory impairment, cerebral hyperemia and oxidative stress after cerebral hypoperfusion and reperfusion in rats. Metab Brain Dis. 35(2), 401-412. doi:10.1007/s11011-019-00527-9.

Simon HU, Haj-Yehia A, Levi-Schaffer F. (2000). Role of reactive oxygen species (ROS) in apoptosis induction. Apoptosis. 5(5), 415-418. doi:10.1023/A:1009616228304.

Su L J, Zhang JH, Gomez H, Murugan R, Hong X, Xu D, Peng ZY. (2019). Reactive oxygen species-induced lipid peroxidation in apoptosis, autophagy, and ferroptosis. Oxid Med Cell Longev. 2019(1), 5080843.17. doi:10.1155/2019/5080843.

Taslimi P, Kandemir FM, Demir Y, İleritürk M, Temel Y, Caglayan C, Gulçin İ. (2019). The antidiabetic and anticholinergic effects of chrysin on cyclophosphamide-induced multiple organ toxicity in rats: Pharmacological evaluation of some metabolic enzyme activities. J Biochem Mol Toxicol. 33(6), e22313. doi:10.1002/jbt.22313.

Temel Y, Kucukler S, Yıldırım S, Caglayan C, Kandemir FM. (2020). Protective effect of chrysin on cyclophosphamide-induced hepatotoxicity and nephrotoxicity via the inhibition of oxidative stress, inflammation, and apoptosis. Naunyn Schmiedebergs Arch Pharmacol. 393, 325-337. doi:10.1007/s00210-019-01741-z.

Temel Y, Çağlayan C, Ahmed BM, Kandemir FM, Çiftci M. (2021). The effects of chrysin and naringin on cyclophosphamide-induced erythrocyte damage in rats: biochemical evaluation of some enzyme activities in vivo and in vitro. Naunyn Schmiedebergs Arch Pharmacol. 394, 645-654. doi:10.1007/s00210-020-01987-y.

Yousef MI, Khalil DK, Abdou H M. (2018). Neuro-and nephroprotective effect of grape seed proanthocyanidin extract against carboplatin and thalidomide through modulation of inflammation, tumor suppressor protein p53, neurotransmitters, oxidative stress and histology. Toxicol Rep. 5, 568-578. doi:10.1016/j.toxrep.2018.04.006.

Zhivotosky, B., & Orrenius, S. (2001). Assessment of apoptosis and necrosis by DNA fragmentation and morphological criteria. Curr Protoc Cell Biol, 12(1), 18-3.

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