The 8th International Electronic Conference on Medicinal Chemistry (ECMC 2022)
01–30 NOVEMBER 2022 | ONLINE

BDDE-Inspired Chalcone Derivatives as New Antimicrobial Adjuvants

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pharmaceuticals MDPI

Ana Jesus [1,2,†], Fernando Durães [1,2,†], Nikoletta Szemerédi [3], Joana Freitas-Silva [2,4], Paulo Martins Costa [2,4], Eugénia Pinto [2,5], Madalena Pinto [1,2], Gabriella Spengler [3], Emília Sousa [1,2,*], Honorina Cidade [1,2,*]

[1] Laboratory of Organic and Pharmaceutical Chemistry, Department of Chemical Sciences, Faculty of Pharmacy, University of Porto, 4050-313 Porto, Portugal;
[2] CIIMAR—Interdisciplinary Centre of Marine and Environmental Research, Avenida General Norton de Matos, S/N, 4450-208 Matosinhos, Portugal;
[3] Department of Medical Microbiology, Albert Szent-Györgyi Health Center and Albert Szent-Györgyi Medical School, University of Szeged, Semmelweis utca 6, 6725 Szeged, Hungary;
[4] ICBAS—Institute of Biomedical Sciences Abel Salazar, University of Porto, Rua de Jorge Viterbo Ferreira 228, 4050-313 Porto, Portugal
[5] Laboratory of Microbiology, Department of Biological Sciences, Faculty of Pharmacy, University of Porto, Rua de Jorge Viterbo Ferreira 228, 4050-313 Porto, Portugal

† These authors contributed equally to this work.
* Corresponding author: esousa@ff.up.pt and hcidade@ff.up.pt

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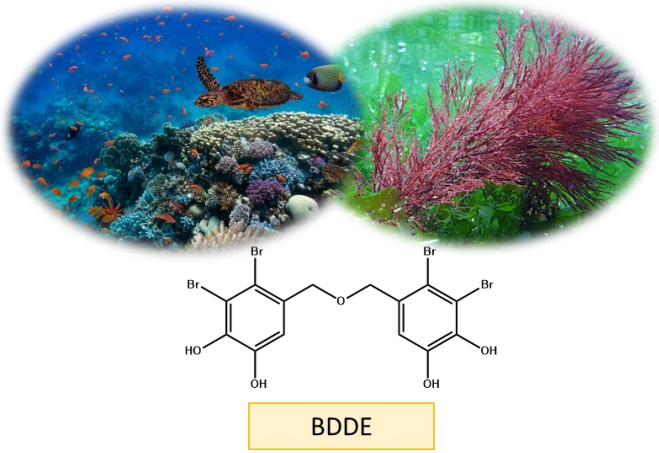
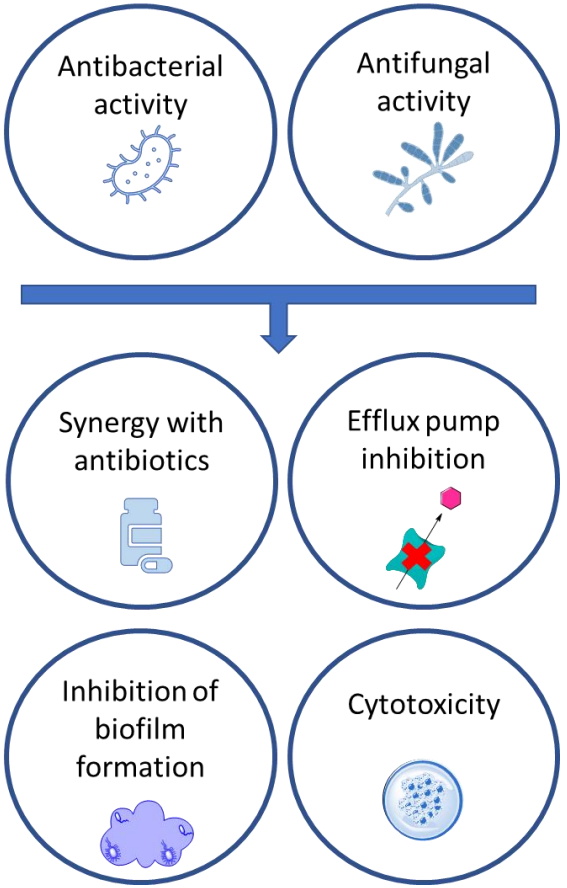
BDDE-Inspired Chalcone Derivatives as New Antimicrobial Adjuvants

Graphical Abstract





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Abstract: The effective response of antibiotics is threatened by the proliferation of microorganisms that manifest resistance mechanisms, leading to an increase of progressively untreatable infectious diseases around the world. One solution to this problem could lie in shifting the strategy from searching for new antibacterials to discover new compounds that potentiate the antimicrobial activity of current antibiotics, therefore reverting resistance, through the interference with several mechanisms, including biofilm formation and efflux pumps (EPs). Using bis(2,3-dibromo-4,5-dihydroxybenzyl) ether (BDDE) as a template, a macroalgae brominated bromophenol with antimicrobial activity, a series of 18 chalcone derivatives was prepared and evaluated for its antimicrobial activity and potential to fight antibiotic resistance. This includes seven chalcones, six dihydrochalcones and five diarylpropanes. Among them, two chalcones exhibited interesting antifungal activity and all compounds reversed resistance to vancomycin in the environmental isolate *Enterococcus faecalis* B3/101. Three compounds caused a four-fold decrease in the minimum inhibitory concentration (MIC) values of vancomycin against *E. faecalis*. All the dihydrochalcones and diarylpropanes displayed inhibition of EPs and biofilm formation in the tested multidrug resistant strain, suggesting that these compounds are EP inhibitors. Notably, dihydro-chalcones and diarylpropanes did not show cytotoxicity in a mouse embryonic fibroblast cell line and they can potentially be regarded as hits for bacterial EPs inhibition.

Keywords: antibiotic resistance; BDDE; halogenated chalcone derivatives; antimicrobial activity; EPs inhibitors

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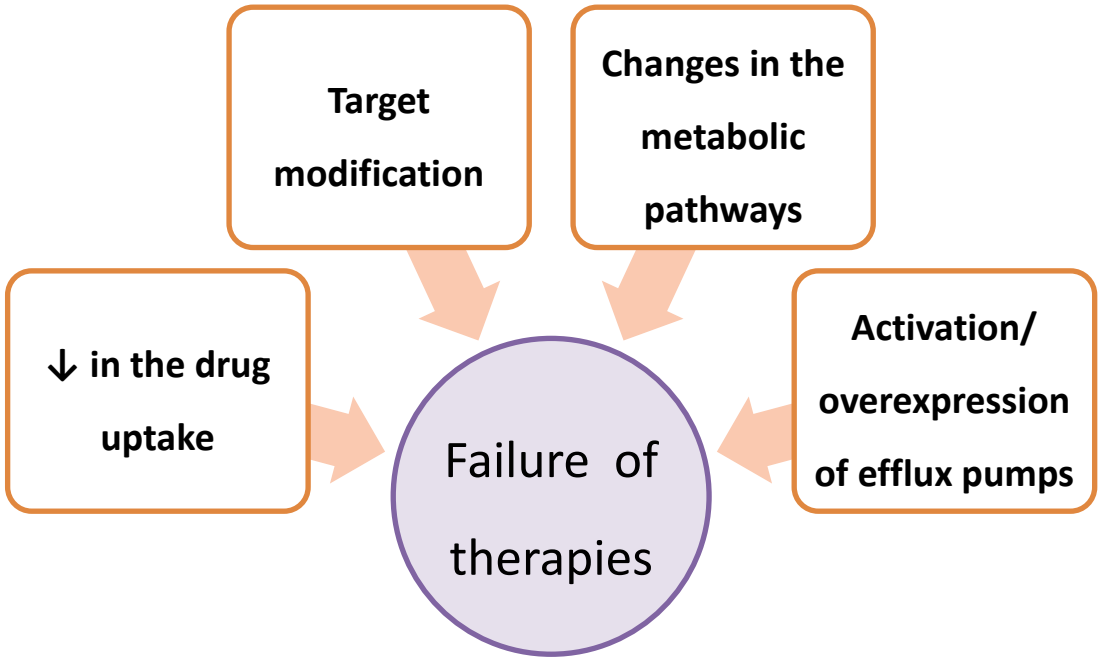
Introduction

Antibiotic resistance

- **Ability of a microorganism to resist to the action of a drug**



Durães F., Pinto M., Sousa E., Current Medicinal Chemistry **2018**, 25(42), 6030-6069
Jesus, A.; Durães, F.; Szemerédi, N.; Freitas-Silva, J.; da Costa, P.M.; Pinto, E.; Pinto, M.; Spengler, G.; Sousa, E.; Cidade, H. BDDE-Inspired Chalcone Derivatives to Fight Bacterial and Fungal Infections. Marine Drugs **2022**, 20, 315.

Introduction

Marine environment

Macroalgae

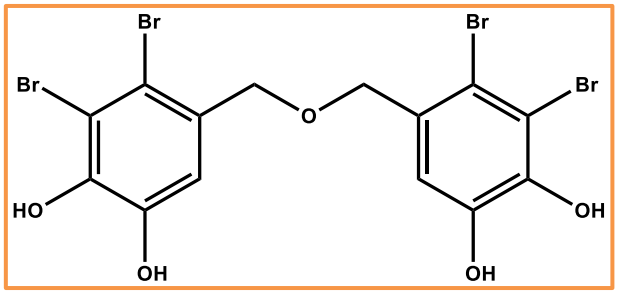
- Great biodiversity
- Discovery of new bioactive compounds

- Bioactive secondary metabolites
- Diversity of chemical structures

Antifungal

4 fungi

% inhibition = 65-80 %



Antibacterial

8 bacterial strains

MIC = 35-140 µg/mL

bis(2,3-dibromo-4,5-dihydroxybenzyl)ether (BDDE)

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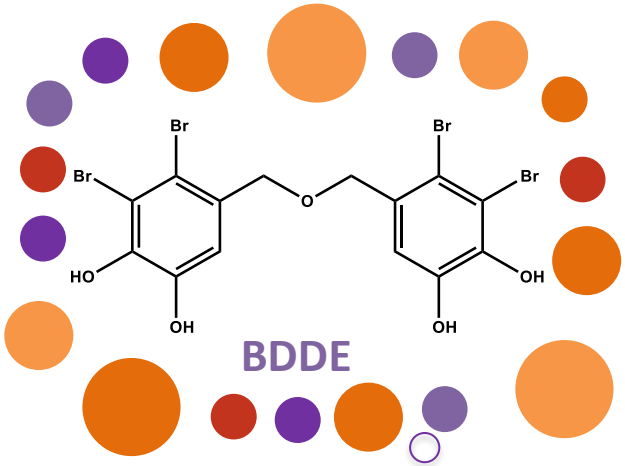
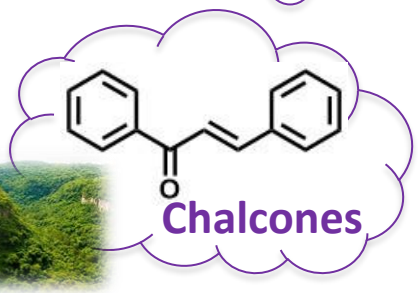
Aim





New BDDE analogues with a chalcone scaffold with antibacterial activity

Synthesis of brominated chalcone derivatives

Evaluation of the antibacterial activity

- **Wide range of bioactivities**
- **Similar structural moieties (two aromatic groups and a linker with 3 carbon atoms)**

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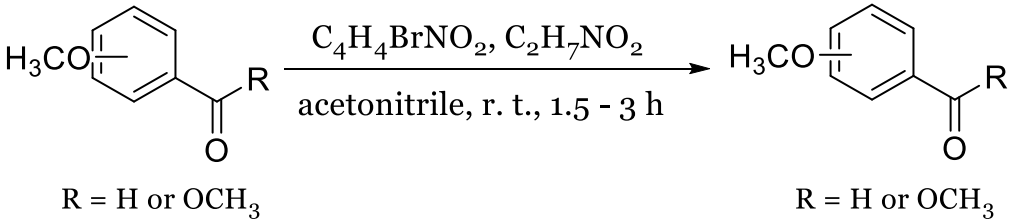
Results and discussion

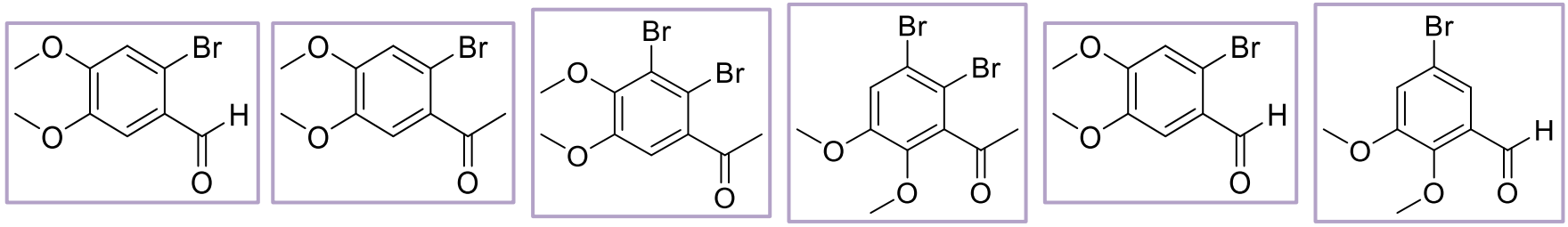
1. Synthesis

Bromination of acetophenones and benzaldehydes

- **With *N*-bromossucimide (NBS)**



R = H or OCH_3 R = H or OCH_3



34OMBBr	34OMABr	34OMABr2	23OMABr2	34OMBBr	23OMBBr
η = 52%	**η = 63%**	**η = 1%**	**η = 52%**	**η = 52%**	**η = 53%**

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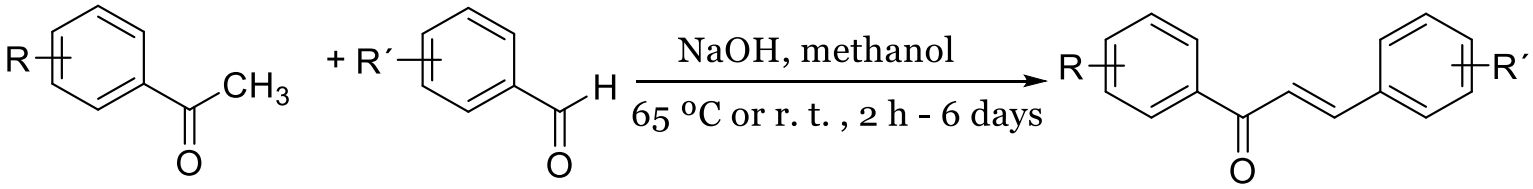
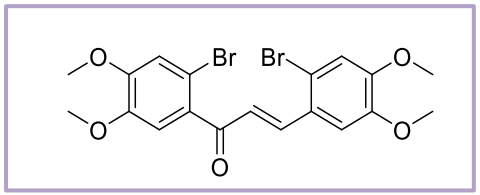
Results and discussion

1. Synthesis

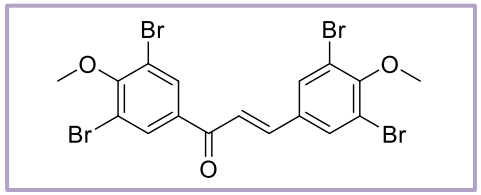
Synthesis of brominated chalcones





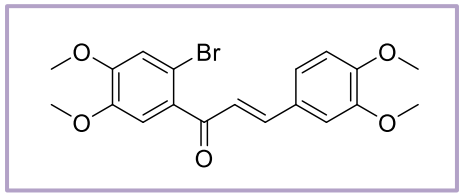
34CBr2

η = 87%



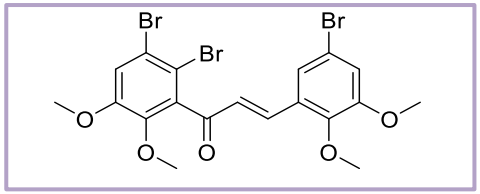
35CBr4

η = 36%



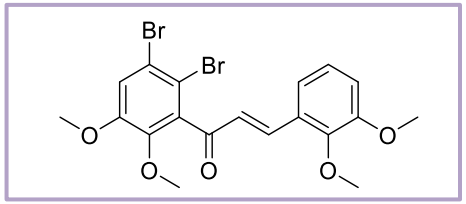
34CR2

η = 80%



23CBr3

η = 55%



23CR1

η = 48%

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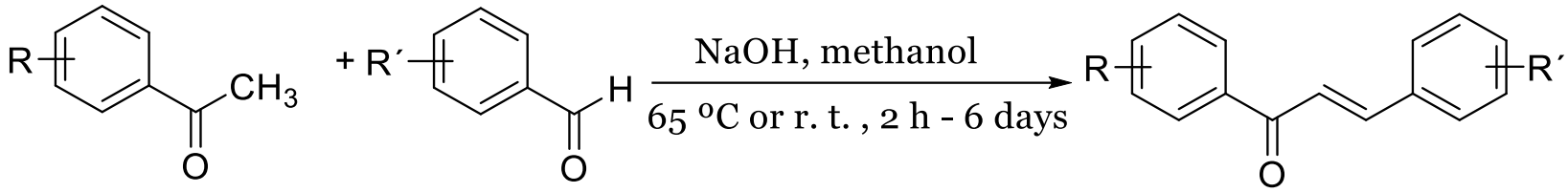
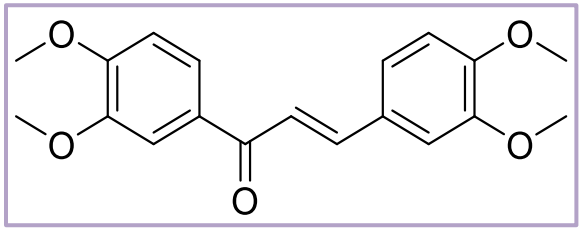
Results and discussion

1. Synthesis

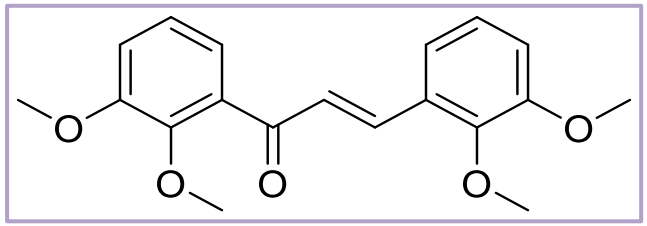
Synthesis of non-brominated chalcones





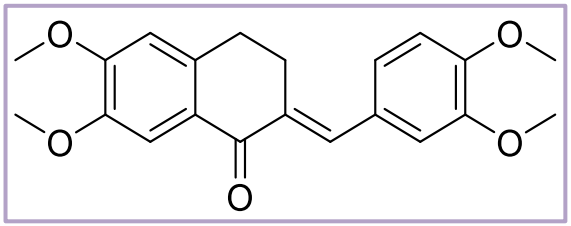
34OMC

η = 74%



23OMC

η = 85%



6734OMC

η = 54%

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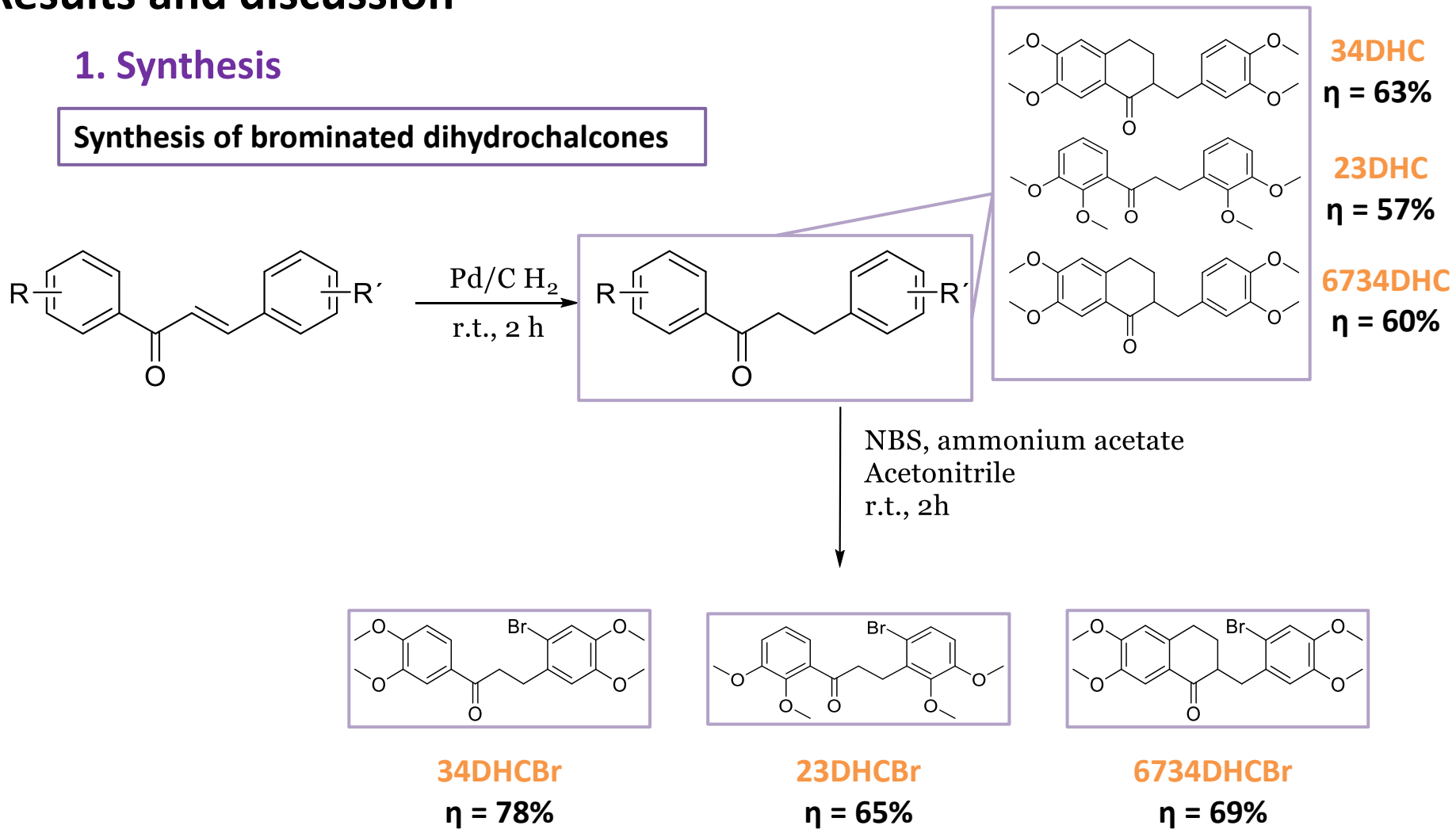
Results and discussion

1. Synthesis

Synthesis of brominated dihydrochalcones



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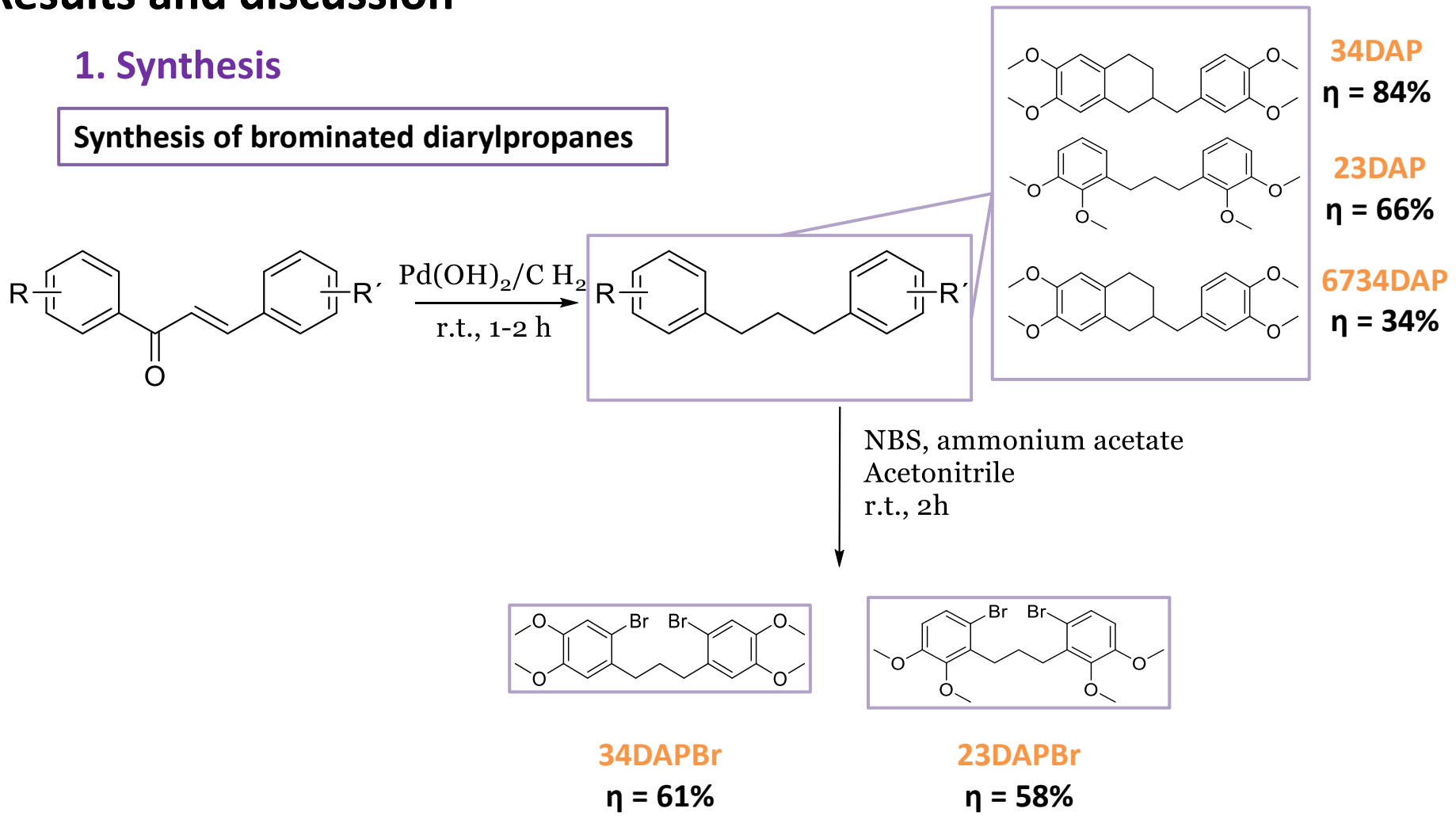
Results and discussion

1. Synthesis

Synthesis of brominated diarylpropanes



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Results and discussion

2. Biological Activity

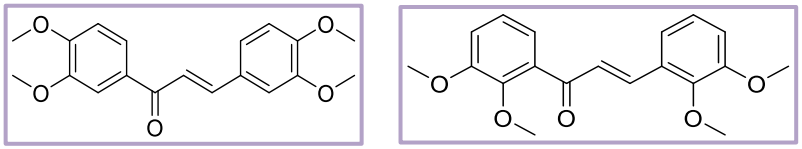
Antifungal activity

Candida albicans ATCC 10231

Aspergillus fumigatus ATCC 204305

Trichophyton rubrum FF5

Chalcones



Active at the maximum concentration tested 128 µg/mL

Dihydrochalcones and diarylpropanes

Did not reveal antifungal activity

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Results and discussion

2. Biological Activity

Antibacterial activity and synergism

- None of the chalcone derivatives showed antibacterial activity (MIC > 64 µg/mL)
- All the compounds exhibited synergy with VAN against *E. faecalis* B3/101
- Fourteen compounds revealed synergy with CTX against *E. coli* SA/2

Compounds	Antibacterial Activity and Potentiation of Antimicrobials (CTX: Cefotaxime; VAN: Vancomycin)	
	E. coli SA/2 MIC CTX = 562 µM	E. faecalis B3/101 MIC VAN = 707 µM
	MIC Reduction	MIC Reduction
34CBr2	2-fold	4-fold
34CR2	No effect	2-fold
23CBr3	No effect	2-fold
23CR1	2-fold	2-fold
34OMC	2-fold	2-fold
23OMC	2-fold	4-fold
6734OMC	2-fold	2-fold
34DHC	2-fold	2-fold
23DHC	2-fold	2-fold
6734DHC	2-fold	2-fold
34DAP	2-fold	2-fold
23DAP	2-fold	2-fold
6734DAP	2-fold	2-fold
34DHCBr	2-fold	2-fold
23DHCBr	2-fold	2-fold
6734DHCBr	No effect	4-fold
34DAPBr	2-fold	2-fold
23DAAPBr	No effect	2-fold

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Results and discussion

2. Biological Activity

Efflux and Biofilm Inhibition

- **All the compounds were tested at concentration of 50 µM**
- **↑ fluorescence → inhibition of ethidium bromide efflux → could be attributed to the inibition of EP**

Compounds	EP Inhibition Assay	Inhibition of Biofilm Formation (%)	
	Relative Fluorescence Index (RFI) ± SD		
	S. aureus 272123	*S. aureus* ATCC 25923	*S. aureus* 272123
34CBr2	0.31 ± 0.03	-	-
34CR2	0.29 ± 0.09	-	-
23CBr3	0.37 ± 0.08	-	-
23CR1	0.66 ± 0.09	3.05 ± 1.41	7.76 ± 2.14
34OMC	0.32 ± 0.04	-	-
23OMC	0.81 ± 0.06	0	87.28 ± 3.84
6734OMC	0.37 ± 0.03	-	-
34DHC	0.23 ± 0.01	-	-
23DHC	0.75 ± 0.04	0	6.89 ± 2.41
6734DHC	0.43 ± 0.02	0	8.15 ± 0.64
34DAP	0.53 ± 0.04	0	49.59 ± 0.39
23DAP	1.30 ± 0.08	0	23.80 ± 0.13
6734DAP	0.70 ± 0.07	0	71.33 ± 1.09
34DHCBr	0.88 ± 0.05	13.28 ± 6.67	7.95 ± 0.65
23DHCBr	1.20 ± 0.01	0	7.08 ± 2.72
6734DHCBr	0.67 ± 0.04	0	10.65 ± 0.76
34DAPBr	0.18 ± 0.02	-	-
23DAAPBr	0.22 ± 0.05	-	-
Control (Reserpine)	0.41 ± 0.01	2.49 ± 1.38	77.62 ± 4.08

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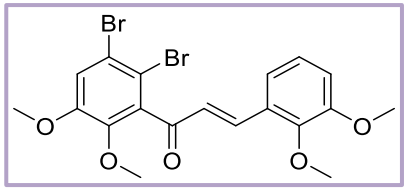
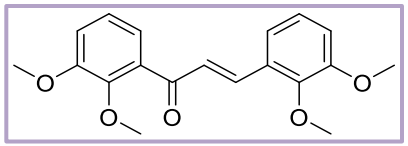
Results and discussion

2. Biological Activity

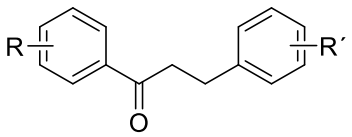
Cytotoxicity in NIH/3T3 mouse embryonic fibroblast cell line

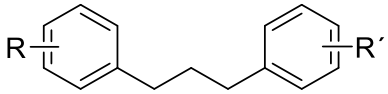




Chalcones

Dihydrochalcones and diarylpropanes





Name	IC_{50} (µM) ± SD
Doxorubicin	12.05 ± 0.81
23CR2	28.31 ± 0,25
23OMC	30.16 ± 1,04
23DHC	>100
6734DHC	>100
34DHCBr	>100
23DHCBr	>100
6734DHCBr	>100
34DAP	>100
23DAP	>100
6734DAP	>100

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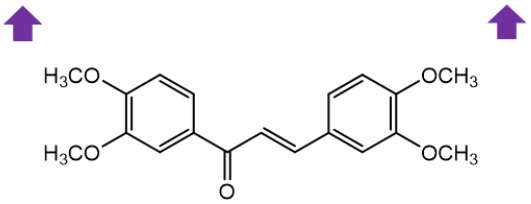
Conclusion

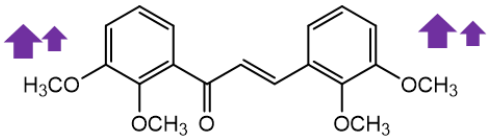
Main Structure-Activity Relationship (SAR) conclusions

Antifungal activity

Only chalcones 13 and 14 were active



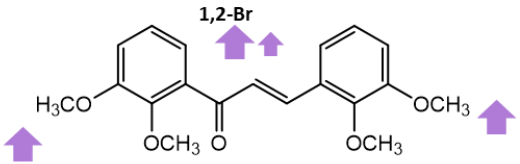
34OMC



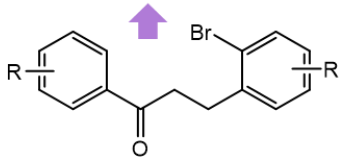
23OMC

OCH_3 are favourable to the activity
Bromo did not potentiate the activity

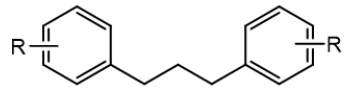
EPs inhibithion



23CR1 and 23OMC



34DHCBr, 23DHCBr, and 6734DHCBr

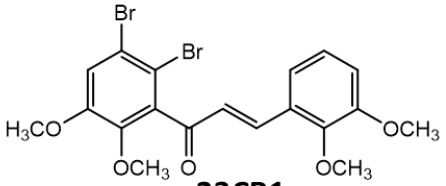


34DAP, 23DAP, and 6734DAP

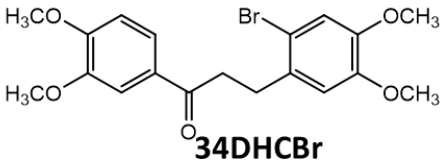
2-Br in DHC potentiate the activity
2,3-OCH_3 increase activity for C and DAP

Biofilm inhibition

Compounds 12 and 22 were more active than reserpine (control)



23CR1



34DHCBr

OCH_3 are favourable to the activity
Bromo could contribute for the activity

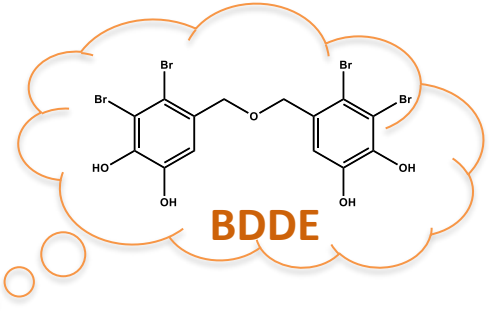
Jesus, A.; Durães, F.; Szemerédi, N.; Freitas-Silva, J.; da Costa, P.M.; Pinto, E.; Pinto, M.; Spengler, G.; Sousa, E.; Cidade, H. BDDE-Inspired Chalcone Derivatives to Fight Bacterial and Fungal Infections. Marine Drugs **2022**, 20, 315.

Conclusions

Synthetic Methods

Claisen-Schmidt condensation, catalytic hydrogenation and bromination

19 chalcone derivatives



Screened their bioactivity

- Antibacterial activity
- Antifungal activity

With exception of two chalcones

- Antibiotic synergistic effect
- EP inhibitors

Hits for EPs inhibitors/ antimicrobial adjuvants

Acknowledgments

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