



The 9th International Electronic Conference on Medicinal Chemistry (ECMC 2023)

01–30 November 2023 | Online

Beta-blockers as potential adjuvants in chemotherapy against melanoma: an *in vitro* study

Chaired by **Dr. Alfredo Berzal-Herranz**
and **Prof. Dr. Maria Emília Sousa**



pharmaceuticals



Laura Rama^{1,*}, Mónica Almeida², Carolina Frazão², Maria de Lourdes Pereira³, Miguel Oliveira²

¹ Department of Biology, University of Aveiro, 3810-193, Aveiro, Portugal

² Centre for Environmental and Marine Studies (CESAM), Department of Biology, University of Aveiro, 3810-193 Aveiro, Portugal

³ Department of Medical Sciences, CICECO—Aveiro Institute of Materials, University of Aveiro, 3810-193 Aveiro, Portugal

* Corresponding author: laurasofiarama@ua.pt





Beta-blockers as potential adjuvants in chemotherapy against melanoma: an *in vitro* study

Individual exposure

β 1 selective beta-blockers

- Atenolol
- Metoprolol

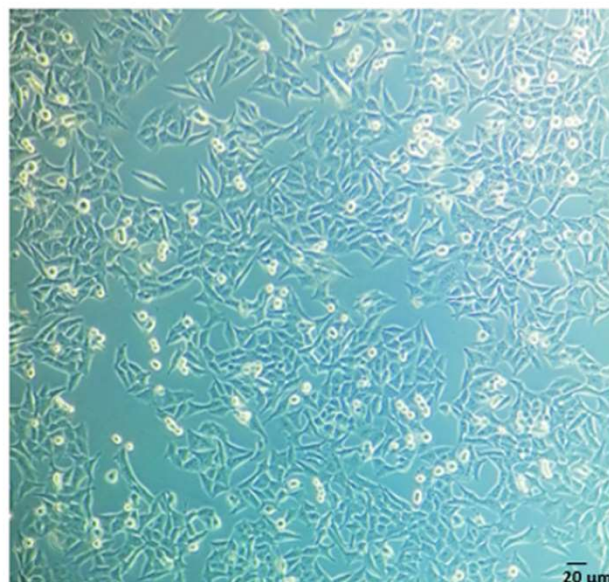
Non-selective beta-blockers

- Carvedilol
- Propranolol

Antineoplastics

- Cisplatin
- 5-fluorouracil

Determination of lethal
concentrations (LC)



A375

melanoma cells

Cell viability

MTT and resazurin assays

Combined exposure

Cisplatin + carvedilol

Cisplatin + propranolol

Carvedilol + propranolol

Cisplatin + metoprolol

Interaction of drugs





The 9th International Electronic Conference on Medicinal Chemistry

01–30 November 2023 | Online



Abstract: Melanoma is an aggressive type of skin cancer, with the number of cases expected to increase in the future. The available treatments show low efficiency highlighting the need to develop new therapies to increase the survival of the patients. Beta-blockers, drugs already known and used for heart conditions; have shown anti-cancer properties and potential to be valuable in conjugation with chemotherapy. This study aimed to evaluate, in vitro, their potential for cancer treatment. A375 cells (melanoma cell line) were exposed to non-selective blockers (carvedilol and propranolol), β 1 selective blockers (atenolol and metoprolol), and antineoplastics drugs (cisplatin and 5-fluorouracil), and viability assessed at 3 timepoints. Selective beta-1 blockers had no significant effects on cell viability. However, the other tested pharmaceuticals affected cell viability allowing the determination of median lethal concentrations (LC50) at 72h and a toxicity ranking: cisplatin (2.46 (1.87 – 3.38)), 5-fluorouracil (4.77 (4.48 – 5.07)), carvedilol (16.91 (15.47 - 18.99)) and propranolol (58.03 (57.08 - 59.11)). Carvedilol and cisplatin were, respectively, the most toxic beta-blocker and antineoplastic. Following these results, a combined exposure of beta-blockers and antineoplastics was performed: cisplatin with metoprolol, propranolol, and carvedilol and also paired both non-selective beta-blockers. The results so far support the potential use of non-selective β -blockers as adjuvants of chemotherapy as a melanoma treatment.

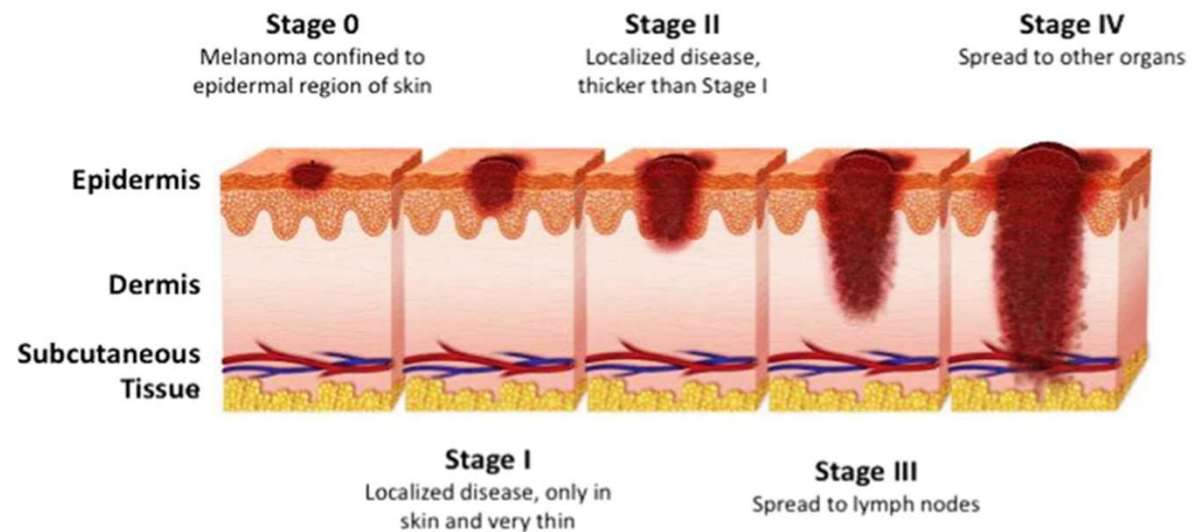
Keywords: melanoma; cancer cell lines; beta-blockers; drug repurposing; combined exposure





Introduction

Melanoma



Mutation in the DNA of melanocytes
causes abnormal growth





Introduction

Epidemiology

2020		324 635 new cases
		57 043 deaths

Source: Globocan

Risk factors

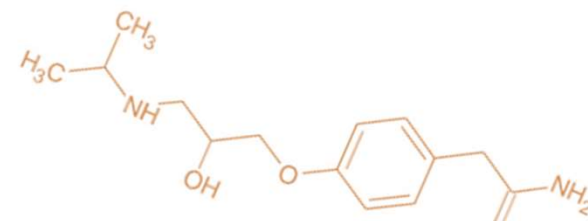
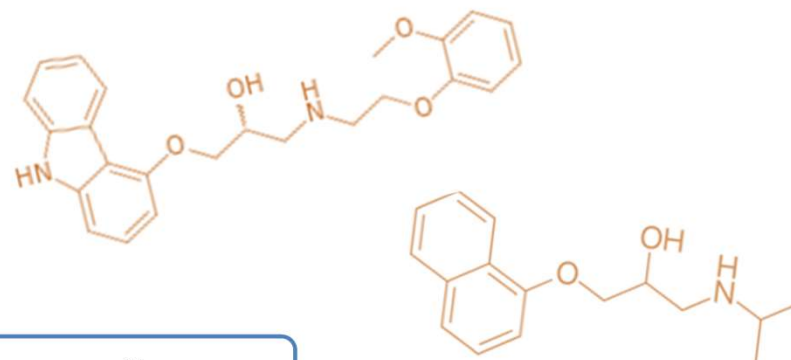




Introduction

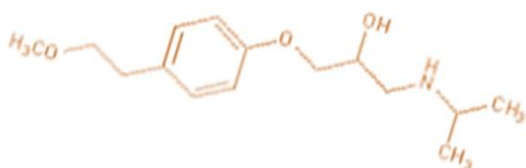
Drug repurposing

A drug that already exists might have a new therapeutic use.



Beta-blockers are used to treat heart diseases

They connect to beta-adrenergic receptors in cells





Objective

Evaluate the effects of different beta-blockers and antineoplastics in melanoma cancer cells in individual and combined exposure

β 1 selective beta-blockers

- Atenolol
- Metoprolol

Non-selective beta-blockers

- Carvedilol
- Propranolol

Antineoplastics

- Cisplatin
- 5-fluorouracil



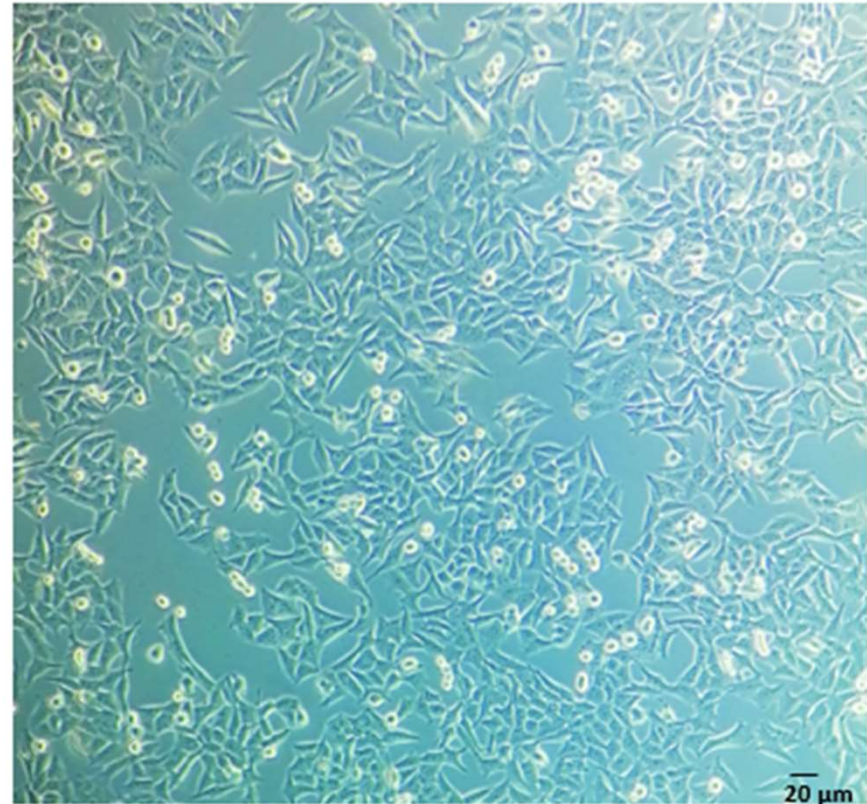


Experimental design

Biological model

A375

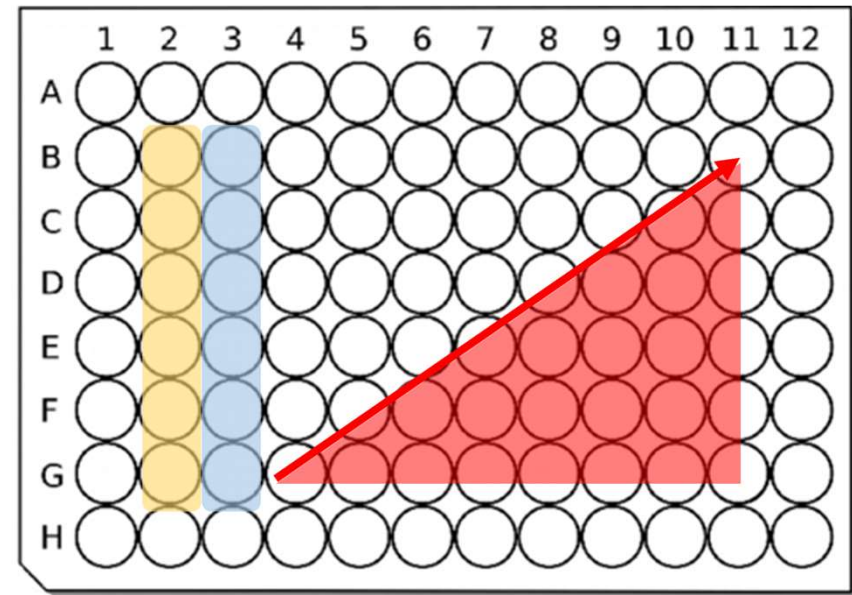
melanoma epithelial cells





Experimental design




Individual exposures



Timepoints: 24h 48h 72h

Viability assays: MTT and resazurin

3 replicates

-  Blank control
-  Negative control
-  Concentrations





Experimental design

Combined exposures

	0	0.25	0.5	0.75	1
0					
0.25					
0.5					
0.75					
1					

Timepoints: 48h

1 toxic unit = LC_{50} of each pharmaceutical

Viability assays: MTT

3 replicates for each condition





Experimental design

Data Analysis



Viability expressed as percentage of control

Non-linear regression (4 parameters)

LCs with confidence intervals

Two-Way ANOVA

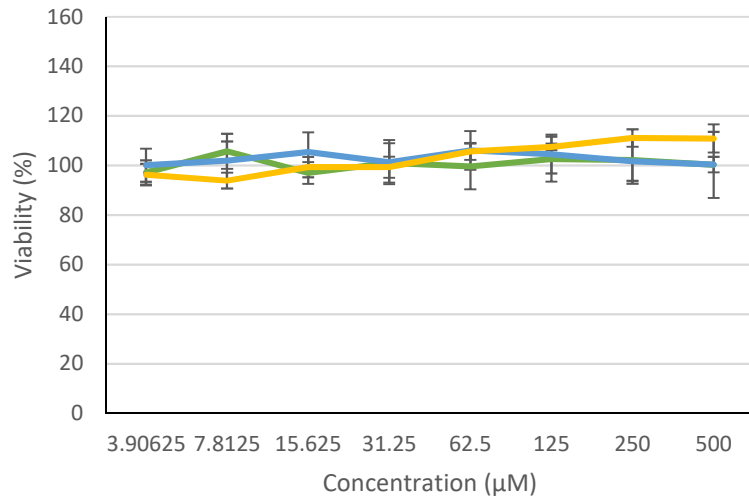




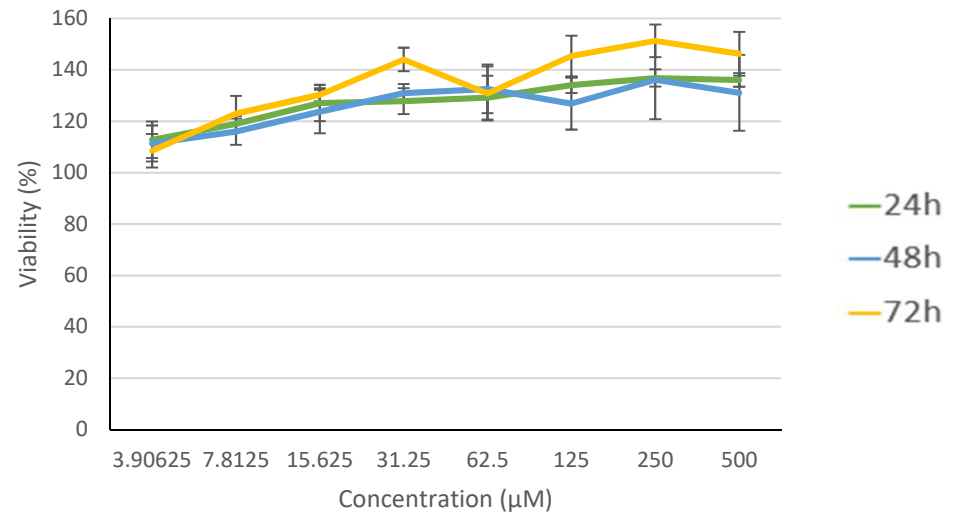
Results and discussion

Atenolol

MTT



Resazurin



No significant effect

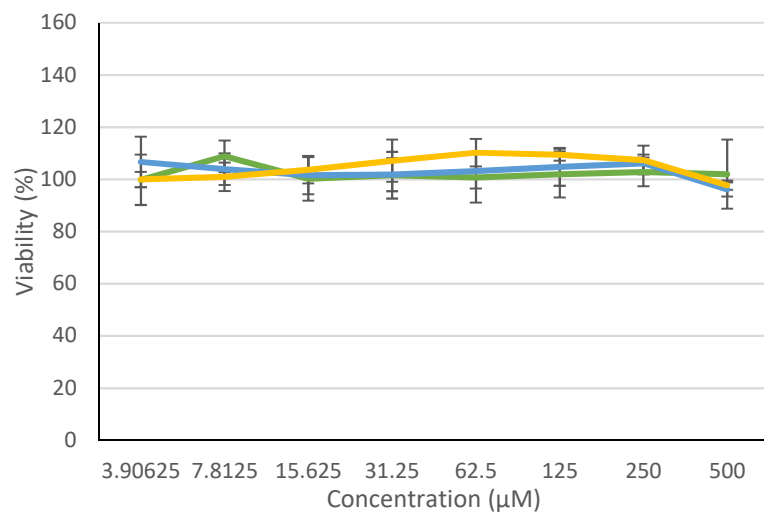




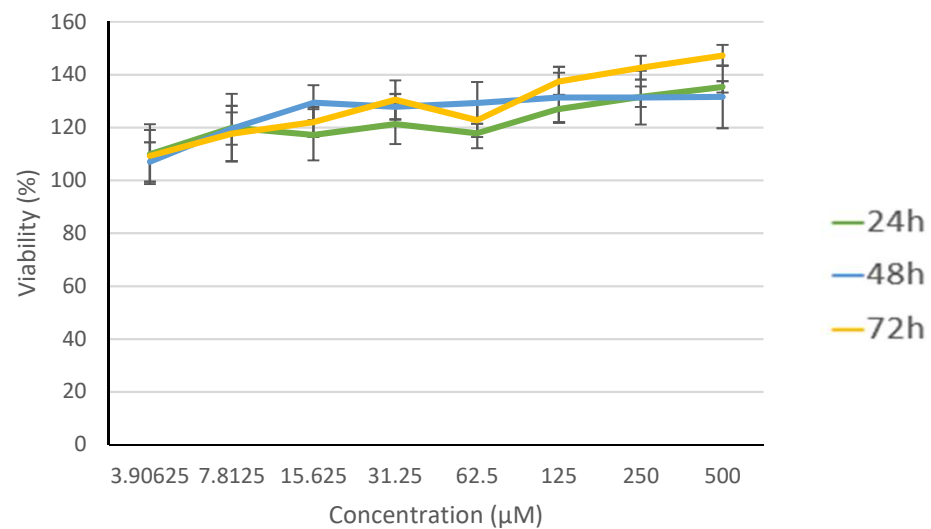
Results and discussion

Metoprolol

MTT



Resazurin



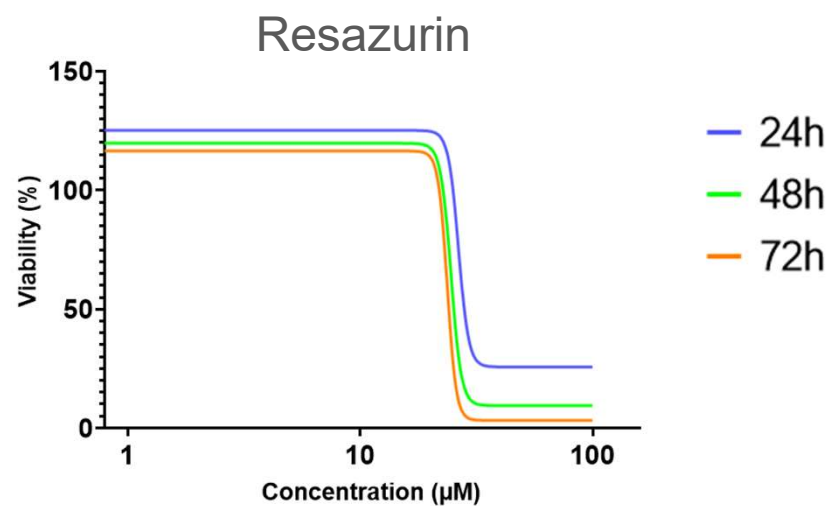
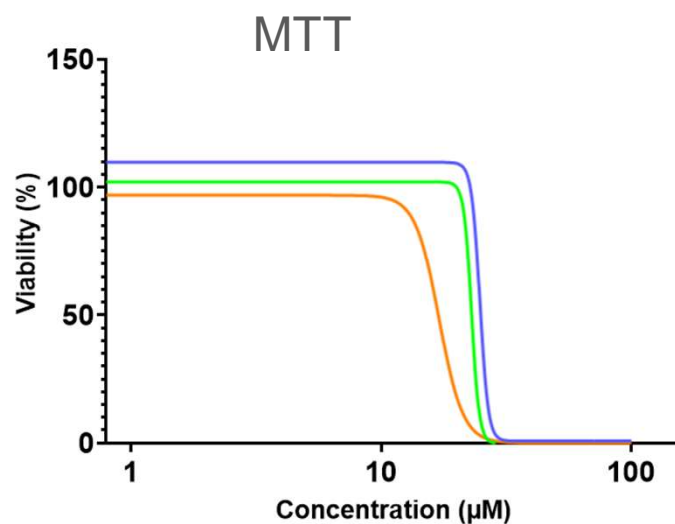
No significant effect





Results and discussion

Carvedilol



Time dependent effects

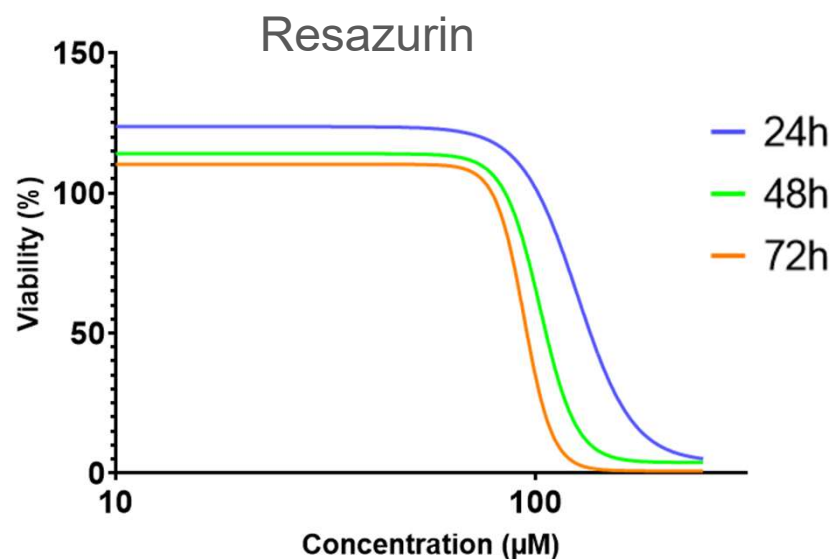
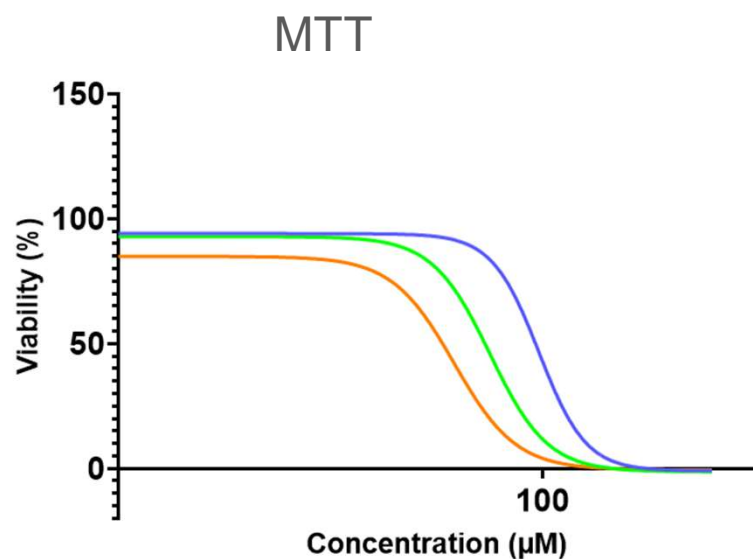
LC ₅₀	24h	48h	72h
MTT	24.95	22.97	16.91
Resazurin	27.97	25.21	23.99





Results and discussion

Propranolol



Time dependent effects

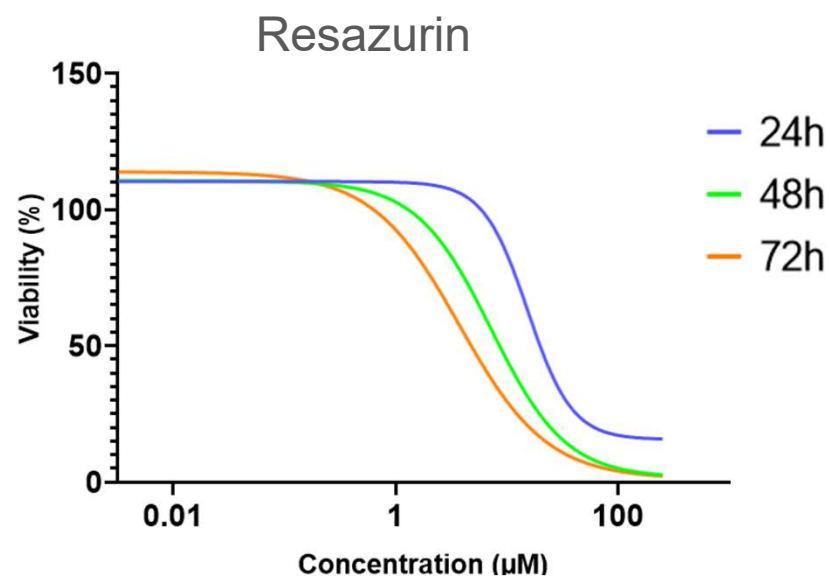
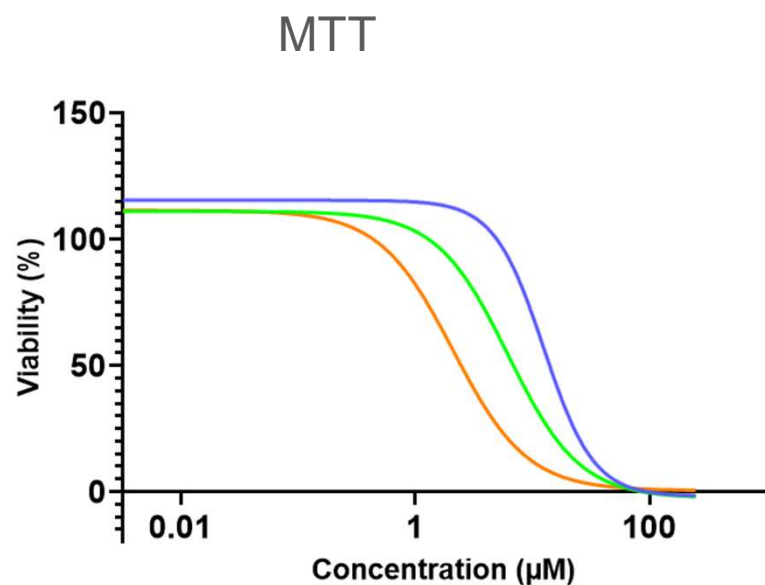
LC ₅₀	24h	48h	72h
MTT	96.04	73.00	58.03
Resazurin	134.81	105.75	95.28





Results and discussion

Cisplatin



Time dependent effects

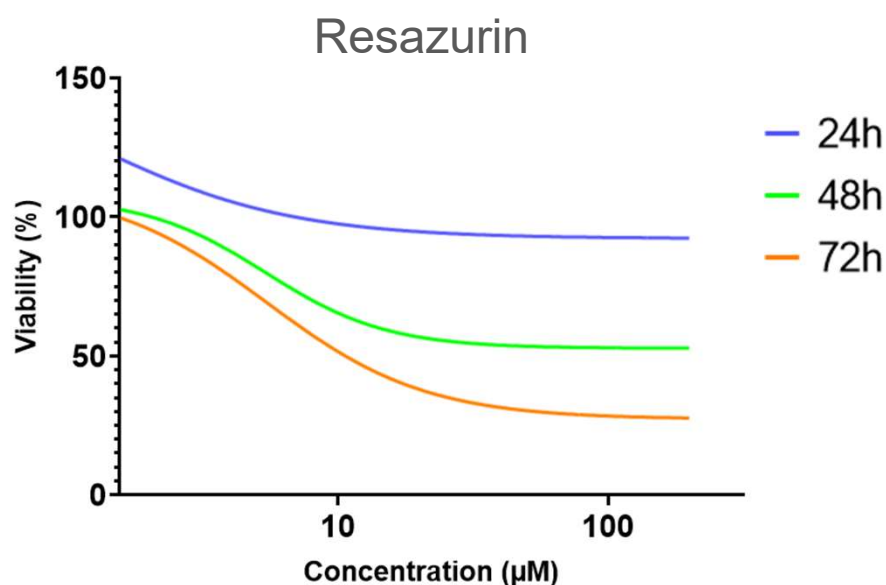
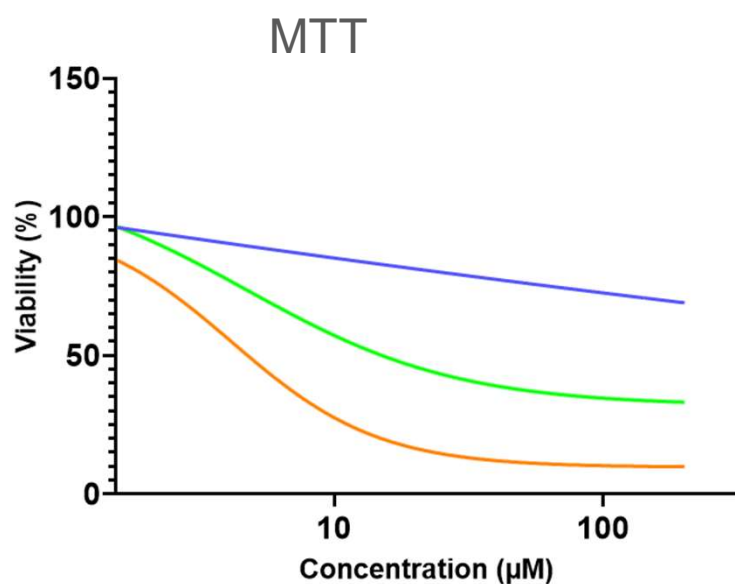
LC ₅₀	24h	48h	72h
MTT	14.01	6.87	2.46
Resazurin	19.79	8.56	4.76





Results and discussion

5-fluorouracil



Time dependent

	LC ₅₀	24h	48h	72h
MTT		--	15.10	4.77
Resazurin		--	--	10.64

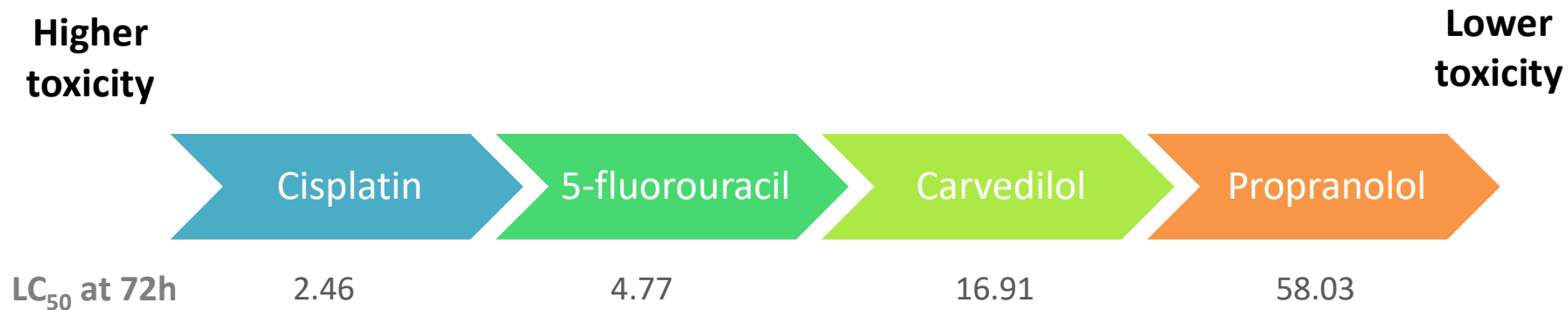




Results and discussion

Sensitivity comparison

Atenolol and metoprolol had no significant effect

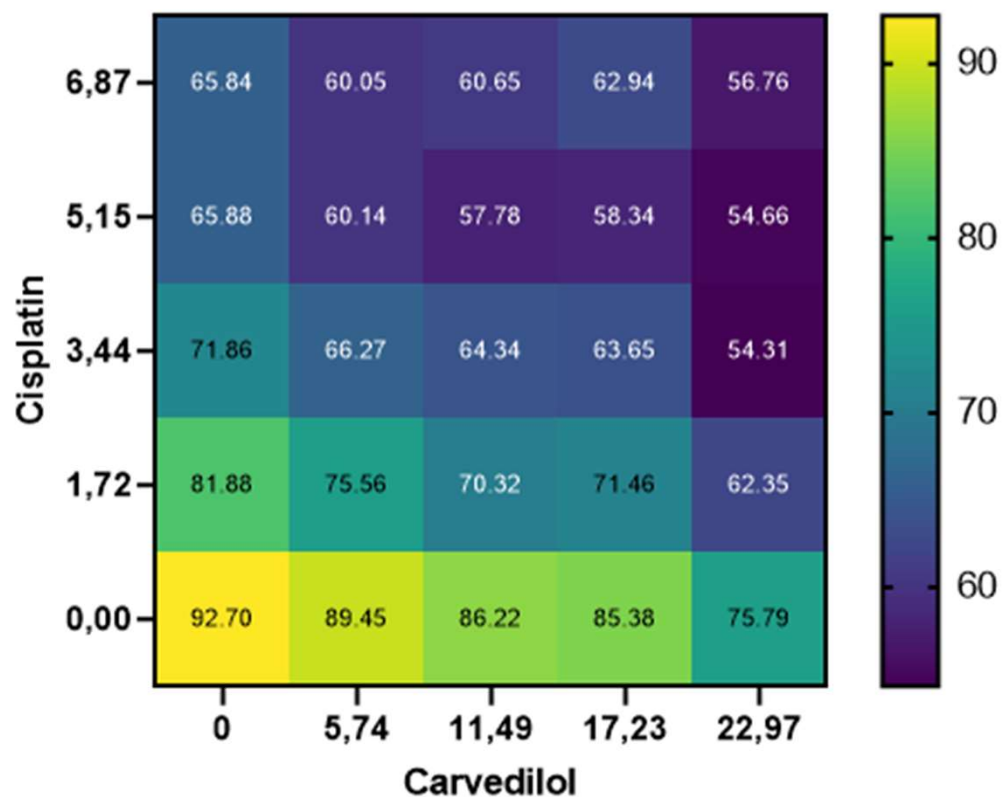




Results and discussion

Cisplatin + Carvedilol

Synergistic effect



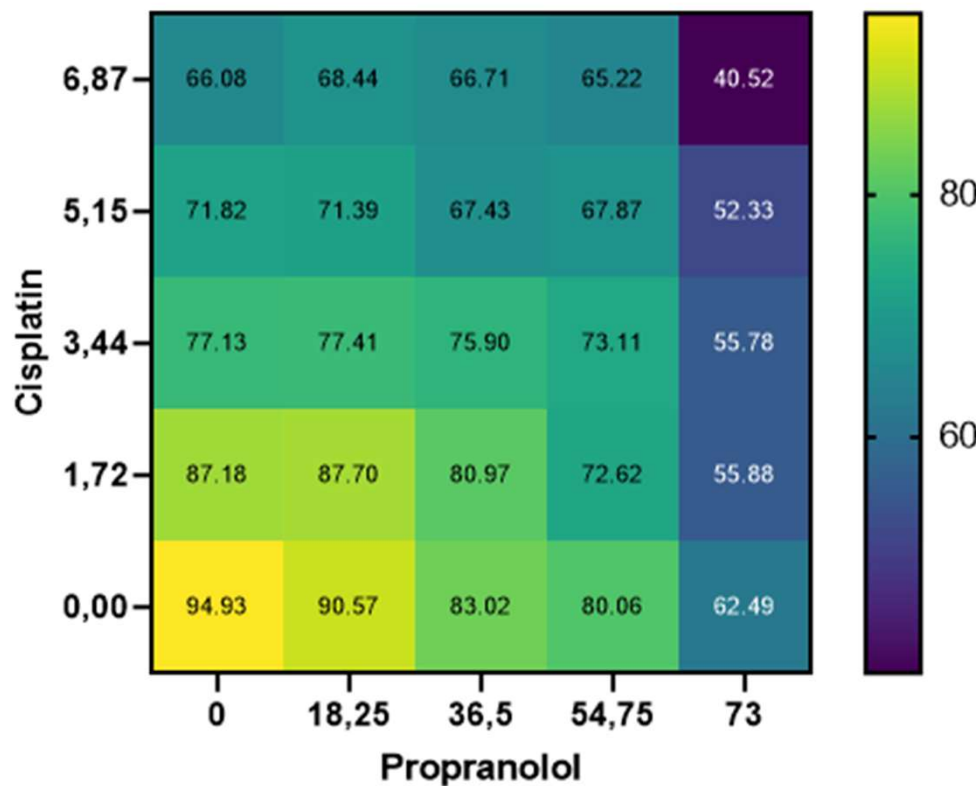


Results and discussion

Cisplatin + Propranolol

Highest propranolol
concentration had a bigger
influence

Synergistic effect



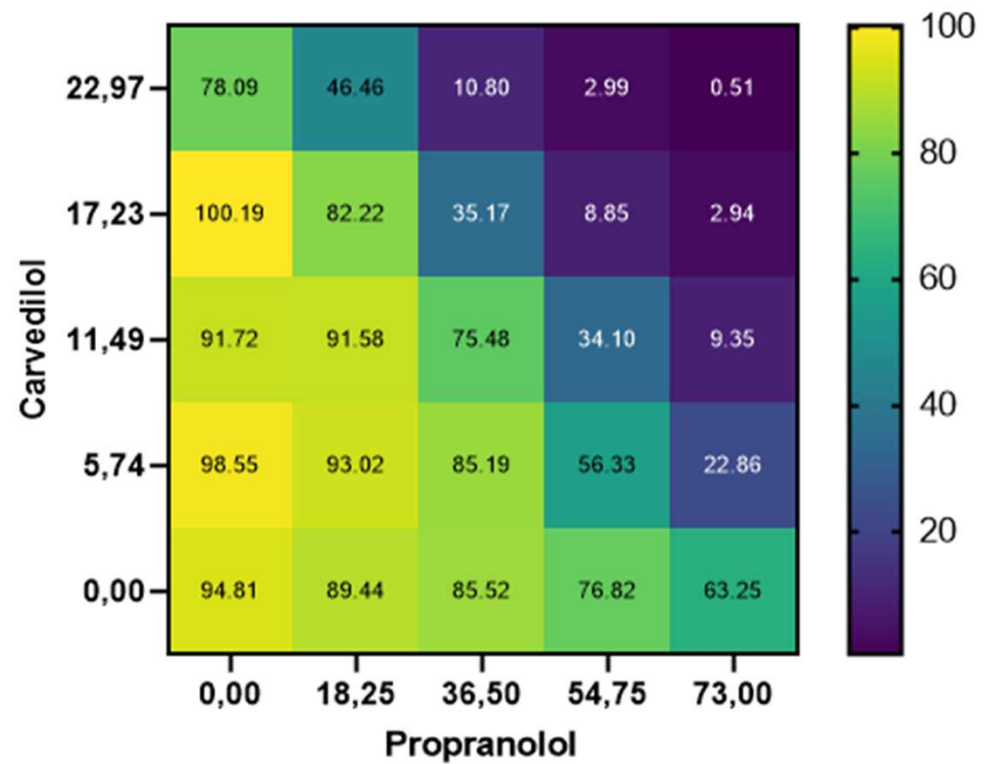


Results and discussion

Carvedilol + Propranolol

No interaction

Additive effect

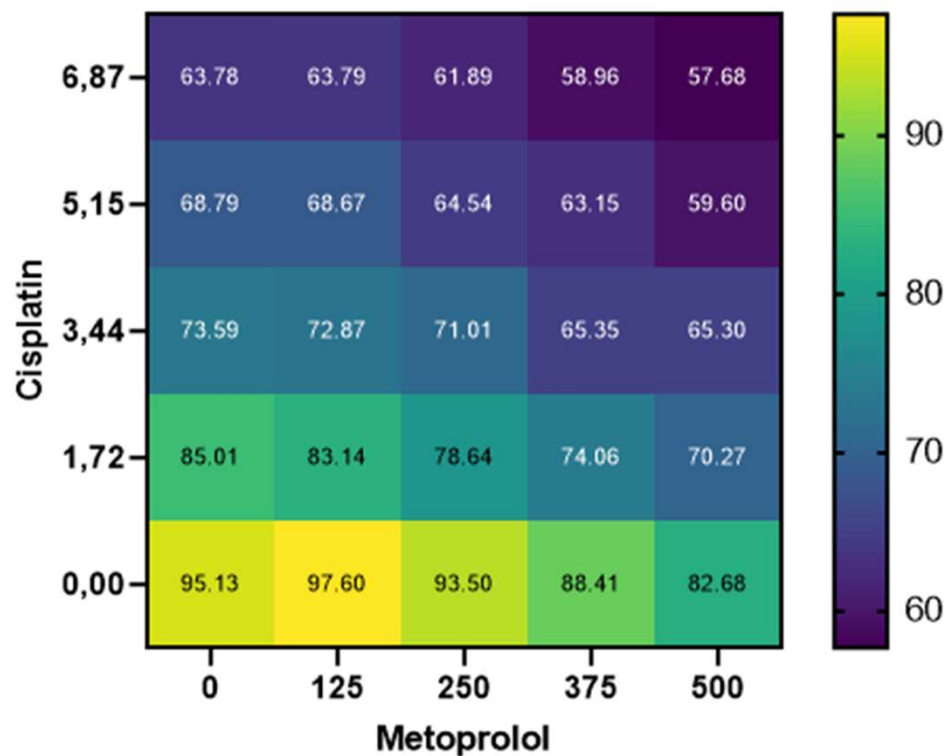




Results and discussion

Cisplatin + Metoprolol

Antagonistic effect





Conclusion

Non-selective beta blockers
help increase cisplatin toxicity

Metoprolol interacts with cisplatin and
diminishes its effect



Future perspective: Normal cell line





The 9th International Electronic Conference on Medicinal Chemistry

01-30 November 2023 | Online



Acknowledgments



Thanks are due to FCT/MCTES for the financial support to the project NanoPlanet (2022.02340.PTDC), and CESAM (UIDP/50017/2020 + UIDB/50017/2020 + LA/P/0094/2020) through national funds.

