Synthesis and Photodecomposition Studies of Photodegradable Antibiotics and Chitin Synthase Inhibitors

by

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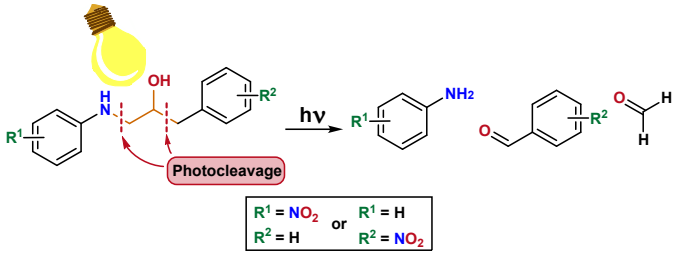
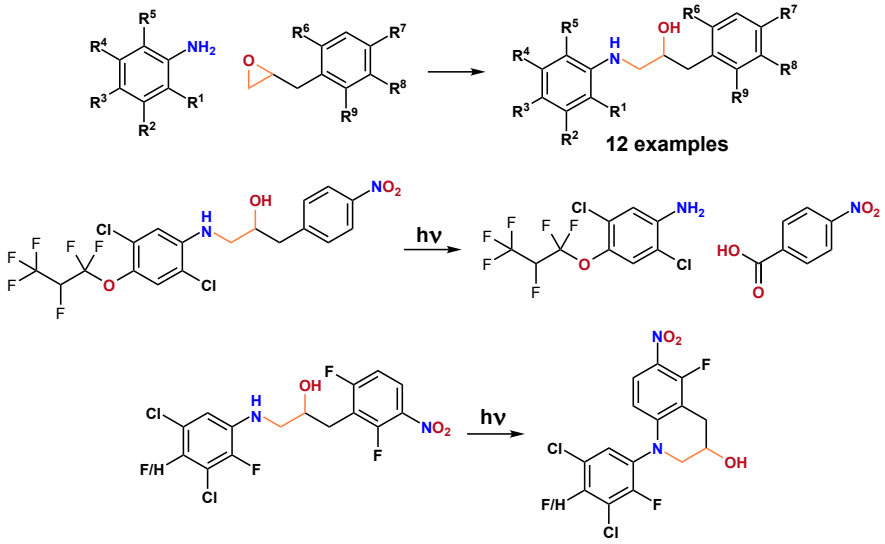
Lastly, I want to thank my family and friends for your constant support, and Jedna Turlag for the annual hiking trips, which provided me with some much needed fresh air.

Graphical abstract

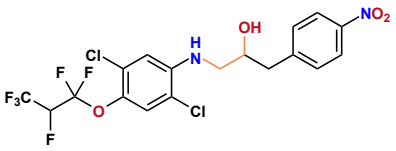
Part II - Development of a photodegradable scaffold



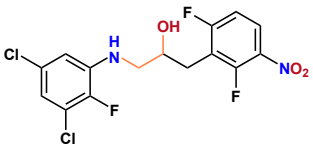
Part III - Chitin synthase inhibitors



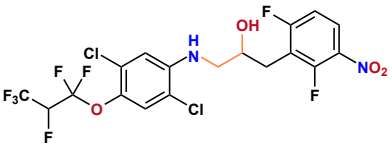
Part IV - Biological studies



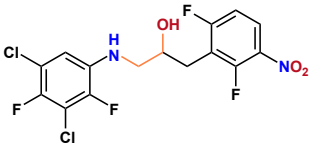
S. agalactiae **6.3 μM**
MRC5 50 μM, HepG2 75 μM



L. salmonis **0.1-0.001 ppt**
S. epidermidis, S. aureus, S. agalactiae **50 μM**
MRC5 50 μM, HepG2 75 μM

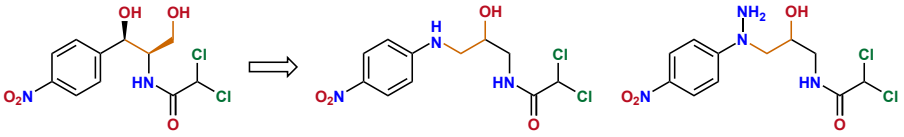


L. salmonis **1 ppt**
S. agalactiae **6.3 μM**
MRC5 25 μM, HepG2 50 μM



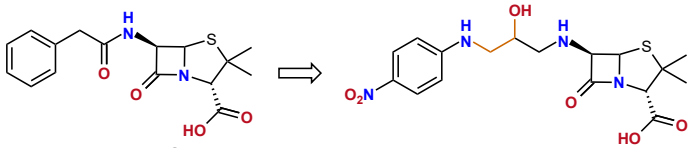
L. salmonis **0.1-0.001 ppt**
S. aureus **50 μM**
MRC5 50 μM, HepG2 50 μM

Part V - Chloramphenicol analogues



Chloramphenicol **Antimicrobially inactive**

Part VI - Towards a photodegradable penicillin



Penicillin G **Not obtained**

Abstract

The release of bioactive compounds to the biosphere, such as chitin synthase inhibitors that are causing negative effects in non-target organisms, or antibiotic agents that contribute to increasing antimicrobial resistance, has become a major problem in recent years. To try to circumvent this, the goal of this project has been to design active compounds that degrade when exposed to light.

Four photodegradable scaffolds based on the core structure 1-(arylamino)-3-arylpropan-2-ol with a nitro group in different positions on the two aromatic rings were designed, and the decomposition products were investigated. The scaffolds were functionalised to give 12 target compounds that resemble commonly used chitin synthase inhibitors, and some of them displayed promising anti-lice activity (0.001-0.1 ppt). The compounds were also tested for their antimicrobial activity (6.3-50 μM), which resulted in the discovery of four active compounds. Following decomposition, all antimicrobial activity was lost and no cytotoxicity was observed, and they represent lead compounds for further development. Synthetic attempts towards several analogues of chloramphenicol were dictated by side reactions and reactivity issues, and ended up in two analogues with no antimicrobial activity. Synthesis of an analogue of the penicillin class of antibiotics was attempted and despite great efforts, problems associated with a final deprotection step was not resolved.

Selected abbreviations and acronyms

6-APA	6-Aminopenicillanic acid
ACN	Acetonitrile
aq.	Aqueous
ATR	Attenuated total reflectance
BDP	Boron-dipyrromethene
BippyPhos	5-(Di-*tert*-butylphosphino)-1',3',5'-triphenyl-1'*H*-[1,4']bipyrazole
Boc	*tert*-Butyloxycarbonyl
Conc.	Concentrated
Cop.	Copepodid(s)
COSY	Correlated spectroscopy
CP	Ciprofloxacin
CSA	(1*S*)-(+)-10-Camphorsulfonic acid
DCC	Dicyclohexylcarbodiimide
DCM	Dichloromethane
DIC	Diisopropylcarbodiimide
DIPEA	Diisopropylethylamine
DMAP	4-Dimethylaminopyridine
DMF	*N*,*N*-Dimethylformamide
DMSO	Dimethyl sulfoxide
EDG	Electron-donating group
Equiv.	Equivalent(s)
EWG	Electron-withdrawing group

FDA	Food and drug administration
FT-IR	Fourier-transform infrared spectroscopy
HMBC	Heteronuclear multiple-bond correlation
hr	Hour(s)
HRMS	High-resolution mass spectrometry
HSQC	Heteronuclear single-quantum correlation
IC	Internal conversion
IR	Infrared spectroscopy
ISC	Intersystem crossing
LG	Leaving group
LPDE	Lithium perchlorate-diethyl ether
LRMS	Low-resolution mass spectrometry
*m*CPBA	*m*-Chloroperoxybenzoic acid
MDR	Multi-drug resistant
MIC	Minimum inhibitory concentration
MS	Mass spectrometry
ms	Molecular sieves
MW	Microwave
Naup.	Nauplius
NIS	*N*-Iodosuccinimide
NMM	*N*-Methylmorpholine
NMR	Nuclear magnetic resonance
OD_{600}	Optical density at 600 nm
PPG	Photolabile protecting group

ppt	Parts per thousand
p-TSA	*p*-Toluenesulfonic acid
Quant.	Quantitative
R_f	Retardation factor
rt.	Room temperature
sat.	Saturated
S_N2	Bimolecular nucleophilic substitution
S_NAr	Nucleophilic aromatic substitution
TBAB	Tetrabutylammonium bromide
TBAI	Tetrabutylammonium iodide
TB Alliance	Global Alliance for Tuberculosis Drug Development
TBDPS	*tert*-Butyldiphenylsilyl
TBS	*tert*-Butyldimethylsilyl
Temp.	Temperature
TFA	Trifluoroacetic acid
THF	Tetrahydrofuran
TLC	Thin-layer chromatography
TLC-MS	Thin-layer chromatography-mass spectrometry
TOX	Cytotoxicity
UV	Ultraviolet
UV-vis	Ultraviolet-visible
WHO	World Health Organization
XDR-TB	Extensively drug-resistant tuberculosis

Science communication

Publications

1. Eikemo, V., Sydnes, L. K., Sydnes, M. O. Photodegradable antimicrobial agents - synthesis, photodegradation, and biological evaluation. *RSC Adv.* **2021**, *11*, 32339.

2. Eikemo, V., Sydnes, L. K., Sydnes, M. O. Photodegradable Antimicrobial Agents - Synthesis and Mechanism of Degradation. *Prepared manuscript.*

3. Eikemo, V., Sydnes, L. K., Sydnes, M. O. Photolytic decomposition, *SE-Patent application 2150492-3.* Filing date 20 April 2021.

Presentations

1. Eikemo, V. Photodegradable antimicrobial agents. BioCat Kick-off Event, 2021. **Oral presentation**.

Table of Contents

Part I

Introduction

In recent years, many pharmaceutical compounds have been detected in drinking water, wastewater, ground water, coastal waters, and marine organisms around the world.[1–4] Many antibiotics are only partially metabolised in the human body,[5–7] and conventional wastewater-treatment processes are not sufficient to completely remove these substances prior to release, leading to environmental exposure of antibacterial agents.[8–14] Combined with agricultural runoff, industrial discharge from production facilities, and the aquaculture industry, this continuous release of bioactive organic compounds to the environment results in negative effects in non-target organisms. This is a major issue especially concerning antibiotics, since it contributes to the buildup of antimicrobial resistance (Figure **1**).[15–21]

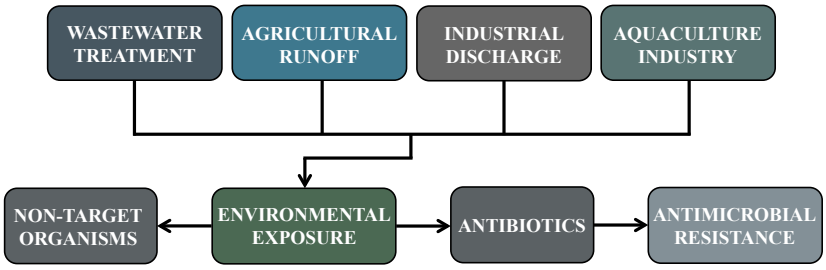


Figure 1. Sources of environmental exposure to bioactive organic compounds, such as antibiotics, which increases antimicrobial resistance.

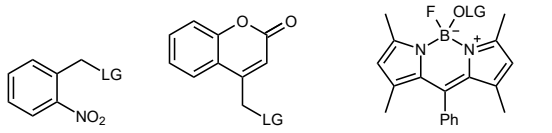
I.1 Photocontrol of drugs

To address these issues, novel methods are being developed that uses light as a tool to enable precise control of the biological activity of pharmaceutical compounds. Achieving spatial and temporal control, i.e. the possibility of deciding where and when activity is wanted, means that a drug can be switched '*on*' or '*off*' at will. Triggering photoresponsive functional groups in a molecule, such as removing a photolabile protecting group (PPG) that inhibits biological activity, reversible activation and deactivation by means of a photoswitch, or initiating an irreversible destructive mechanism, allows for different ways of optically controlling a drug's properties.

I.1.1 Photoprotecting groups

While the removal of traditional protecting groups require chemical reagents that could lead to unwanted side reactions, PPGs only use light in the deprotection process.[22] The mild conditions used to photochemically remove protecting groups has made them attractive targets in the synthesis of natural products, and they are, for example, involved in the total syntheses of *ent*-fumiquinazoline G and (-)-diazonamide A.[23,24] There are several different types of PPGs, such as the 2-nitrobenzyl, coumarin-4-yl methyl, and boron-dipyrromethene types (Figure **1**).[22,25–29] The wide availability and structural variation of PPGs means that different wavelenghts are required for deprotection to occur. This opens the possibility of im-

plementing orthogonal protection and deprotection strategies with complete control of selectivity.[25]

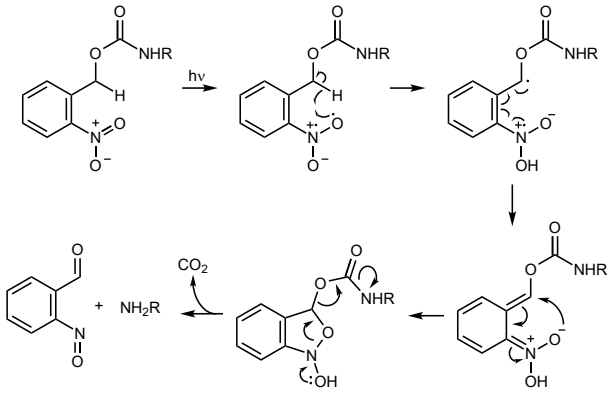


2-Nitrobenzyl Coumarin-4-yl methyl Boron-dipyrromethene (BDP)

Scheme 1. Some commonly used PPGs.[22,25–29] LG = Leaving group.

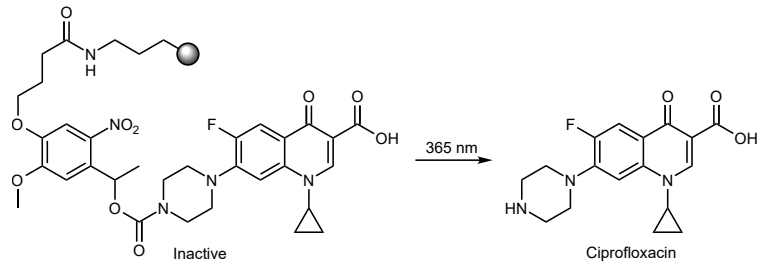
The idea of using photoprotecting groups to activate pharmaceutical compounds is an important strategy in photopharmacology, which is an emerging approach in medicine with the goal of preventing side effects.[30,31] A PPG can be installed on important functional groups that are critical for biological activity, such as the carboxylic acid in β-lactams and quinolone antibiotics, thereby masking the activity.[32,33] One method to incorporate a PPG into an active drug is through a carbamate linker, which is useful in pharmaceutical compounds containing an amine functionality. The mechanistic pathway of photorelease of such a compound is illustrated using the 2-nitrobenzyl group linked to an amine as a carbamate (Figure **2**). The reaction takes place with initial excitation of the nitro group with irradiation with UV light. Proton abstraction by the hydroxyl radical followed by radical combination leads to ring closure to a five-membered ring. Subsequent ring opening releases CO_2, *o*-nitrosobenzaldehyde, and the free amine. An identical mechanism will occur for carboxylic acid substrates linked as an ester instead

of a carbamate, with release of *o*-nitrosobenzaldehyde and the free acid without decarboxylation.[22]



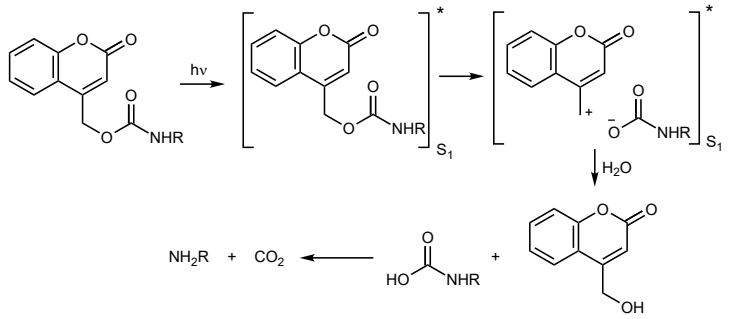
Scheme 2. Mechanism of photorelease of 2-nitrobenzyl PPG.[22]

This strategy of structure design is used for the photorelease of ciprofloxacin using the 2-nitrobenzyl moiety as a photolabile leaving group. Forsythe and co-workers reported liberation of the antibacterial compound upon irradiation at 365 nm at low intensity (Scheme **3**).[26] Ciprofloxacin linked to the hydrogel makes this a potential material to be used in wound dressings, which would slowly release the antibiotic through exposure to light, thereby improving the healing process.



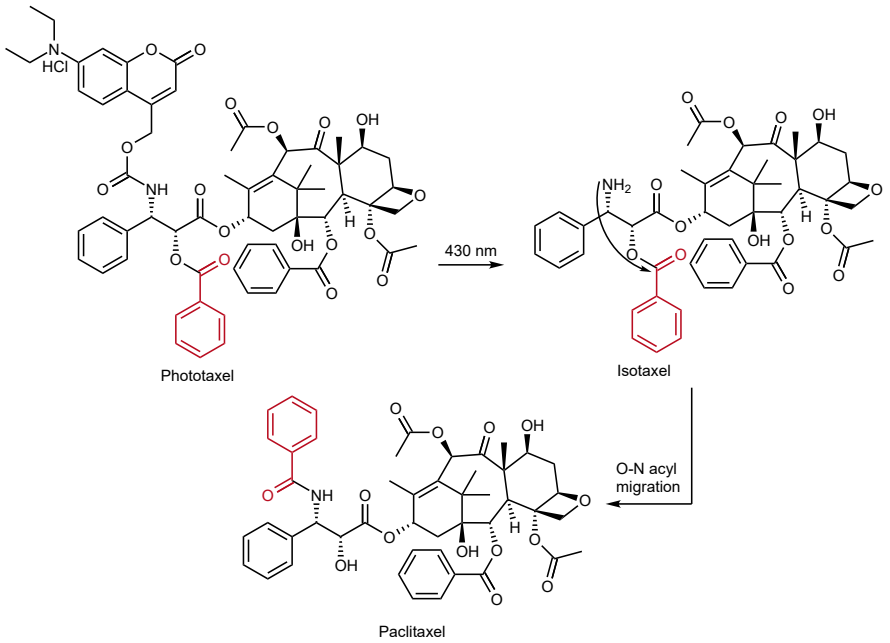
Scheme 3. Photodeprotection of ciprofloxacin as reported by Forsythe and co-workers.[26] The sphere represents a linker to a hydrogel network structure.

The coumarin-4-yl methyl PPG can be installed in a similar fashion through a carbamate with amine-containing substrates. Irradiation with light forms the first excited singlet state (S_1), which in turn, can either get deactivated by loss of excitation energy through fluorescence or undergo heterolytic cleavage to form an ion pair. Reaction with water forms coumarin-4-yl methanol and a carbamic acid which decarboxylates to give the free amine (Scheme **4**). Carboxylic acid substrates liked as an ester displays an identical photorelease mechanism with the exception of the final decarboxylation step.[34,35]



Scheme 4. Photorelease of coumarin-4-yl methyl-protected amines.[34,35]

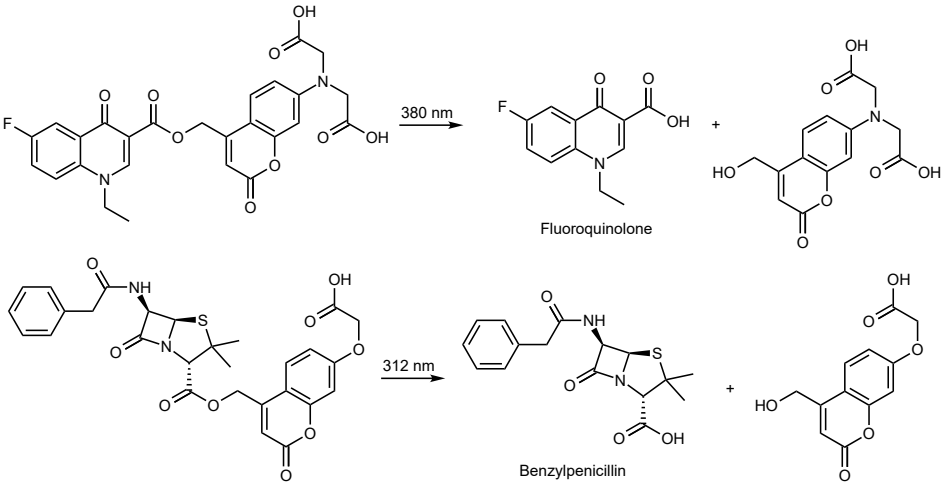
This strategy is used by Kiso and co-workers for photoactivating the inactive prodrug phototaxel into paclitaxel, a chemotherapy medication. Irradiation at 430 nm releases the coumarin-4-yl methyl group, CO_2, and isotaxel through the mechanism described above, and subsequent O-N acyl migration yields the active drug (Scheme **5**).[27] The acyl migration step is critical since paclitaxel does not contain any amine functionality, which is required for the carbamate linker.



Scheme 5. Photoactivation of phototaxel as reported by Kiso and co-workers.[27]

Similar coumarin protecting groups have been used by Feringa and co-workers for the photorelease of a fluoroquinolone antibiotic and benzylpenicillin (Scheme **6**).[28] In these examples, the PPG is linked with an ester instead of a carbamate, and upon irradiation, the active

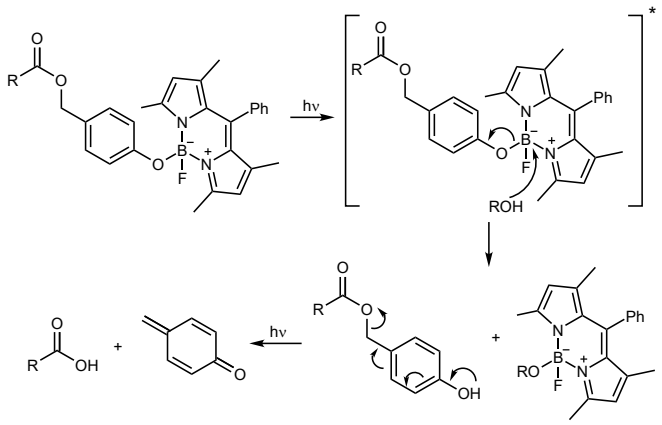
antibiotic compuonds are released as the free carboxylic acids along with the respective coumarin-4-yl methanol, following the same mechanism as described earlier (Scheme **4**). Additionally, these examples illustrate the effect the substituent on the coumarin phenyl ring has on the wavelength required for cleavage to occur. By exchanging the aminediacetic acid with the oxyacetic acid group, a difference of nearly 70 nm is seen. This opens the possibility for fine tuning the absorption maxima by choosing the right substituents.



Scheme 6. Photoactivation of phototaxel as reported by Kiso and co-workers.[27]

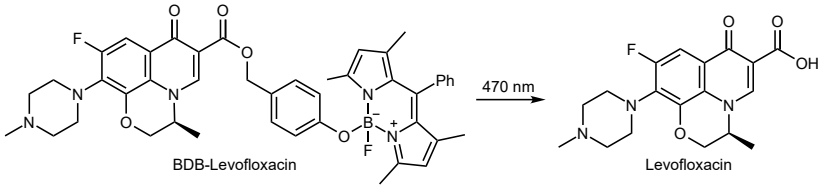
The BDP PPG is installed somewhat differently, with a phenoxy linker between the photosensitive moiety and the substrate. Carboxylic acids are protected this way through a simple esterification with the PPG containing the phenoxy linker. The B-O bond in aryloxy-BDP derivatives cleave efficiently upon irradiation with visible light, forming the carboxylic acid substrate with the phenol

linker still intact. Further irradiation leads to self-immolation of the phenolate yielding the free acid and 4-methylenecyclohexa-2,5-dien-1-one (Scheme **7**).[29,36]



Scheme 7. Photodeprotection mechanism of the BDP group.[29,36] R = H, alkyl.

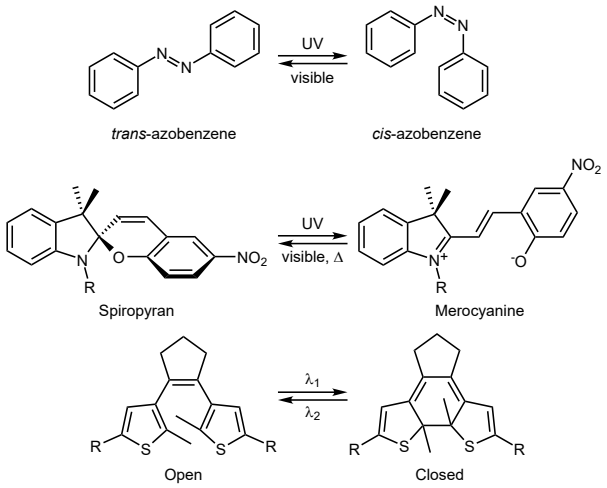
This strategy was used by Chakrapani and co-workers for the photorelease of levofloxacin. The B-O bond is photolytically cleaved by irradiation at 470 nm, which forms a phenolate that self-immolates into the desired antibiotic (Scheme **8**), following the mechanism described above.[29]



Scheme 8. Photorelease of levofloxacin, as reported by Chakrapani and co-workers.[29]

I.1.2 Photoswitching drugs

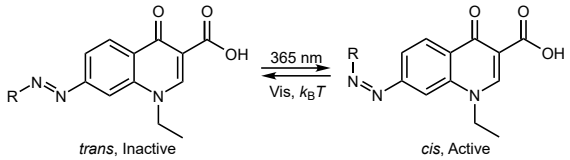
In order to achieve even more control of a drug's potential, a photoswitchable moiety can be incorporated into a biologically active substance. This would allow for the drug to be reversably switched between two states of different geometries and electronic properties. Examples of such photoswitches are azobenzenes, spiropyrans, and diarylethenes (Scheme **9**).[30,37]



Scheme 9. Photoisomerisation of azobenzenes, spiropyrans, and diarylethenes.[30,37]

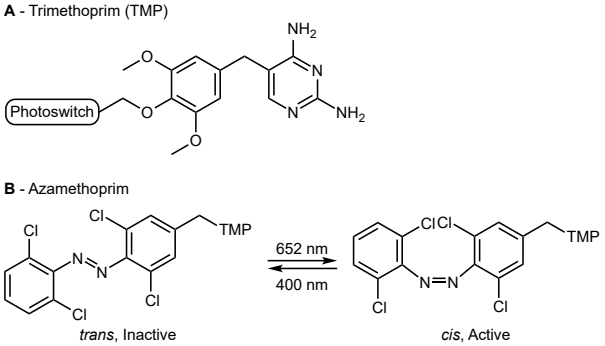
Feringa and co-workers developed a photoswitchable antibacterial compound based on the quinolone antibiotic class, with the usual piperazine exchanged with an aryldiazo moiety and the fluorine substituted for a proton. Upon irradiation with 365 nm, conversion from the less biologically active *trans* to the more active *cis*-isomer

occurs. The reversible reaction occurs either through exposure to visible light or thermally at room temperature (Scheme **10**).[38]



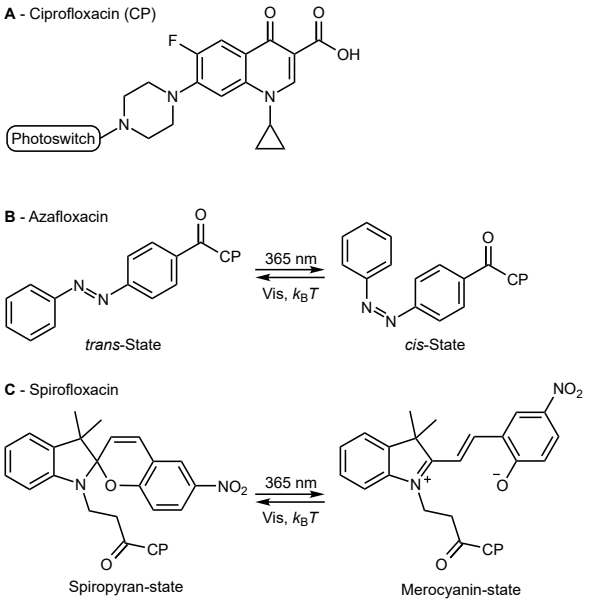
Scheme 10. Photoswitching of a quinolone antibiotic by Feringa and co-workers.[38] R = 3-Methyl-4-methoxyphenyl.

Another example of an antibacterial compound that takes advantage of the *cis-trans* isomerisation of azobenzenes, is the photoswitchable analogue of trimethoprim (Figure **11**, **A**). Feringa and co-workers functionalised the widely used, broad-spectrum antibiotic with an azobenzene moiety that switches from the antimicrobially inactive *trans*-state to the active *cis*-state by irradiation at 652 nm. The *trans*-state can be regenerated by irradiating at 400 nm (Figure **11**, **B**), enabling this compound to be switched '*on*' or '*off*' using different light sources.[39]



Scheme 11. Photoswitching of a trimethoprim antibiotic by Feringa and co-workers.[39]

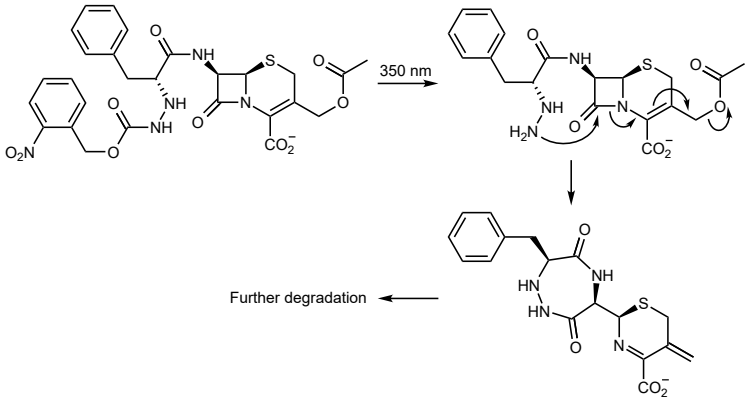
Analogues of ciprofloxacin with photoswitches attached at the secondary nitrogen in the piperazine ring has also been developed. By using acid chloride derivatives of azobenzene and spiropyran, the photoswitch is easily incorporated (Scheme **12**, **A**), and they readily alternate between the two different states (Scheme **12**, **B** and **C**). Azafloxacin in its thermally relaxed *trans*-state shows a 50 fold increase in activity against *M. luteus* compared to ciprofloxacin, whereas the *cis*-state shows a 25 fold increase. Spirofloxacin was found to be 5 times more active than the original antibiotic for both states. Against *E. coli*, azafloxacin displayed a 40 fold decrease in activity compared to ciprofloxacin but no change for its *cis* and *trans*-state. For spirofloxacin however, a doubling in activity was observed from the thermally stable spiropyran to the irradiated merocyanin-state. Compared to the unmodified ciprofloxacin they were 100 and 50 times less active, respectively.[37]



Scheme 12. Ciprofloxacin photoswitch with azobenzene and spiropyran reported by Feringa and co-workers.[37]

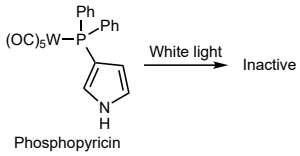
I.1.3 Photodecomposition

Another method of deactivating drugs, and perhaps the most efficient one, is to completely destroy the core structure, rendering it inactive. By incorporating a destructive mechanism into the structure, Mobashery and co-workers accomplished complete decomposition of a cephalosporanic-acid antibiotic triggered by light (Scheme **13**). They applied the familiar 2-nitrobenzyl PPG, that when photolytically removed, releases a hydrazine which rapidly attacks and opens the β-lactam ring, leading to inactivation of the antibiotic.[40]



Scheme 13. Photodecomposition of a cephalosporanic-acid antibiotic.[40]

Photodegradation also occured for an inorganic batericidal compound termed phosphopyricin. Sterenberg and co-workers described a phosphine-tungsten complex, which displayed activity against *S. aureus*, that completely degraded by white light over the course of several hours (Scheme **14**). However, the photodegradation products were not investigated.[41]

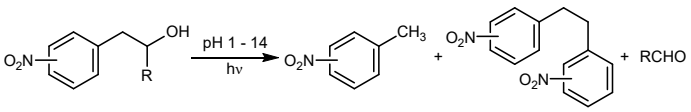


Scheme 14. Photodegradation of an inorganic antibacterial agent.[41]

I.1.4 Photo-retro-aldol type reaction

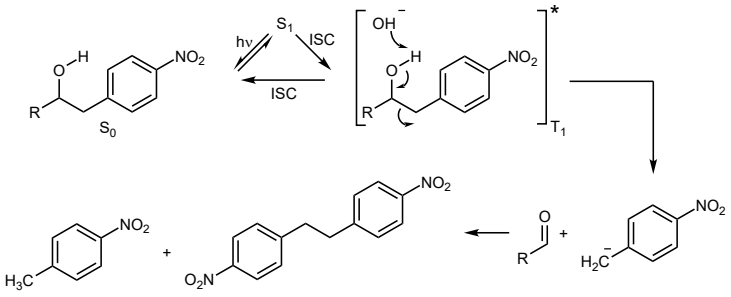
Another interesting and less recognised example of photodegradation, is the photo-retro-aldol type mechanism proceeding through

heterolytic cleavage of a benzylic C-C bond. Wan and Muralidharan observed this transformation with *p*- and *m*-nitro-substituted phenethyl alcohols with varying substituents (R) in 1-position at different pH values (Scheme **15**).[42,43]



Scheme 15. Photolytic cleavage of nitrophenethyl alcohols.[42,43] R = H, Ph.

With 1-phenyl substituted phenethyl alcohols, the reactions were more or less pH independent as conversions of 30-60% were achieved throughout the pH range. The mechanism suggests that the reactive triplet state undergoes a retro-aldol reaction through abstraction of a proton, by either water or hydroxide, forming an aldehyde and *p*-nitrobenzyl carbanion, which in turn gives *p*-nitrotoluene and 4,4'-dinitrobibenzyl (Scheme **16**).[42]



Scheme 16. Mechanism for the photo-retro-aldol reaction as outlined by Wan and Muralidharan.[42]

I.1.5 The excited state

For any of the aforementioned photochemical processes to occur, the compounds need to be able to absorb electromagnetic radiation, resulting in excitation of an electron to a higher energy level. The energy of the excited state can be sufficiently high enough to rival the bond dissociation energy of a carbon-carbon σ-bond, which makes it reasonable that certain chemical transformations are initiated by light.[44] Excitation occurs from a bonding or non-bonding molecular orbital (σ, π, n) to an antibonding orbital (σ*, π*), and the transitions are illustrated as σ-σ*, n-σ*, π-π* or n-π*.[44] There are two possible electron arrangements following excitation; the singlet state S_1 with paired electron spins, or the triplet state T_1 with parallel electron spins. These are formed according to the Jablonski diagram (Figure **2**), where the ground and higher vibrational levels of each electronic state are indicated by black and blue lines, respectively.

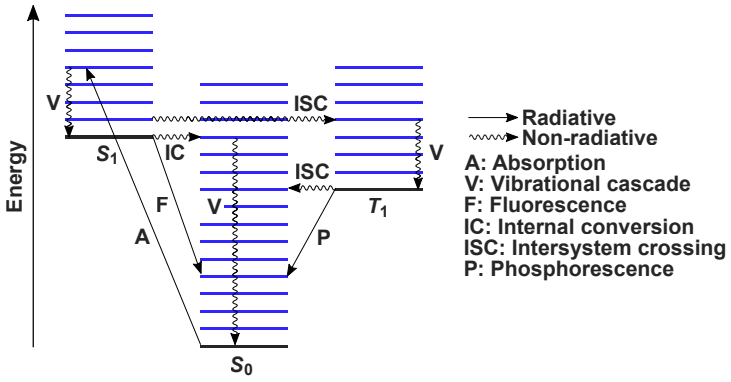
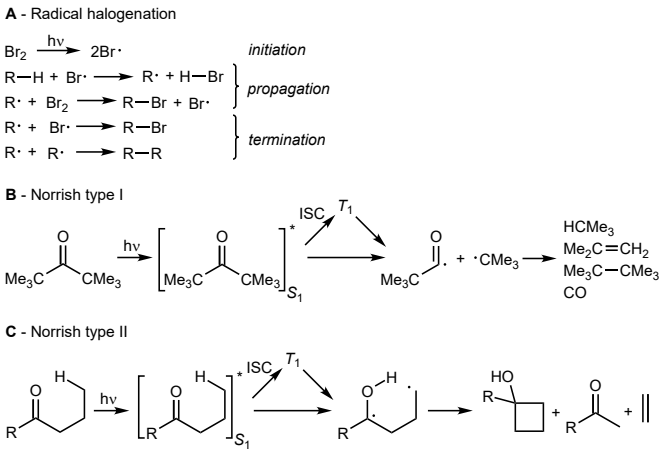


Figure 2. Jablonski diagram illustrating excitation and deactivation routes.[44]

Following this illustration, a molecule in its ground state S_0 is absorbing electromagnetic radiation (A), forming the first excited singlet state S_1, which is high in vibrational energy. A process termed vibrational cascade (V) occurs, dissipating energy as heat. From this point there are three possibilities; loss of excitation energy by emission in the form of fluorescence (F), internal conversion (IC) S_1-S_0 followed by vibrational cascade (V) to the ground state, or intersystem crossing (ISC) S_1-T_1 with subsequent vibrational cascade (V) and emission of radiation as phosphorescence (P).[44] Once an excited state is populated sufficiently, a wide range of interesting chemical reactions can take place, such as alkene/azobenzene *E*-*Z* isomerisation, cyclisation of conjugated dienes, oxidation and reduction reactions, and photo-retro-aldol type reactions, that would not take place in the abscence of light.[42–45] Often, the excited species transform into free radicals, which in turn opens up the possibility for a whole range of transformations, such as the radical halogenation of alkanes (Scheme **17**, **A**) or the Norrish type I (**B**) and II reactions (**C**).[44,46,47]



Scheme 17. A few examples of free-radical reactions initiated by light.[44,46,47]

I.2 Persistant drugs in the aquaculture industry

The aquaculture industry in Norway is a huge economic resource with export values for the nine first months of 2021 exceeding 84 billion NOK. That is a 10% increase compared to the same period in 2020, and this is estimated to continue growing.[48] In fact, with 1.4 million tonnes of Atlantic salmon farmed annually as of 2019, Norway is the largest producer in the world.[49] That means that this industry represents the largest export in Norway after oil and gas and it is important to tackle any problems that are associated with it. One such problem is salmon lice (*Lepeophtheirus salmonis*), feeding on skin, mucus, and blood from the host, which may also induce epizootics in wild fish.[50] Since the salmon farms are using open-net pens, the lice infestations can move to adjacent farms and may also infect the local populations of fish, such as sea trout (*Salmo trutta*)

and wild Atlantic salmon (*Salmo salar*).[51–53] To solve the lice problem, fish farms are using large amounts of parasiticides, such as the benzoyl urea compounds diflubenzuron and teflubenzuron (Figure **3**). These compounds act as chitin synthase inhibitors, preventing the lice from forming the chitin-rich exoskeleton during its frequent ecdyses (change of exoskeleton) until adulthood, ultimately leading to death.[54] There are several other treatments for lice infestations, such as emamectin benzoate, hydrogen peroxide, azamethiphos, and pyrethroids, but reports of resistance towards all these compounds are emerging.[55] However, resistance against the benzoyl ureas are not reported for *L. salmonis*, but there are observations of resistance in other arthropods.[55] In recent years, another benzoyl urea, lufenuron, has been launched as a new louse treatment (Figure **3**).[56]

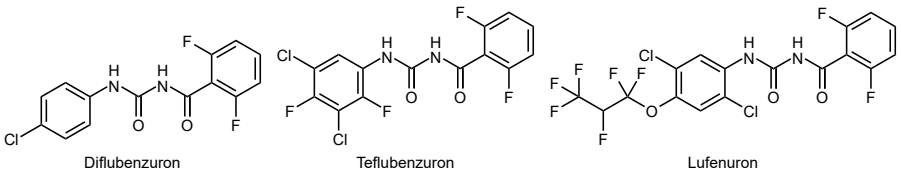


Figure 3. Two commonly used anti-lice drugs, diflubenzuron and teflubenzuron, and the more recent addition, lufenuron.[54,56]

Since the flubenzurons and lufenuron disrupt the chitin synthesis, they are highly toxic to non-target crustaceans.[17] A laboratory study by Samuelsen showed that teflubenzuron and diflubenzuron showed no significant decomposition when associated to organic particles in marine sediment after 24 weeks.[57] Another study revealed a stipulated half-life of 170 days for teflubenzuron in sediments collected under a fish farm.[21] These compounds have been detected as far

away as 1400 m from the farm and have proven to exhibit negative impacts on king crab, shrimp, squat lobster, and European and Norway lobster in laboratory studies.[18–21,58] If moulting was imminent, the detected levels of teflubenzuron was sufficient to induce mortality. The maximum-residue level value in saithe and the crustaceans mentioned above exceeded that of Atlantic salmon, meaning that food safety has to be considered.[21]

I.3 Antibiotic resistance

Overuse of antibiotic agents in human medicine and agriculture has led to the emergence of resistant bacteria strains, and many illnesses that were once easily manageble, such as pneumonia and post-operative infections, are gradually becoming untreatable.[59,60] Tuberculosis, today's number one infectious disease with 10 million cases and 1.4 million deaths in 2019, is becoming multi-drug resistant (MDR), requiring prolonged treatment with highly toxic and expensive second-line antibiotics. Resistance to these second-line drugs are also occuring, giving rise to what the WHO defines as extensively drug-resistant tuberculosis (XDR-TB).[60,61] MDR bacteria are already a widespread problem across the globe and several public health organisations have described the situation as a crisis or nightmare scenario that could have catastrophic consequences.[62–64] It is estimated that 700.000 people die every year from infections caused

by drug-resistant bacteria, and despite this, pharmaceutical companies are reluctant to develop new antibiotics.[65] The risk of resistance buildup means that new antibiotics are used only when necessary, which completely ruins the profit. Of the 15 new FDA-approved antibiotics since 2010, five of these are now scarcely available since the companies that produced them were either sold off or filed for bankruptcy.[65] This negative development is contributing to driving the *"evolution of resistance"* forward, rendering more and more antibiotics ineffective.[62] As seen in Figure **4**, there is a discovery void of new classes of antibiotics during the last few decades. From the early 1900s up to the late 1980s, new classes appeared frequently, but this development has declined dramatically.[66–70]

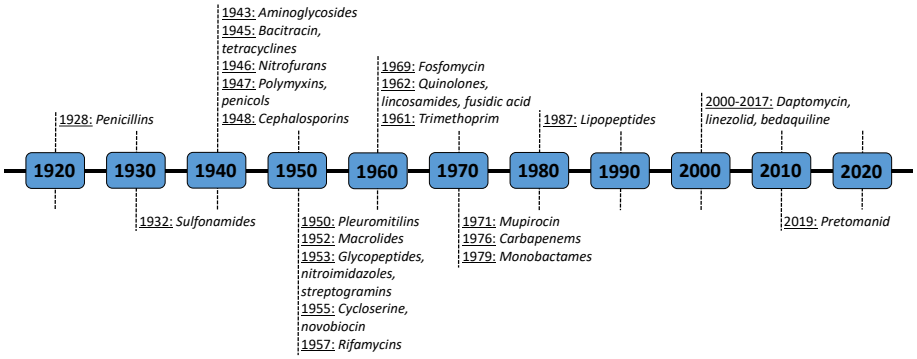


Figure 4. Classes of antibiotics discovered through the 20th century up to 2017.[66–70]

In the period 1998-2017, three new antibacterial agents with novel mechanisms of action have been approved by the FDA, however, all of them show activity only against Gram-positive bacteria.[68,69] In 2019, the Global Alliance for TB Drug Development (TB Alliance)

gained FDA approval for Pretomanid, a new drug for the treatment of MDR and XDR-TB.[70] Following this drug, no new classes of antibiotics against Gram-negative bacteria have been approved.[60] Additionally, studies by Fischback and Walsh in 2009 uncovered that if microorganisms are resistant to a particular drug, they rapidly develop resistance to the entire class, making development of new classes of antibiotics even more urgent.[71]

I.4 Aim of the project

The first aim of this project has been to design and synthesise a light-sensitive compound based that can undergo a photo-retro-aldol decomposition. The fragmentation products resulting from photocleavage of the proposed 1-(arylamino)-3-arylpropan-2-ol scaffold will be investigated (Figure **5**).

Secondly, the scaffold will be functionalised into compounds expected to display activity against *L. salmonis*, a parasite that is pestering Atlantic salmon farming. This will be done by mimicking the chemical structure of the benzoyl urea drugs diflubenzuron, teflubenzuron, and lufenuron, and evaluating their potential as chitin synthase inhibitors (Figure **5**).

The third aim of this thesis has been to apply the light-sensitive scaffold to make photodegradable antimicrobial compounds, based on the structures of the already existing antibiotics chloramphenicol and the penicillins. Antibacterial activity will be evaluated against a number of Gram positive and Gram negative bacteria.

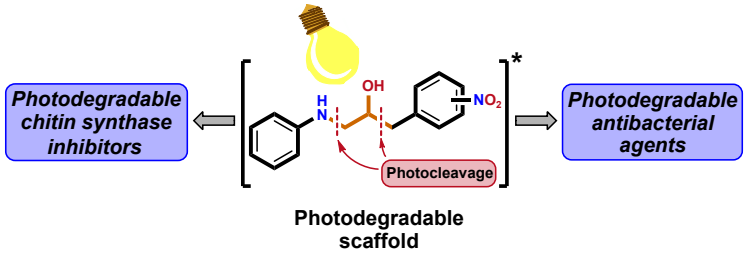


Figure 5. Aim of the study.

Part II

Development of a photodegradable scaffold

Inspired by the photo-retro-aldol type chemistry described by Wan and Muralidharan,[42,43] and the environmental impacts of the benzoyl urea parasiticides diflubenzuron and teflubenzuron (described in Part I), a series of model compounds were designed to act as photodegradable scaffolds (Figure **6**). The suggested 1-(arylamino)-3-arylpropan-2-ol derivatives with the two aromatic groups linked by a chain of equal length and a nitro group positioned in *para* or *meta*-position on either ring, are reasonable candidates for this study. After synthesising the compounds, the photochemical properties and photodegradation patterns were investigated.

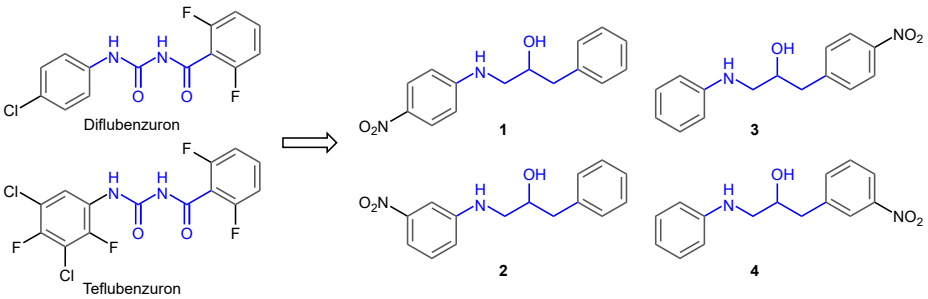
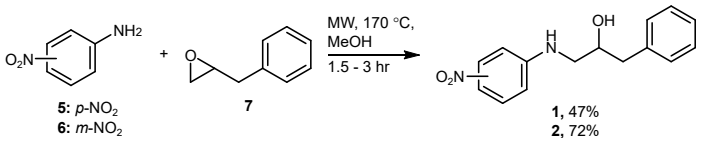


Figure 6. Analogues of teflubenzuron and diflubenzuron for a proof-of-concept study.

II.1 Synthesis of model compounds

Amino alcohols **1** and **2** were prepared according to a literature procedure by Lindsay and co-workers,[72] which yielded the two target molecules in 47 and 72% yields, respectively. The difference in isolated yields is explained by the inferior nucleophilicity of *p*-nitroaniline compared to the *m*-isomer, as indicated by the pK_a-values of their conjugate acids of 1.0 and 2.5, respectively.[73]

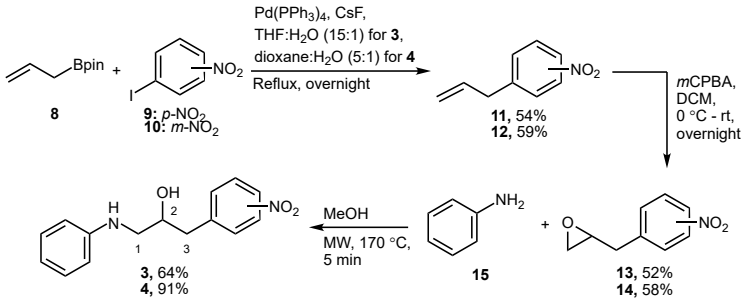


Scheme 18. Synthesis of amino alcohols **1** and **2** through a microwave-assisted epoxide ring opening.

The yield of amino alcohol **1** was increased to 64% when the reaction was performed in 5 M lithium perchlorate-diethyl ether (LPDE) solution, following a procedure reported by Heydari and co-workers.[74] The rate enhancement is due to a high concentration of Li^+, which acts as a weak Lewis acid, activating the epoxide.[75]

In order to synthesise the remaining two analogues with the nitro group positioned on the phenyl moiety at C3 (**3** and **4**, Scheme **19**), a few additional steps were necessary. A similar epoxide ring opening as described for **1** and **2** was envisioned, but as the nitrated epoxides were not commercially available, they had to be synthesised from the corresponding iodonitrobenzenes **9** and **10** (Scheme **19**). A

Suzuki-Miyaura cross-coupling reaction with allyl boronate **8**, using a method described by Kotha and co-workers,[76] yielded the allylated product **11**, but disappointingly compound **12** was not formed under these conditions. Electron-withdrawing groups (EWG) activates the aryl halide towards oxidative addition,[77] and this effect is less prominent in *m*-position, which explains the poorer reactivity of **10**. However, by switching solvent from THF to 1,4-dioxane and increasing the temperature to 100 °C, the allylated coupling product **12** was succesfully formed in good yield (59%). Subsequent treatment with *m*CPBA gave epoxides **13** and **14**, and a microwave-assisted epoxide ring-opening reaction[72] with aniline (**15**) gave the desired amino alcohols **3** and **4** in 18% and 31% overall yields, respectively (Scheme **19**).



Scheme 19. Synthesis of amino alcohols **3** and **4** through Suzuki-Miyaura cross coupling followed by epoxidation and subsequent microwave-assisted ring opening.

II.2 Decomposition studies and elucidation of the decomposition mechanism

With the four model compounds in hand, their λ_{max} and molar attenuation coefficient ϵ was determined by UV-vis spectroscopy (Figure **7**). Compounds **1** and **2**, with the nitro group attached to the aniline, have similar absorption spectra with a slight bathochromic shift for the *m*-isomer (384 to 400 nm). For amino alcohols **3** and **4**, a large hypsochromic shift is seen towards 248 nm. Due to the large difference in λ_{max}, these compounds are expected to behave differently in the degradation study.

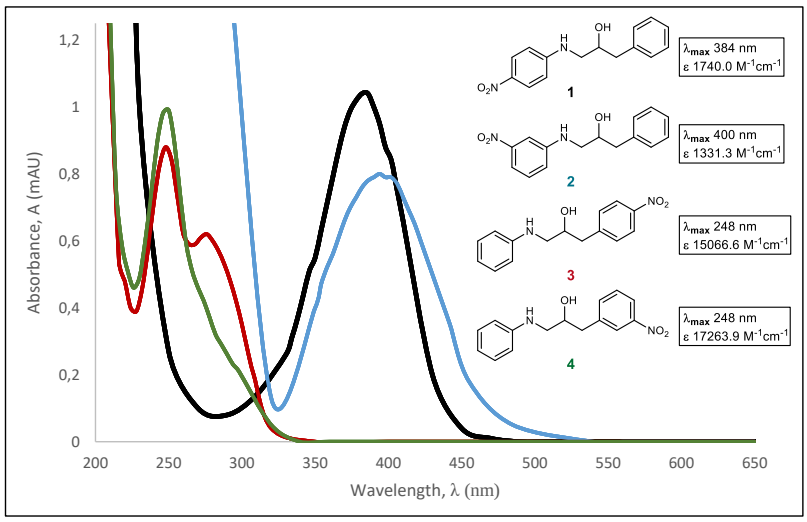


Figure 7. Overlay of the UV-vis spectra of the four model compounds **1-4**. Concentrations were 0.6 M for **1** and **2** and 0.06 M for **3** and **4**. ϵ was determined from the slope of standard curves with four data points and R^2-values > 0.999.

Following the procedure by Wan and Muralidharan,[42] amino alcohol **1** was photolysed in acetonitrile (ACN) and water (7/3, V/V) at

various pH using a 125 W medium-pressure mercury-vapour lamp with strong emissions at 315, 365, 436, 546, and 578 nm. Seven experiments were performed from pH 13 to pH 1 with a concentration of ∼0.68 mM and 2 hr irradiation time. No special care was taken to exclude oxygen from the reaction mixture. Following liquid-liquid extraction with DCM, all reaction mixtures were analysed with ^{1}H-NMR spectroscopy (Figure **8**). Reactions performed at pH lower than 7 did not show sufficient decomposition and are therefore not shown in Figure **8**. ^{1}H-NMR analysis of control experiments performed in the dark at pH 11 revealed that no reaction is taking place in the abscence of light (Figure **9**). Compounds **2**-**4** show identical stability.

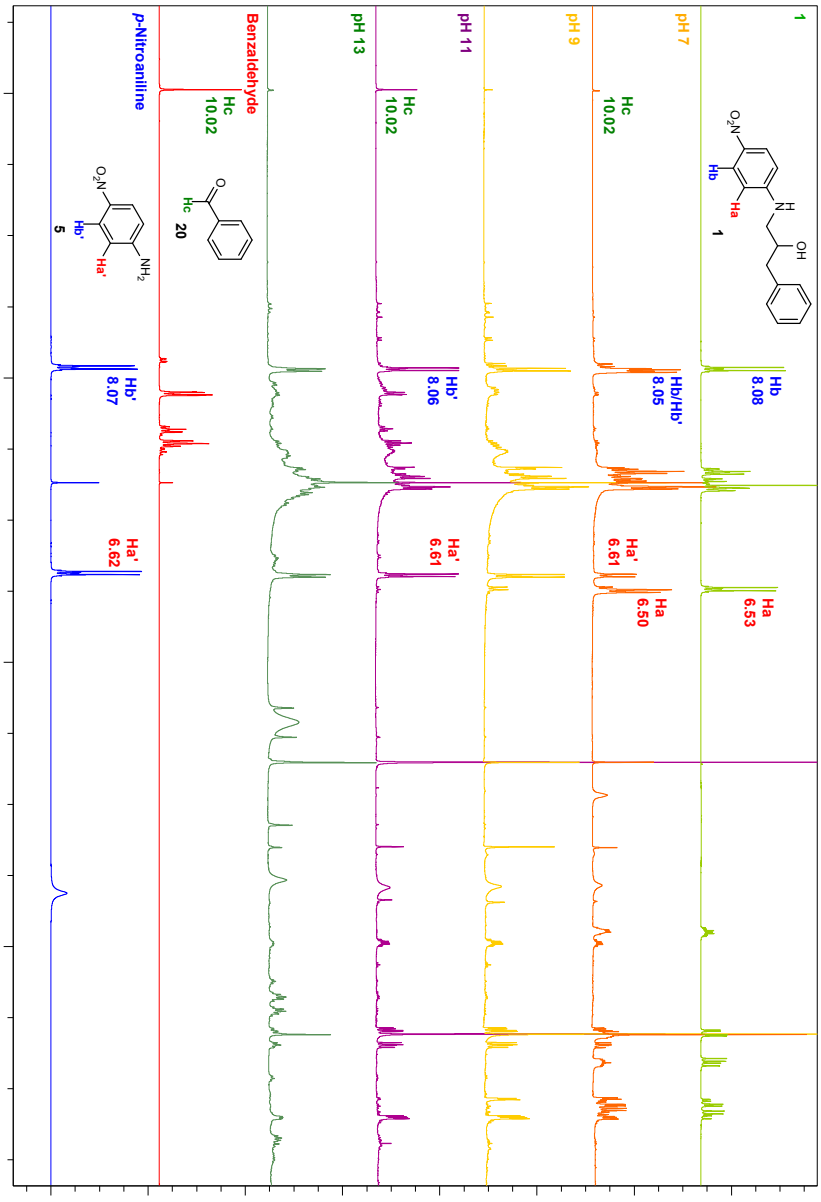


Figure 8. ^{1}H-NMR spectra displaying photolysis of amino alcohol **1** at varying pH values. Irratiation time: 2 hr, Lamp: 125 W medium-pressure mercury.

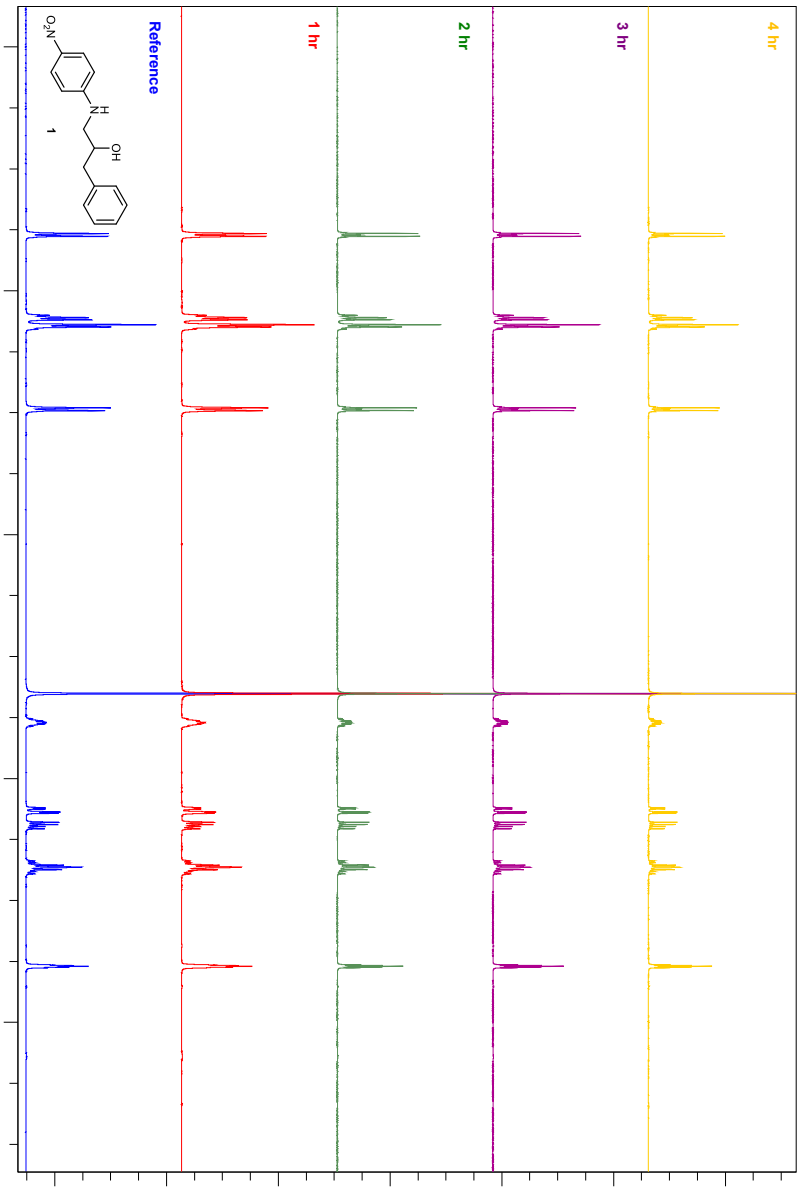
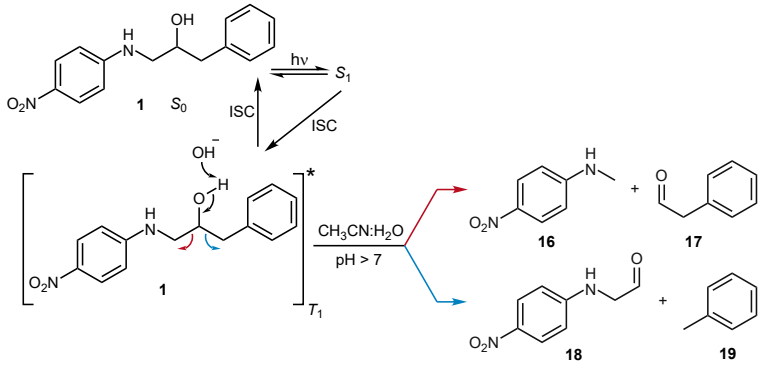


Figure 9. ^{1}H-NMR spectra displaying complete stability of amino alcohol **1** at pH 11 after 4 hr with abscence of light. Lamp: 125 W medium-pressure mercury.

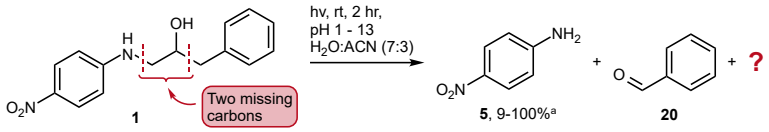
Initially it was expected that the decomposition would take place through a mechanism similar to the nitrophenethyl alcohols reported by Wan and Muralidharan (Scheme **16**).[42] Expected products would therefore be *N*-methyl-*p*-nitroaniline (**16**) and phenylacetaldehyde (**17**) following the **red** mechanistic pathway, or 2-(*p*-nitroaniline)acetaldehyde (**18**) and toluene (**19**), illustrated by the **blue** mechanism (Scheme **20**). Intermolecular reactions are highly unlikely to occur due to the low reaction concentration.



Scheme 20. Expected behaviour of amino alcohol **1** when subjected to the conditions described by Wan and Muralidharan.

However, analysis of ^{1}H-NMR spectra of the reaction mixtures revealed none of these products. For photodecomposition reactions performed with compound **1** at pH $\geq$ 7, an aldehydic signal is observed at δ 10.02 ppm, corresponding well with a reference spectrum of benzaldehyde (**20**) ($\Delta\delta$ = 0.001 ppm) (Figure **8**). Additionally, a doublet at δ 6.61 ppm is seen, correlating to the *o*-proton in *p*-nitroaniline ($\Delta\delta$ = 0.019 ppm). The remaining signals of these two compounds are present and illustrated in an overlay of the spec-

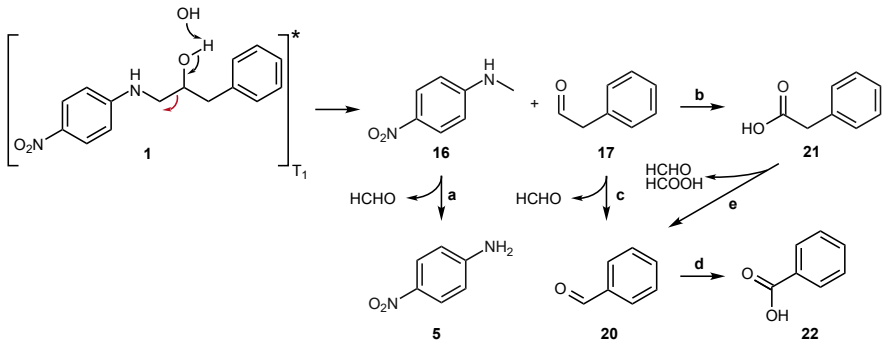
tra (Figure **8**). The decomposition rate is clearly pH dependent, reaching a peak somewhere around pH 11, and gradually decreasing with lower pH values. As two of the products are undoubtedly *p*-nitroaniline (**5**) and benzaldehyde (**20**), this means that there are two carbons that are unaccounted for in the decomposition (Scheme **21**).



Scheme 21. Photodecomposition of compound **1**. [a]Estimated from normalised integral values for *o*-protons on the aniline for each experiment at pH 1, 3, 5, 7, 9, 11, and 13 (Table **2**).

Additionally, it is worth noting that the ratio between benzaldehyde and *p*-nitroaniline is much lower for the experiment performed at pH 13 than for the one at pH 11 (6:94 and 26:74, respectively). This suggests that benzaldehyde reacts further, and that a reaction according to the **red** pathway occurs (Scheme **20**), initially forming *N*-methyl-*p*-nitroaniline (**16**) and phenylacetaldehyde (**17**). Photochemical *N*-demethylation is known to occur for anilines,[78,79] yielding *p*-nitroaniline (**5**) and formaldehyde (Scheme **22**, **a**). Phenylacetaldehyde can either undergo oxidation in the presence of oxygen to form phenylacetic acid (**21**) (Scheme **22**, **b**), or react in a Norrish type I photocleavage reaction, forming formaldehyde and a benzyl radical, which is oxidised to benzaldehyde (**20**) when exposed to oxygen (Scheme **22**, **c**). Photolysis of pure phenylacetaldehyde at pH 11

under an air atmosphere did indeed yield benzaldehyde, confirming that this is an intermediate in the reaction. This can in turn undergo further oxidation to benzoic acid (**22**) (Scheme **22**, **d**), giving rise to the different ratios observed. Additionally, phenylacetic acid can undergo cleavage to yield benzaldehyde, formaldehyde, and formic acid (Scheme **22**, **e**).[80] In fact, when the reaction was repeated with degassed solvents under nitrogen atmosphere, benzaldehyde was not detected. However, all starting material was still consumed and formation of *p*-nitroaniline still ocurred. With no oxygen present, the benzyl radical can either dimerise, which is unlikely due to the low concentration in the reaction mixture, or simply abstract a proton, forming toluene. This would then evaporate during concentration under reduced pressure and elude detection. This mechanism would explain the two missing carbons, since formaldehyde would either remain in the aqueous phase as its hydrate or it would disappear in the rotary evaporator during concentration. The aromatic region in the ^{1}H-NMR spectra is messy with overlapping peaks and it is therefore difficult to assign any other signals.



Scheme 22. Proposed mechanism for the photodegradation of compound **1**.

Identical experiments were performed for the three other analogues (**2**-**4**), and dramatic differences in decomposition conversions were observed (Table **2**). When the nitro group is in *para*-position on the aniline (**1**), a 100% conversion is achieved at pH $\geq$ 11 (Table **2**, entries 6 and 7), whereas for the *meta*-analogue **2** an unsatisfactory 11% degradation occurs (Table **2**, entry 7). At pH values lower than 7, only trace amounts are detected, and for **3** and **4** a pH $\geq$ 11 is required for any degradation to occur at all (Table **2**, entries 6 and 7). These results clearly show that *para*-nitro substitution is optimal for photodecomposition, especially when positioned on the aniline (**1**), which even display 32% degradation at pH 7 and 75% at pH 9 (Table **2**).

Table 2. Percent conversion for photodecomposition reactions for compound **1-4**. Conversions are obtained from normalised integral values for *o*-protons on the aniline.

Entry	pH	1	2	3	4
1	1	9	trace	-	-
2	3	9	trace	-	-
3	5	9	trace	-	-
4	7	32	4	-	-
5	9	75	5	-	-
6	11	100	11	38	20
7	13	100	17	40	56

To shed some more light on the decomposition mechanism during the photochemical reaction, a mechanistic study following the reaction with ^{1}H NMR and by performing the photolysis in an NMR tube was performed. Photolysis was done by placing a 28.0 mM solution of **1** in 50:50 deuterated acetonitrile:deuterium oxide adjusted to pH 13 with sodium deuteroxide, next to the irradiation source, and recording a ^{1}H-NMR spectrum with increasing intervals (Figure **10**). As expected, the aldehydic signal at 9.91 ppm belonging to benzaldehyde starts appearing with increasing intensity together with the doublet corresponding to *p*-nitroaniline at 6.62 ppm. A slight difference in chemical shift for the aldehyde is observed since the solvent used is a mixture of acetonitrile and water instead of chloroform. Interestingly, an additional singlet at 8.34 ppm appears, which suggests formation of a formamide, perhaps *N*-(4-nitrophenyl)formamide. However, the aromatic signals that should accompany this singlet are not seen, and there is no carbon signal

around 160 ppm, thereby disproving this theory. There is, however, a sharp peak at 171.1 ppm in the ^{13}C-NMR spectrum recorded after 240 min of irradiation, corresponding well with benzoic acid ($\Delta\delta$ = 0.913 ppm), confirming the suspicion that benzaldehyde is further oxidised to the corresponding acid. However, the reaction never went to completion, and after 24 hours of irradiation, around half of the starting material remained unreacted. As the reaction progresses, an insoluble dark film starts forming on the glass wall, preventing light from penetrating into the solution. This was not observed in any previous studies because the concentration was around 50 times lower and there was continuous stirring, which was not possible when doing the reaction inside the NMR tube. Nonetheless, this experiment provides valuable insight into what is actually taking place in the reaction.

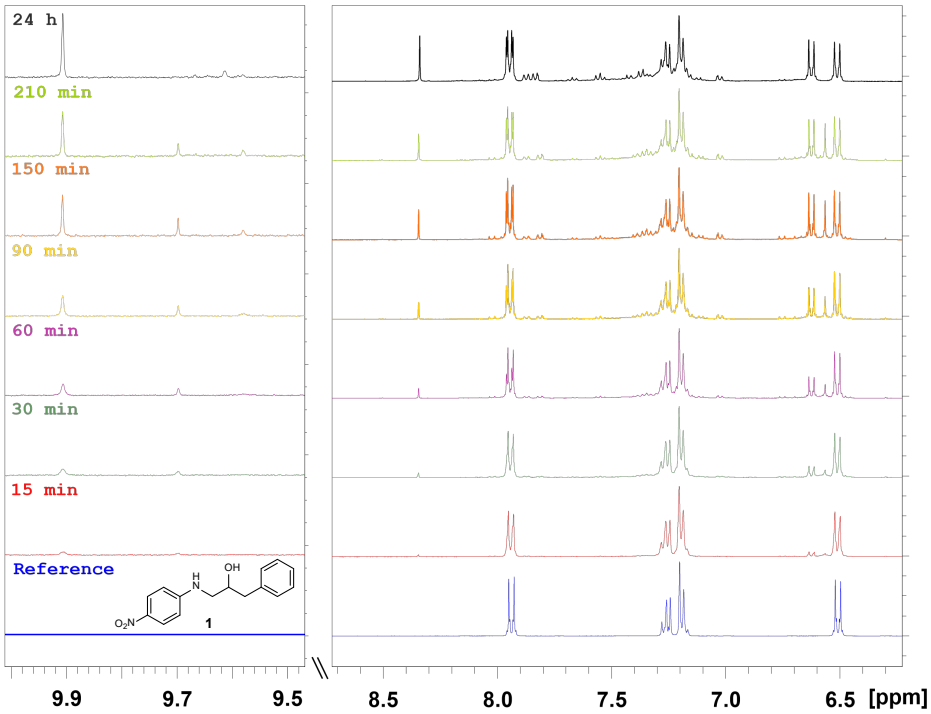


Figure 10. ^{1}H-NMR study of the photodecomposition of **1**. Lamp: medium-pressure mercury vapour.

When examining the region between 3-5 ppm in the ^{1}H-NMR spectra and comparing with a reference spectrum of formaldehyde, it seems likely that this is one of the decomposition products. The singlet at 4.59 ppm corresponds perfectly with formaldehyde, but the other peaks from 4.54 to 4.90 are not matching (Figure **11**). In aqueous solution formaldehyde exists mainly as the hydrate, which can also form dimethylene glycol and oligomers, which is why more than one signal is observed.[81] Based on these signals it is highly likely that formaldehyde is, indeed, a product of the photodegradation reaction.

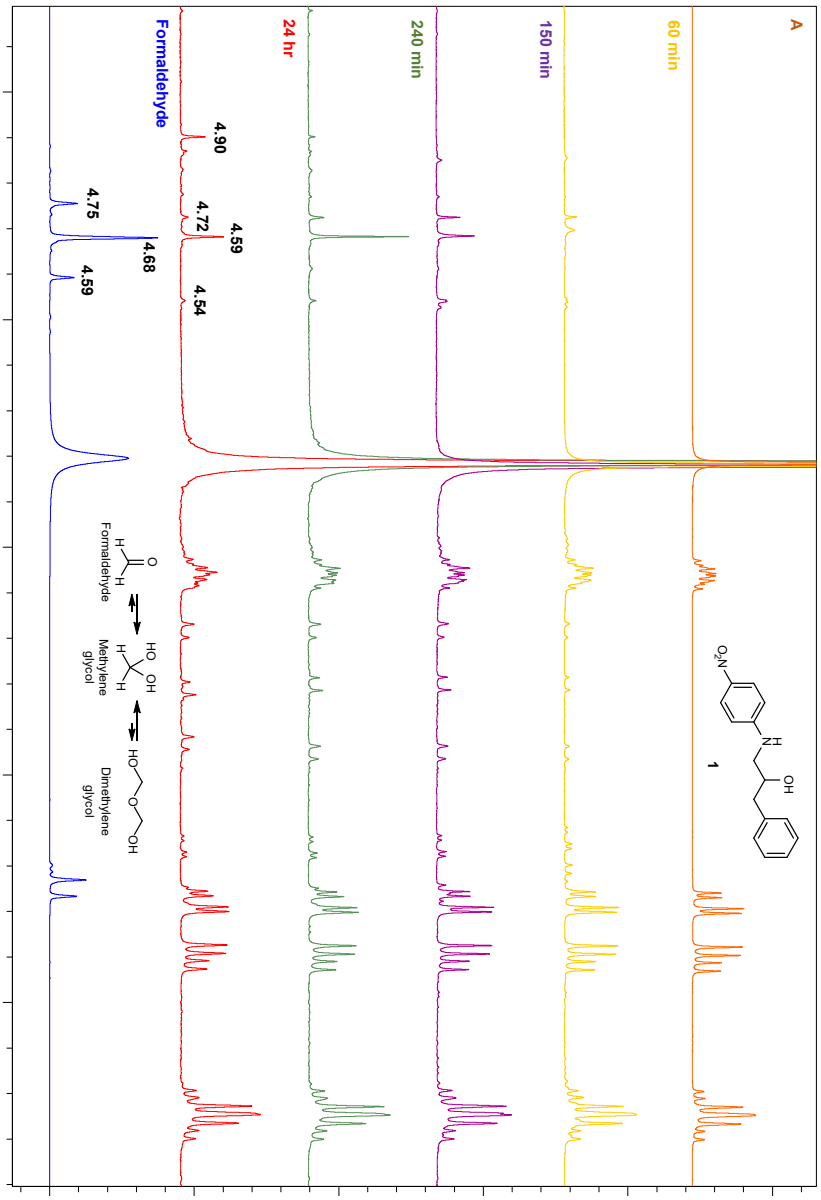
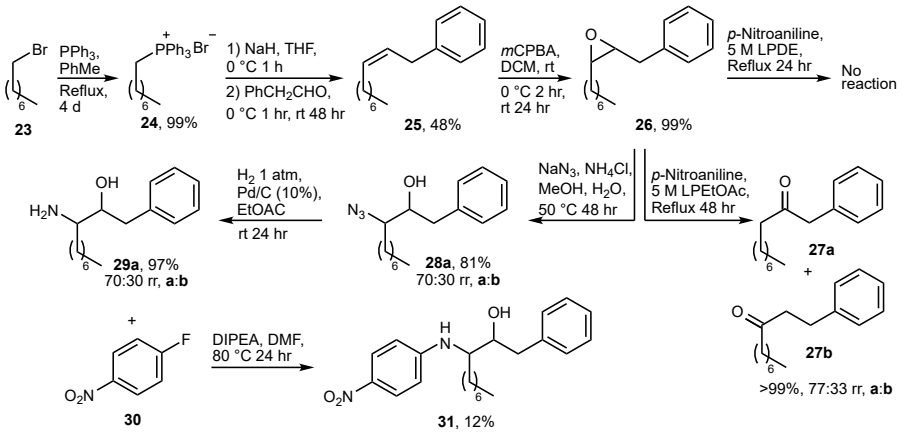


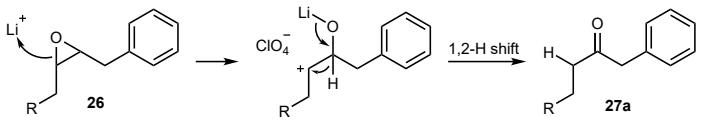
Figure 11. ^{1}H-NMR spectra of pure compound **1** before photolysis and degradation mixture after 60, 150, 240 min, and 24 hr, and formaldehyde.

To be absolutely certain that formaldehyde is formed in the reaction, an analogue of compound **1** with a long alkyl chain at C1 was synthesised (Scheme **23**). Assuming that an analoguous photodegradation mechanism occurs, this reaction would yield an alkyl aldehyde with a lipophilic and non-volatile nature with no risk of forming hydrates or disappearing in the rotary evaporator, which would be easy to detect with NMR spectroscopy. To this end, a Wittig reaction with octyltriphenylphosphonium bromide (**24**) and phenylacetaldehyde yielded *Z*-alkene **25** in 48% yield. Oxidation with *m*CPBA gave epoxide **26** in near quantitative yield, but when attempting a ring-opening reaction with *p*-nitroaniline using the 5 M LPDE conditions, no reaction was observed. 5 M LPDE was substituted with 5 M LPEtOAc and the reaction mixture was heated for 2 days at reflux, but the desired product was again not detected. Instead, a 77:33 regioisomeric mixture of ketones **27** was formed in 99% combined yield. These products are the result of a Lewis-acid promoted epoxide-ketone rearrangement, where the ring is opened by lithium and a subsequent 1,2-hydride shift yields the ketone (Scheme **24**). Since the epoxide can be opened at two positions, there are two regioisomers forming.



Scheme 23. Complete synthesis of compound **31**.

Instead, the epoxide was successfully opened with sodium azide giving a 70:30 mixture of regioisomers **28** in favor of the desired product in 81% combined yield, which was incredibly difficult to separate by column chromatography. Therefore the compounds were kept as a mixture in the following step, which was a simple palladium-catalysed hydrogenation giving a 70:30 mixture of amines **29** in 97% combined yield. Again, separating the isomers was difficult and the next step was carried out in the hope that the target compound would be isolable from the product mixture. Luckily, after a nucleophilic aromatic substitution reaction with 1-fluoro-4-nitrobenzene (**30**), the target compound (**31**) was isolated from a 70:30 mixture of regioisomers by column chromatography in 12% yield (Scheme **23**). The two different isomers were identified by the ratios of ^{1}H NMR using the signals for the benzylic methylene protons at 2.82 and 2.94 ppm.



Scheme 24. Lewis-acid promoted epoxide-ketone rearrangement.

Amino alcohol **31** was then photolysed at pH 11 with a medium-pressure mercury-vapour lamp for 4 hours, which achieved 64% conversion. A control experiment performed in the dark revealed that there are no reactions taking place in the basic environment in the absence of light. The ^{1}H-NMR spectrum of the crude reaction mixture after aqueous workup again shows formation of both *p*-nitroaniline and benzaldehyde by appearance of a doublet at δ 6.61 ppm and a singlet at δ 10.02 ppm, respectively (Figure **12**). This time however, an additional aldehydic signal is visible at δ 9.76 ppm, appearing as a triplet with a coupling constant of 1.9 Hz. This could belong to either phenylacetaldehyde (**17**), which is confirmed to be an intermediate for formation of benzaldehyde (**20**), or to octanal (**32**), as their chemical shift values are indistinguishable. However, phenylacetaldehyde displays a coupling constant of 2.3 Hz, and the methylene group has a characteristic doublet at δ 3.69 ppm,[82] which is not visible in the ^{1}H-NMR-spectrum of the crude reaction mixture.

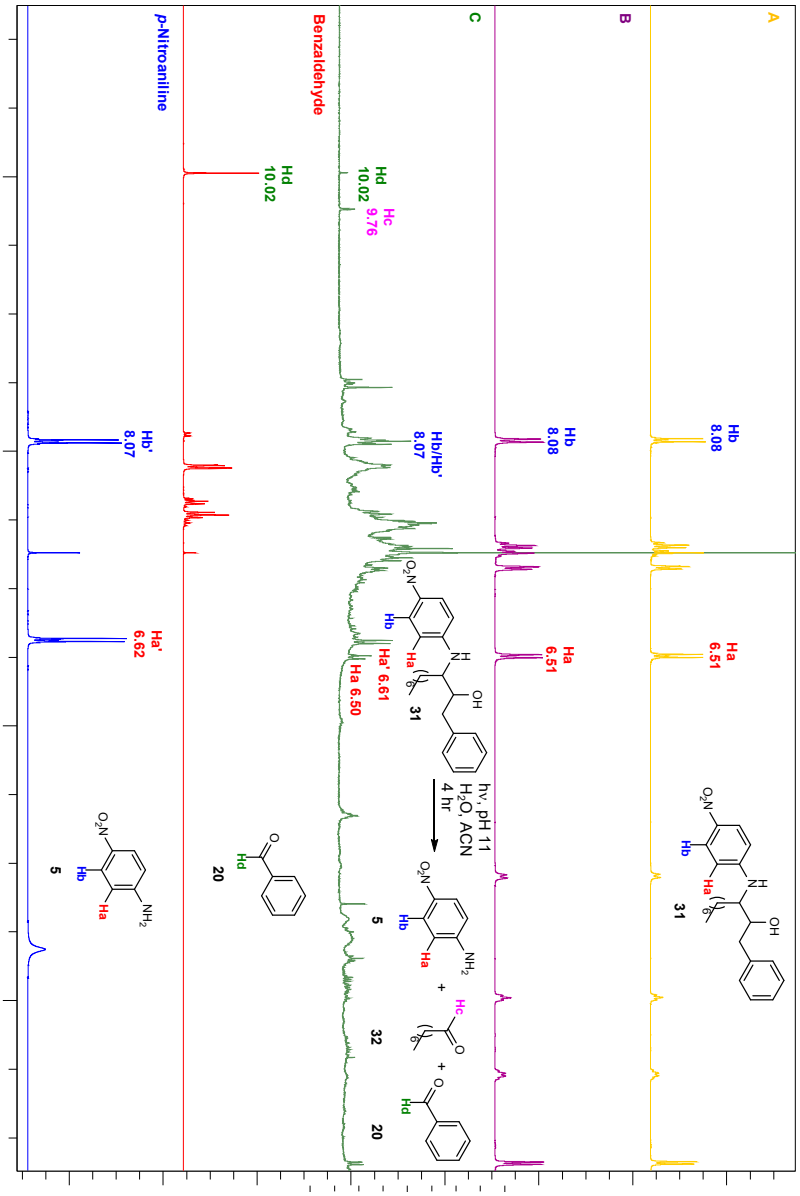


Figure 12. ^{1}H-NMR spectra of pure compound **31** (A), control experiment performed in the dark for 4 hr at pH 11 (B), crude degradation mixture after 4 hr photolysis at pH 11 (C), benzaldehyde and *p*-nitroaniline.

Additionally, a COSY correlation from the aldehyde triplet at δ 9.76 ppm to an aliphatic multiplet at δ 2.42 ppm is observed (Figure **13**), which is characteristic for octanal (**32**).[82,83] After 3 days at room temperature in the dark, only traces of the aldehyde signal remained. Almost full conversion to octanoic acid had ocurred, confirmed by an HMBC correlation from δ 2.33 ppm to δ 174.0 ppm, corresponding well with literature values,[84] and TLC-MS (Exact mass 144.12, detected m/z 143.67 [M-H]$^-$ and 179.03 [M+Cl]$^-$).

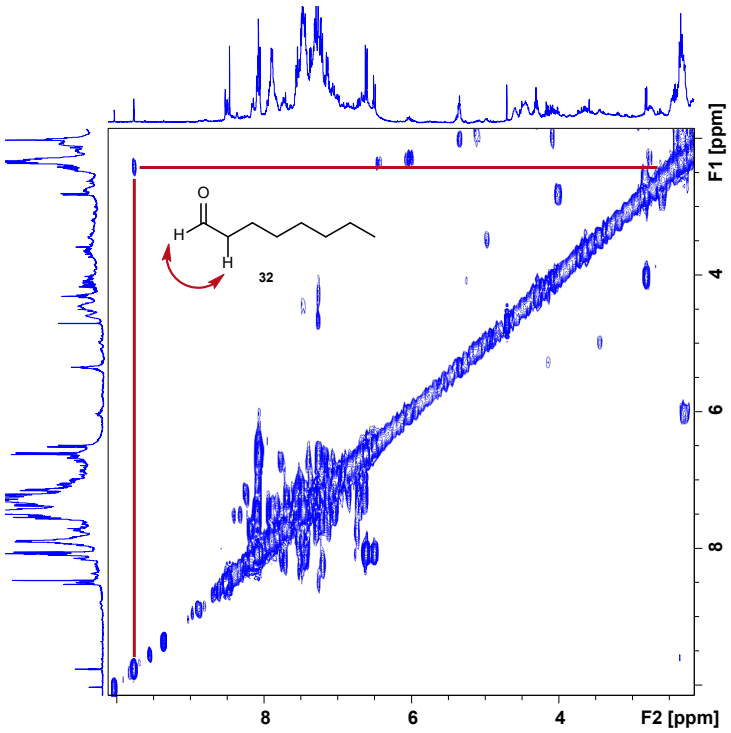


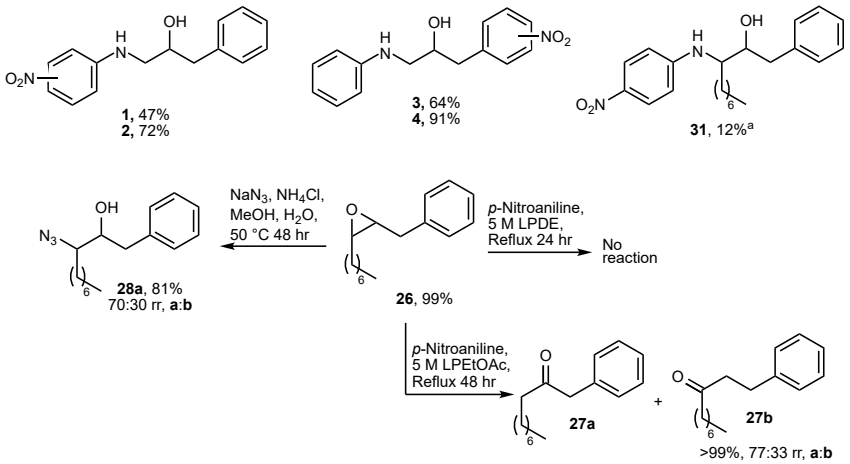
Figure 13. COSY correlation confirming the formation of octanal (**32**) in the photodecomposition of compound **31**.

These results clearly confirm that formaldehyde is one of the degradation products in the photolysis of amino alcohol **1**, together with

p-nitroaniline (**5**), and that benzaldehyde (**20**) is formed from phenylacetaldehyde (**17**) or phenylacetic acid (**21**) in a secondary photolytic reaction. This means that the other missing carbon in the mechanism has to be either a second unit of formaldehyde or formic acid.

II.3 Summary of synthetic outcomes

Aminols **1-4** was successfully synthesised throught epoxide ring-opening reactions in yields ranging from 47-91%. Compound **31**, with a heptyl alkyl chain, was achieved through six steps in 12% yield yield based on the final reaction. An ring-opening reaction with epoxide **26** and *p*-nitroaniline was attempted, but either there was no reaction and recovery of starting materials, or an epoxide-ketone rearrangements occurred, giving ketones **27a** and **27b** (Scheme **25**). Compound **31** was therefore synthesised from azide **28a** through palladium-catalysed reduction to the amine followed by a nucleophilic aromatic substitution reaction with 1-fluoro-4-nitrobenzene.



Scheme 25. Summary of synthetic outcomes in Part II.

II.4 Concluding remarks

Part II presents the successful development of four photodegradable scaffolds based on photo-retro-aldol chemistry and the chitin synthase inhibitors diflubenzuron and teflubenzuron. The syntheses of these compounds are described in detail, as well as the photodegradation conversions. The compound that displayed superior degradation, amino alcohol **1**, was selected for a mechanistic investigation, and during the photodegradation studies, this compound fragmented into *p*-nitroaniline (**5**) and phenylacetaldehyde (**17**), which further degraded into benzaldehyde (**20**), by cleaving off a unit of formaldehyde or formic acid. This left one carbon atom in the structure unaccounted for, and it was hypothesised that this carbon also ended up as formaldehyde, either remaining in the aqueous phase as its hydrate, or it evaporated in the rotary evaporator, eluding detec-

tion. This suspicion was investigated by synthesising an analogue with a heptyl chain at C1 (**31**), which retained the suspected aldehyde in the organic phase and prevented evaporation. Octanal (**32**) was detected by ^{1}H NMR, which over time oxidised to octanoic acid, confirmed by both ^{1}H NMR and LRMS. These results clearly indicate that formaldehyde was indeed formed in the photodegradation of compound **1**.

Part III

Chitin synthase inhibitors

By functionalising the photodegradable scaffolds described in Part II with halogens in the appropriate positions, a series of analogues of these benzoyl urea parasiticides were envisioned. Following the synthesis of these compounds, they were tested for anti-lice activity before and after photodegradation. Since the *para*-nitro substituted 1-(arylamino)-3-arylpropan-2-ol derivatives displayed superior photodecomposition compared to the *meta*-substituted ones, the analogues were solely based on these two scaffolds (Figure **14**).

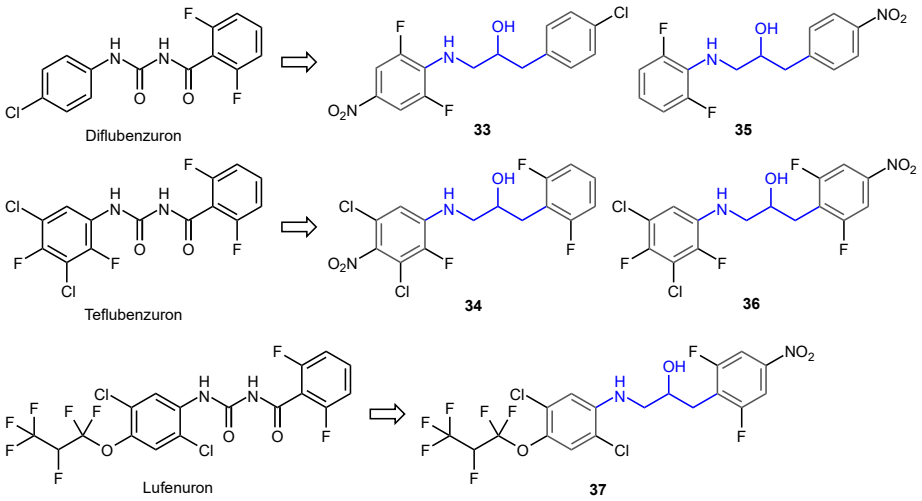
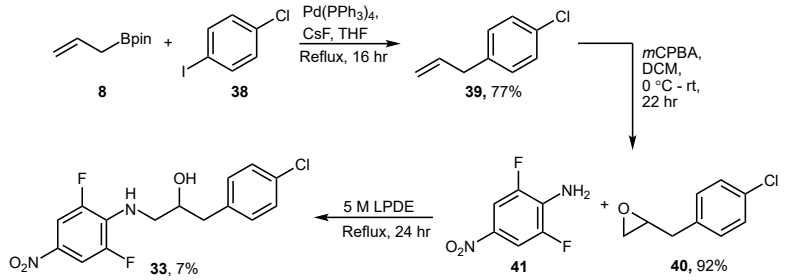


Figure 14. Photodegradable analogues of the benzoyl urea parasiticides diflubenzuron, teflubenzuron, and lufenuron.

III.1 Synthesis of flubenzuron analogues

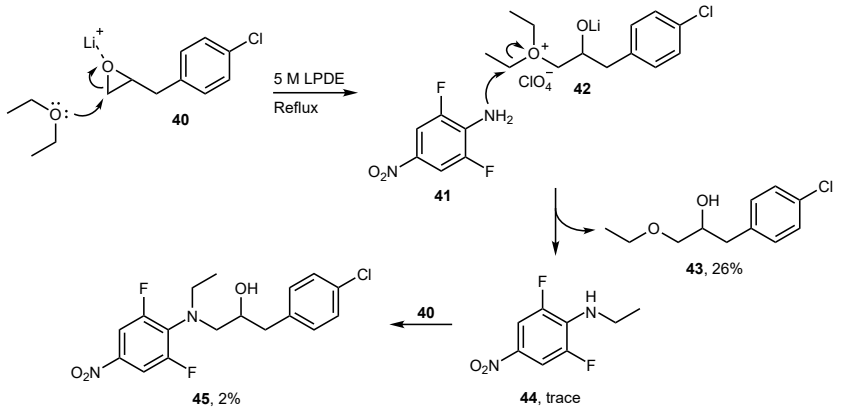
A synthetic route similar to the one used for compound **3** and **4** was planned for diflubenzuron analogue **33**. A Suzuki-Miyaura cross-coupling reaction under anhydrous conditions gave the allylated product **39**, and treatment with *m*CPBA yielded epoxide **40** in 71% overall yield. However, when attempting to react the epoxide with aniline **41** in 5 M LPDE medium, the desired amino alcohol **33** was obtained in only 7% yield after a challenging purification (Scheme **26**). Aniline **41**, which is highly deactivated due to the nitro group and the two fluorine atoms, is not nucleophilic enough to efficiently open the epoxide under these conditions.



Scheme 26. Synthesis of diflubenzuron analogue **33** through palladium-catalysed allylation, epoxidation, and lewis acid-promoted epoxide ring opening.

Three unexpected products were also formed; ethoxylated epoxide **43**, ethylated aniline **44**, and ethylated target molecule **45** (Scheme **27**). A proposed mechanism for the unexpected epoxide ring opening proceeds through nucleophilic attack by diethyl ether at the lithium-coordinated epoxide to form trialkyloxonium ion **42**. These

compounds act as strong alkylating agents,[85] ethylating the starting aniline **41**, yielding **44** (trace amounts), while simultaneously liberating the ring-opened product **43** (26%). Additionally, compound **44**, which is sufficiently nucleophilic due to positive inductive effect from the ethyl group, reacts with the remaining epoxide yielding amino alcohol **45** (2%). Unreacted aniline **41** was also present in the reaction mixture, as indicated by TLC analysis. The reaction was performed again over 72 hr, resulting in 36% ethoxylated epoxide **43**, 50% unreacted epoxide **40**, and trace amounts of ethylated product **45**. Additionally, there were at least three unidentified compounds in the mixture that were too difficult to isolate by column chromatography, and structure elucidation from crude NMR spectra was challenging.



Scheme 27. Proposed mechanism for epoxide ring opening by diethyl ether.

Initial analysis by ^{1}H NMR revealed the ethoxy group, and studying the cross peaks in the ^{1}H-^{13}C-HMBC spectrum, confirmed formation

of compound **43** (Figure **15**). The carbons were identified using ^{1}H-^{13}C-HSQC NMR, and couplings $H_4 \rightarrow C_3$ (**green**) and $H_3 \rightarrow C_4$ (**purple**) reveals the structure. Compound **44** and **45** were identified by HMBC cross peaks from the ethyl group to the aromatic *ipso* carbon.

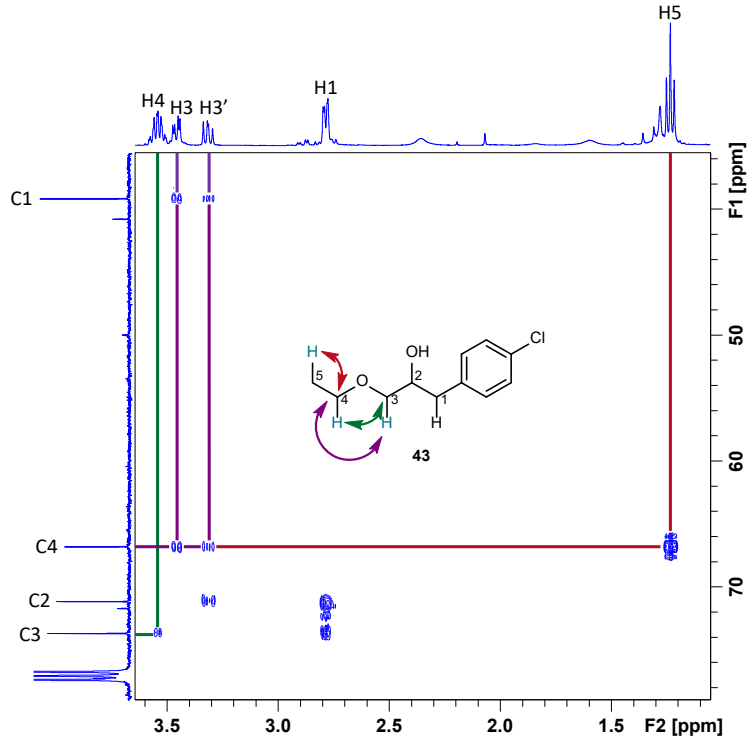
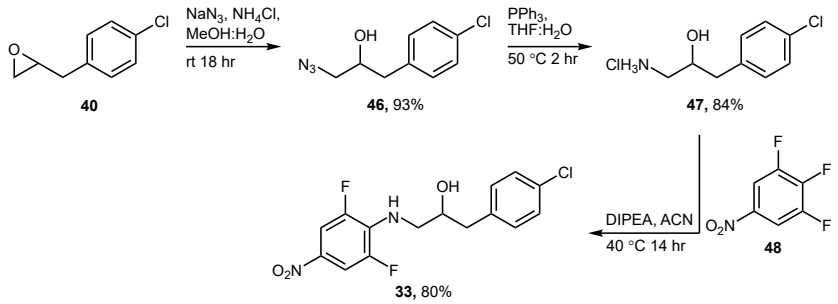


Figure 15. ^{1}H-^{13}C-HMBC couplings proving formation of ethoxylated side product **43**.

In order to avoid all these side products, a different synthetic route was developed starting from the same epoxide **40**. Ring opening by sodium azide in methanol and water[86] yielded 1,2-azido alcohol **46** in 93% yield, which was readily reduced to the primary amine under Staudinger conditions[87,88] and isolated as hydrochloride salt **47** in

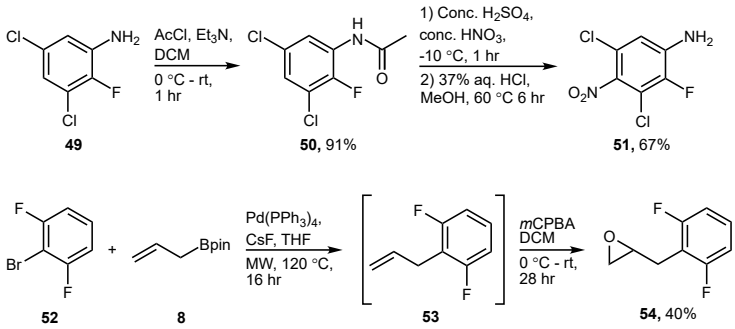
excellent yield (84%). A nucleophilic aromatic substitution reaction with the heavily activated fluorobenzene **48** gave the desired amino alcohol **33** in 44% overall yield through five steps (Scheme **28**).



Scheme 28. Synthesis of diflubenzuron analogue **33** through epoxide ring opening with sodium azide, Staudinger reduction, and nucleophilic aromatic substitution.

For teflubenzuron analogue **34**, the starting nitroaniline was not commercially available and had to be synthesised from aniline **49**. Since the nitro group is introduced by nitration, the amino group had to be protected in order to avoid deactivation and tarry oxidation products.[89] Nitroaniline **51** was therefore obtained through acetylation, nitration, and hydrolysis in 61% overall yield (Scheme **29**). Epoxide **54** was synthesised starting with a microwave-assisted palladium-catalysed allylation[76] of difluorobromobenzene **52**, followed by direct epoxidation with *m*CPBA in 40% yield over two steps. Special care had to be taken when handling allylbenzene **53** due to its volatile nature and its ability to evaporate off the TLC plate. For this reason it was not characterised by NMR, but it could

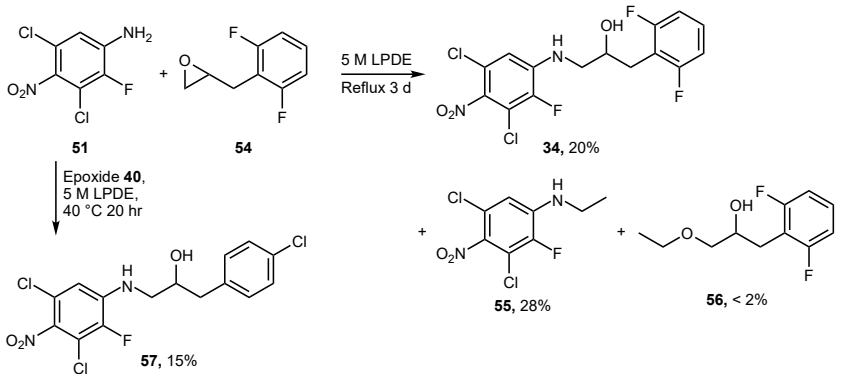
be identified by its sharp pine-like smell, which is characteristic for some phenylpropanoids.[90]



Scheme 29. Synthesis of the two key building blocks (**51** and **54**) for teflubenzuron analogue **34**.

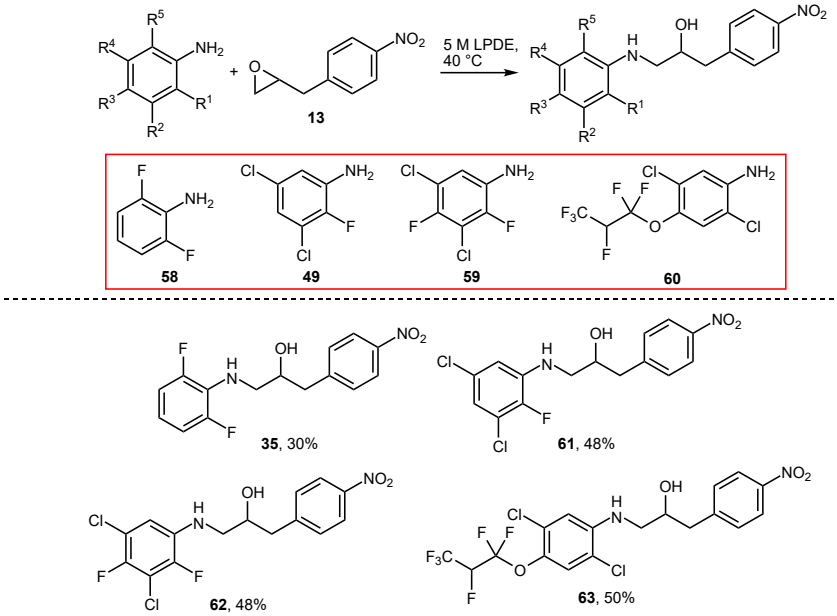
With aniline **51** and epoxide **54** in hand, teflubenzuron analogue **34** was successfully synthesised in 20% yield by refluxing in 5 M LPDE medium. As expected, a substantial amount of ethylated aniline **55** was isolated, along with 34% recovery of starting material **51** (Scheme **30**). Although the nucleophile is heavily deactivated leading to poor yield and side reactions, it seems to be more reactive than **41** since the target molecule is formed in greater quantity. Additionally, ethoxylated epoxide **56** was detected, meaning that an analogous mechanism to the synthesis of diflubenzuron analogue **33** is taking place (Scheme **27**). The isolated yield of compound **56** is reported as less than 2% because the sample was contaminated with aniline **51**, ethyl acetate, and small amounts of grease. Additionally, by reacting aniline **51** with epoxide **40**, amino alcohol **57** was syn-

thesised under the same conditions and isolated in 15% yield after a challenging purification.



Scheme 30. Synthesis of teflubenzuron analogue **34**.

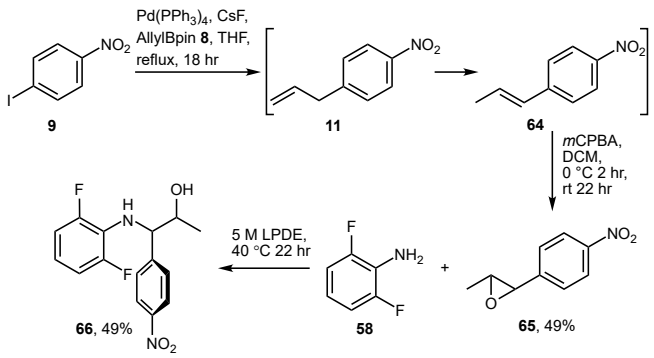
By reacting commercially available anilines **49** and **58-60** with nitrophenylepoxide **13** using the 5 M LPDE conditions, a group of aminols were successfully synthesised and isolated in moderate yields of 30-50% (Scheme **31**). The reactions were purposefully not driven to completion in order to avoid any aniline ethylation and epoxide ethoxylation, which made the purification process simpler. Epoxide **13** was recovered in yields ranging from 24-43%.



Scheme 31. Synthesis of flubenzuron analogues with the 4-nitrophenyl group.

During a period where the NMR instrument was out of order, epoxide **65** was accidentally synthesised when attempting to make more of compound **13**. It was discovered that if no water is added to the Suzuki-Miyaura cross coupling, and the reaction is left stirring overnight, complete isomerisation from allylbenzene **11** to 1-propenylbenzene **64** occurs (Scheme **32**). Transition-metal catalysed allylbenzene to 1-propenylbenzene isomerisation is a well known phenomenon,[91] but this was not detected in any of the other cross-coupling reactions. Subsequent oxidation with *m*CPBA gave *trans*-epoxide **65** in 49% overall yield, distinguishable from a theoretically possible *cis*-isomer by a coupling constant of 1.9 Hz, which is characteristic for the *trans*-configuration.[92] However, due to the lack of

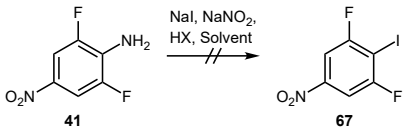
NMR analysis, this change of events was not discovered until after aminol **66** was synthesised, and no NMR data was recorded for 1-propenylbenzene **64**.



Scheme 32. Isomerisation of allylbenzene **11** into 1-propenylbenzene **64** and synthesis of aminol **66**.

Synthesising teflubenzuron analogue **36** however, proved to be more challenging, as an appropriately substituted aryl halide for palladium-catalysed allylation was not readily commercially available. It was envisioned that this compound, aryl iodide **67**, could be obtained through a Sandmeyer reaction from aniline **41** (Scheme **33**). At first, a diazotisation-halogenation reaction was attempted following a procedure described by Billamboz and co-workers for the iodination of 5-amino-2-carboxymethylbenzoic acid using hydriodic acid, sodium nitrite, and cuprous iodide in water.[93] The target product was formed in a disappointing 8% yield along with 67% recovery of starting material (Table **3**, entry 1). The amounts of sodium iodide and sodium nitrite were increased, and the temperature was increased slightly, but this led to complete decomposition of aniline

41 (Table **3**, entry 2). The reactions were repeated a couple of times, but the outcome remained unchanged.



Scheme 33. Attempted Sandmeyer-type reaction to form compound **67**.

A third attempt with sodium iodide, sodium nitrite, and sulfuric acid in acetonitrile and water did not give any of the desired product, and analysis by TLC-MS revealed formation of 2,6-difluoro-4-nitrophenol (Exact mass 175.01, detected m/z 174.02 [M-H]$^-$) (Table **3**, entry 3). When performing the reaction with hydrochloric acid in water or a mixture of water and acetonitrile, following a modified procedure reported by Holmes and co-workers,[94] the product was not detected, and complete decomposition of the starting material was observed (Table **3**, entry 4 and 5). Presumably, one of the products was the corresponding phenol, but TLC-MS analysis was not used to confirm this. Procedures with hydrobromic acid in DMSO[95] and *N*-iodosuccinimide (NIS) in DMF[96] gave no reaction at all, and the starting material was recovered (Table **3**, entry 6 and 7). A few attempts using *p*-toluenesulfonic acid (*p*-TSA) and cuprous iodide in acetic acid mainly resulted in decomposition of the starting material, and the best yield obtained was 9% (Table **3**, entry 8 and 9). In one attempt, following a procedure described by Chi and coworkers for the diazotisation of a wide scope of *p*-nitro, cyano, chloro,

bromo, and iodosubstituted anilines,[97] quantitative conversion was observed (Table **3**, entry 10), but when scaling up from 0.30 to 0.90 mmol, only traces of the product were detected. The reaction was repeated many times, and it became apparent that the starting material was converted into many other products instead, one of which was identified as 2,6-difluoro-4-nitrophenol by TLC-MS. The reaction mixtures turned a deep purple colour in most of the attempts, indicating that the iodide (I^-) in solution is oxidised to iodine (I_2). This is known to occur in acidic solution with nitrous acid,[98] which is exactly the case in this reaction. If this process is dominating, it would consume both the iodide and the nitrite, preventing the desired reaction from taking place. The reaction was repeated many times with variations in the order the reagents were added, such as initial addition of sodium nitrite to the substrate in acidic solution followed by sodium iodide, or the other way around. However, only trace amounts of product were detected every time (Table **3**, entry 11-13). The required amount of aryl iodide **67** was never achieved, and focus was shifted towards finding an alternative synthesis of this compound.

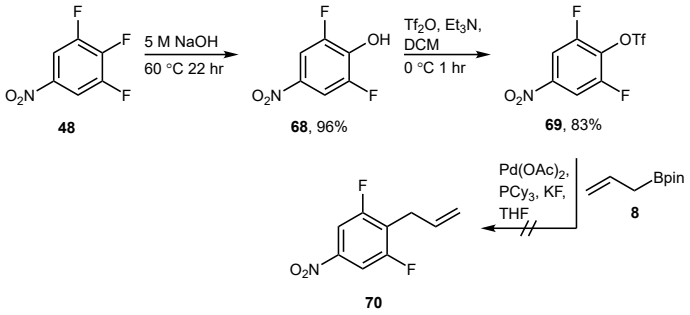
Table 3. Reaction conditions for the attempted Sandmeyer-type reaction.

Entry	NaI (equiv.)	$NaNO_2$ (equiv.)	CuI (equiv.)	Acid	Solvent	Temp. (°C)	Time (hr)	Yield (%)[a]
1[93]	1	4	1	HI (57%)	H_2O	0 - rt	24	8 (67)
2[93]	5	5	1.5	HI (57%)	H_2O	10 - 30	20	-[b]
3	5	5	-	H_2SO_4	ACN:H_2O	-10	1	-[c]
4[94]	2.5[d]	2	-	HCl (37%)	H_2O	0 - rt	1	-[b]
5[94]	3[d]	3	-	HCl (37%)	ACN:H_2O	0 - rt	24	-[b]
6[95]	4	5	-	HBr (32%)	DMSO	rt	24	- (Quant.)
7[96]	1[e]	1.5	-	-	DMF	rt	4	- (Quant.)
8	2.5	2	0.1	*p*-TSA	AcOH	rt	20	9[b]
9	-	20	1.25	*p*-TSA	AcOH	10 - 50	24	-[b]
10[97]	2	3	-	CSA	AcOH	rt	1.5	Quant.[f]
11[97]	2.5	3	-	CSA	AcOH	rt	1.5	Traces[b,c]
12[97]	2.5	1.5	-	CSA	EtOH:H_2O	0	20	- (Quant.)
13[97]	5	1.5	-	CSA	AcOH	0 - rt	20	50 (50)[f]

[a] Yields in parentheses represent recovered starting material. All conversions are estimated by ^{1}H NMR.
[b] Starting material decomposed.
[c] 2,6-Difluoro-4-nitrophenol was detected by LRMS.
[d] KI was used instead of NaI.
[e] NIS was used instead of NaI.
[f] Yield was not reproducable even after several attempts and only traces of the product were detected.

Starting from the activated fluoronitrobenzene **48**, it was possible to synthesise phenol **68** in a nucleophilic aromatic substitution reaction by simply heating compound **48** in 5 M aqueus sodium hydroxide solution in 96% yield. Following triflation under standard conditions,[99] a Suzuki-Miyaura palladium-catalysed allylation was attempted, in order to synthesise allylbenzene **70**. However, when using the conditions described earlier with $Ph(PPh_3)_4$ and CsF in THF, no product formation was observed. Another procedure, developed by Fu and coworkers,[100] with $Pd(OAc)_2$, PCy_3, and KF in THF at room temperature, also failed to give allylated product **70**. A third attempt with heterogeneous ligand-free conditions described

by Sajiki and coworkers,[101] using Pd/C (10%) and sodium phosphate in isopropanol and water at room temperature, resulted in the same outcome.



Scheme 34. Alternative approach to form allyl **70**.

In all cases, the reaction mixture turned a deep yellow color, which is characteristic for the deprotonated form of phenol **68** (Figure **16**). Both phenol **68** and triflate **69** are completely colourless compounds, making it highly likely that the colour change is due to decomposition into phenolate **71**. Additionally, conversion into the corresponding phenol is a known phenomenon in Suzuki-Miyaura cross-coupling reactions of electron-poor aryl triflates.[102]

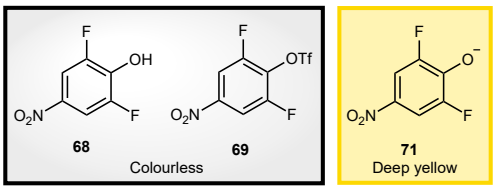
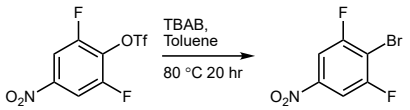


Figure 16. Deep yellow colour of phenolate **71** compared to the corresponding phenol **68** and triflate **69**.

This led to the development of a third approach, starting from the same aryl triflate **69**. Simply reacting 3,4,5-trifluoronitrobenzene

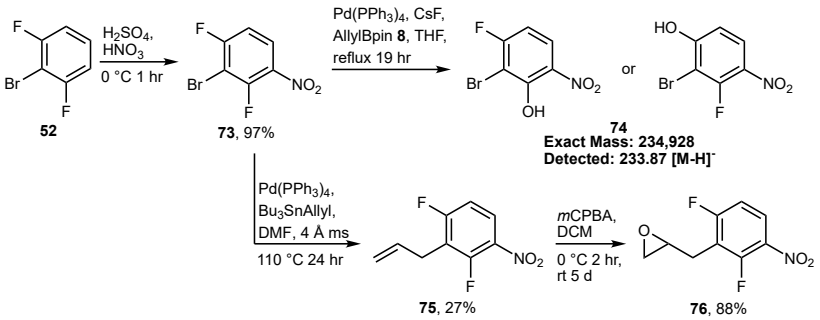
(**48**) with a halide source, such as LiBr, TBAB, or NaI, gave either no conversion at all or complete decomposition of starting material when attempting various conditions. However, Katritzky and coworkers developed a procedure for converting activated aryl triflates into aryl bromides by treatment with TBAB in refluxing toluene.[103] These conditions led to aryl bromide **72** in 6-14% yield after isolation by column chromatography (Scheme **35**). However, when attempting to scale up the reaction, the target compound was only detected in trace amounts. According to TLC analysis, there are at least three other products, and a strong yellow colour suggests formation of phenolate **71**. Continuing this method would consume significant amounts of 3,4,5-trifluoronitrobenzene (**48**), and was therefore considered non-viable.



69 **72**, 6-14%

Scheme 35. Alternative approach to form aryl halide **72**.

Another attempt at obtaining the elusive bromonitrobenzene **72** by nitrating bromide **52** was done in the hopes that a mixture of both nitration products would form. The reaction was highly selective with 97% yield and no need for column chromatography, but unfortunately compound **73** was the only product (Scheme **36**). Even though this was not the desired product, it would still be possible to make some interesting aminols with the formed isomer **73**.



Scheme 36. Synthesis of 2-(2,6-difluoro-3-nitrobenzyl)oxirane (**76**).

However, with the nitro group in this position, the compound can act as a strong electrophile in nucleophilic aromatic substitution reactions. This was indeed observed when attempting a Suzuki-Miyaura cross-coupling reaction. Instead of the allylated product, a simple S_NAr reaction with hydroxide gave phenol **74**, confirmed by TLC-MS (Figure **17**) and appearance of a bright yellow colour in the reaction mixture. Special care was taken to keep the reaction conditions anhydrous, but the extremely hygroscopic nature of cesium fluoride makes this difficult when not working in a glove box. This means that some water inevitably ends up in the vessel, destroying the reaction. No attempts were made at isolating this compound. Instead, a Stille coupling with allyltributylstannane in anhydrous DMF with $Pd(PPh_3)_4$ and 4 Å molecular sieves (4 Å MS) yielded the desired allylated product **75** in 27% isolated yield. Subsequent oxidation with *m*CPBA gave epoxide **76** in a satisfactory 88% yield.

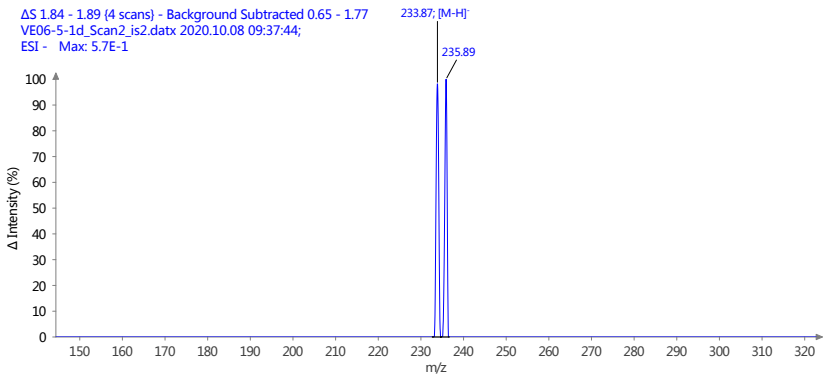
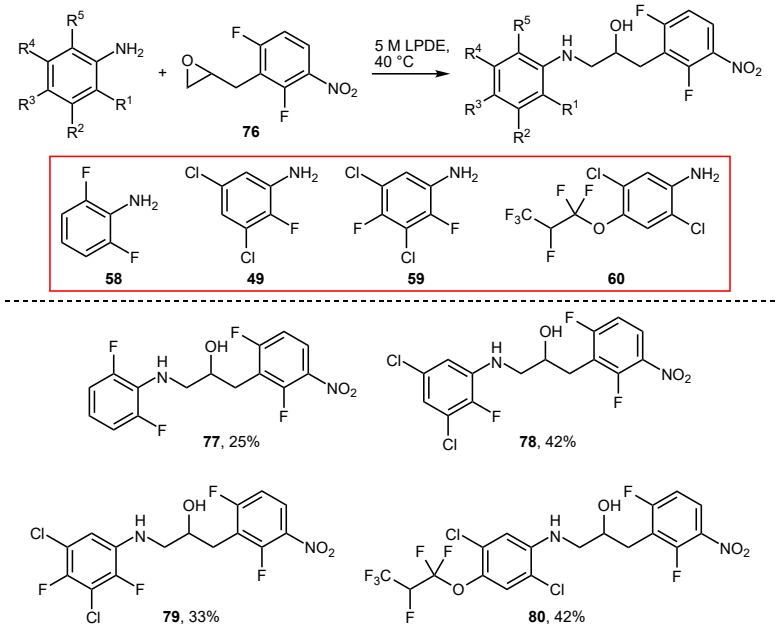


Figure 17. Low-resolution mass spectrum revealing formation of phenol **74**

Epoxide **76** was then ring opened with anilines **49** and **58-60** to form another series of 1-(arylamino)-3-aryl-propan-2-ols in moderate to good yields (Scheme **37**), along with 7-46% recovery of **76**. Again, the reactions were purposefully stopped before completion due to risk of aniline ethylation and epoxide ethoxylation.



Scheme 37. Synthesis of flubenzuron analogues with the 2,6-difluoro-3-nitro group.

III.2 Photodegradation of flubenzurons

In Part IV, the activity against salmon lice, the minimum inhibitory concentration (MIC) against a selection of Gram-positive and Gram-negative bacteria, and cytotoxicity against MRC5 and HepG2 cell lines of the target molecules are discussed. Four of them, **63**, **78**, **79**, and **80**, displayed antimicrobial activity at concentrations ranging from 6.3-50 μM and anti-lice activity at 0.1-0.001 ppt, and based on these promising results, the four compounds were subjected to photodegradation studies. In order to choose the optimal photochemical conditions, UV-vis spectroscopy was used to determine λ_{max} and the

molar attenuation coefficient (ϵ), revealing absorption maxima at around 250 nm (Figure **18**). Therefore, a 6 W low-pressure mercury-vapour lamp was chosen as the irradiation source for the degradation experiments, since it emits light mainly at 254 nm.

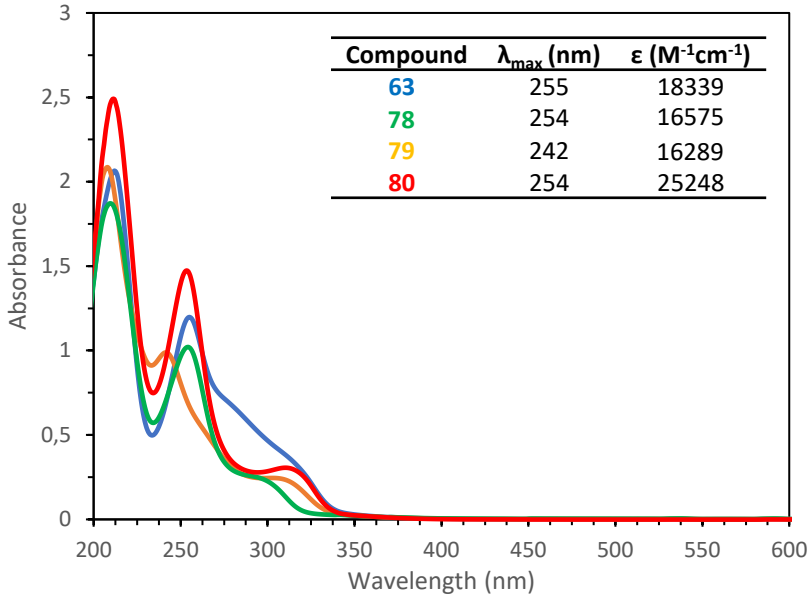


Figure 18. UV-vis spectra displaying λ_{max} and molar attenuation coefficient ϵ.

The photodecomposition study was started with compound **63**, and the reaction was carried out at pH 13 in acetonitrile and water. Photolysis after two and four hours did not achieve full conversion, but after 24 hours of continuous irradiation, the reaction was completed. The crude reaction mixture was analysed with ^{1}H-NMR spectroscopy, and aniline **60** and 4-nitrobenzoic acid (**81**) was revealed as two of the degradation products (Figure **19**), which was expected based on previous experiments. This is illustrated by the

complete disappearance of the two singlets at δ 7.32 and 6.80 ppm from aminol **63**, and the appearance of two new singlets at δ 7.30 and 6.93 ppm, corresponding perfectly with aniline **60** ($\Delta\delta = 0.00$ ppm). Additionally, the two doublets at δ 8.27 and 8.19 ppm fits well with 4-nitrobenzoic acid (**81**) ($\Delta\delta = 0.01$ and 0.00 ppm, respectively).

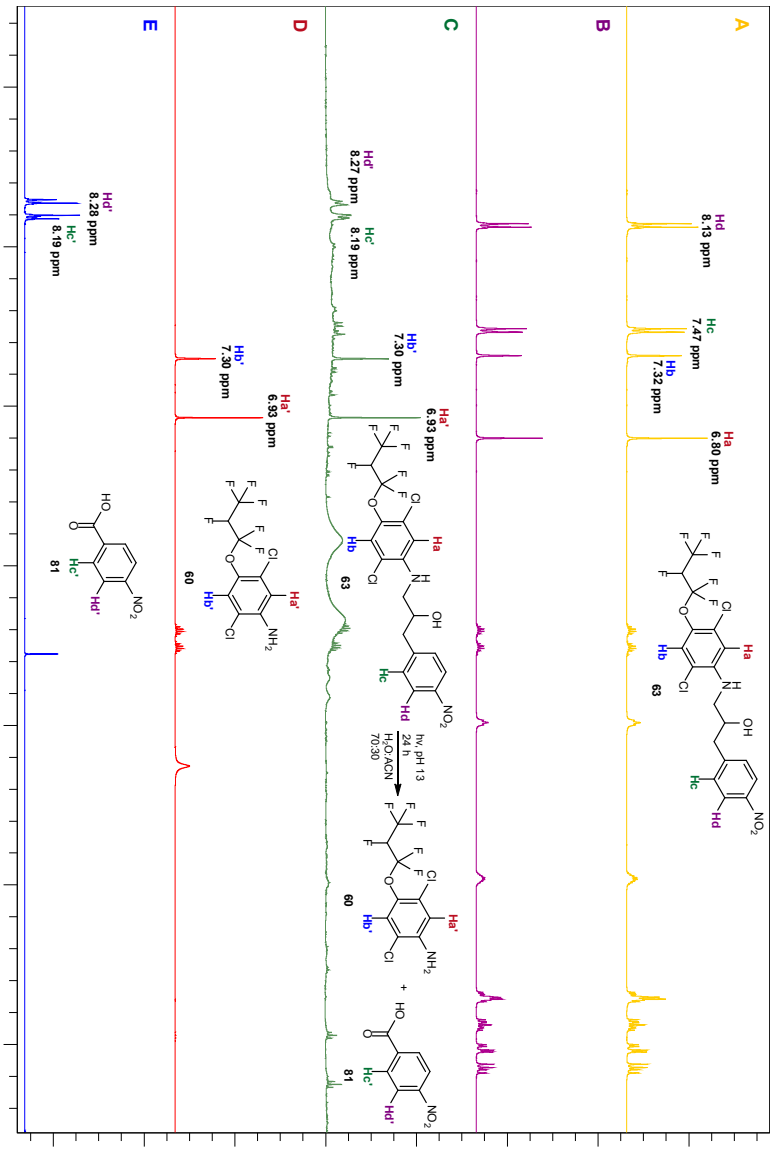


Figure 19. ^{1}H-NMR spectra of compound **63** before photolysis (A), control experiment after 24 hr at pH 13 in the dark (B), crude degradation mixture (C), degradation products **60** (D) and **81** (E) in CD_3CN at 400.13 MHz.

The NMR spectrum is quite complicated and there are several other peaks that were difficult to assign, suggesting further degradation of these fragments. Since the aniline and the carboxylic acid is formed, it can be assumed that a similar decomposition mechanism to the one taking place for compound **1** is occuring, and that formaldehyde and possibly formic acid are also products in the reaction. Identical photodegradation experiments at pH 13 were performed for the other three active compounds **78**, **79**, and **80**, but control experiments performed in the dark revealed that these compounds are somewhat unstable in basic environment at room temperature, with the exception of compound **63**, which was completely stable (Table **4**). They undergo a nucleophilic aromatic substitution reaction with hydroxide, which is illustrated by formation of phenol **82** and detected by mass spectrometry (Exact mass 392.01, detected m/z 391.35 [M-H]$^-$) in the control experiment of compound **78** (Figure **20**). This is further confirmed by ^{19}F NMR, and the ^{1}H-NMR spectrum of the control experiment showed that around 50% of the starting material underwent this unwanted reaction. This means that in the photodegradation at pH 13, there is an additional reaction taking place that are not initiated by light.

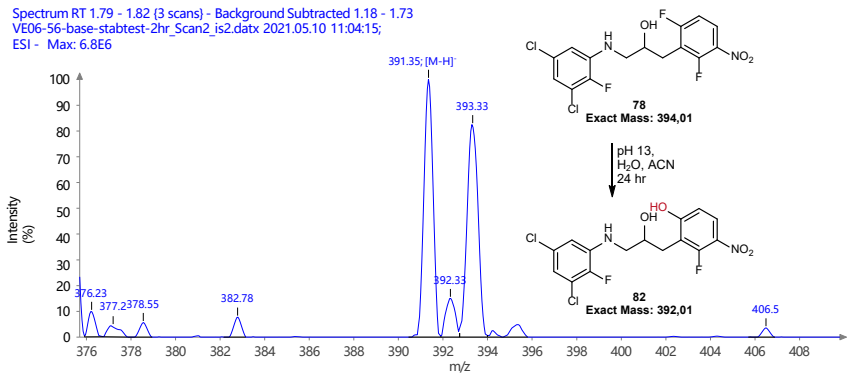


Figure 20. Mass spectrum showing instability of compound **78**.

When comparing the ^{1}H-NMR spectra of unreacted **78** (Figure **21**, **A**) with the crude reaction mixture after 24 hr stability test (Figure **21**, **B**) and after 24 hr photolysis (Figure **21**, **C**), this is indeed observed. The signals from the S_NAr reaction to give phenol **82** at δ 7.91 and 6.86 ppm are present in the mixture, along with aniline **49** at δ 6.76-6.71 ppm, as well as several other signals that at this point are not possible to assign. Similar results were obtained for compound **79** and **80**.

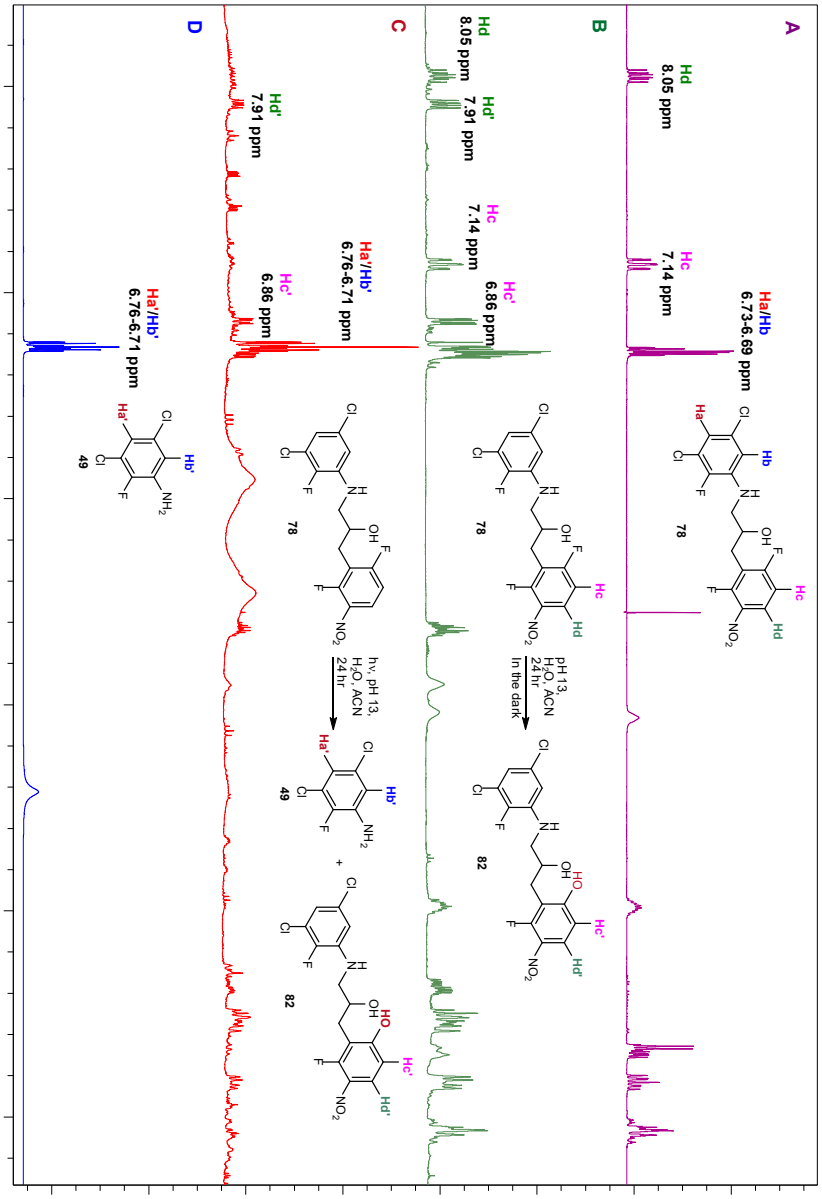


Figure 21. ^{1}H-NMR spectra of aminol **78** before photolysis (A), after 24 hr stability test at pH 13 (B), after 24 hr photolysis (C), and aniline **49** (D) in CD_3CN at 400.13 MHz.

Due to the instability of compounds **78**, **79**, and **80**, a series of photodegradation experiments were performed at pH 8. Control experiments in the dark revealed that all the compounds were stable for at least 24 hr at room temperature and pH 8. As the pH in seawater is around 7.5-8.4 depending on the region,[104] these experiments should simulate how the compounds would behave in a natural environment. Delightfully, 100% conversion was observed for compounds **78** and **80**, and compounds **63** and **79** had conversions of 25 and 19%, respectively (Table **4**). In comparison, diflubenzuron underwent 66% conversion at pH 8 and is therefore more photostable than compounds **78** and **80** (Table **4**, entry 5).

Table 4. Percent conversion for photodecomposition reactions for **63**, **78**, **79**, and **80**.

		Photolysis		**Stability**	
Entry	**Compound**	**pH 8 (%)[a]**	**pH 13 (%)[a]**	**pH 8**	**pH 13**
1	**63**	25	100	Yes	Yes
2	**78**	100	100	Yes	No
3	**79**	19	100	Yes	No
4	**80**	100	56	Yes	No
5	Diflubenzuron	66	-	-	-

[a] Conversions are estimated from ^{1}H-NMR spectra of the crude reaction mixtures after aqueous workup.

The photodegradation reactions that were performed at pH 8 were much cleaner than the ones at pH 13, which is illustrated in the ^{1}H-NMR spectra of compound **78** before (Figure **22**, **A**) and after (Figure **22**, **B**) photolysis. As seen in the spectra, the signals from the starting material at δ 8.05, 7.14, and 6.73-6.69 ppm have

completely disappeared, and there are four sharp signals appearing in the aromatic region at δ 7.76 (dd), 7.58 (dd), 7.41 (dd), and 6.23 (dd), which were also present in the spectrum after degradation at pH 13 (Figure **21**, **C**). According to ^{19}F NMR there are only two fluorine atoms present in the new product (Figure **23**, **B**) and TLC-MS gave a clear ion with m/z 419.36. This suggests that an intramolecular nucleophilic aromatic substitution reaction ocurrs, yielding tetrahydroquinoline **83**, meaning the detected ion corresponds to an [M+HCOO]$^-$ adduct. Since there are two fluorine atoms in suitable positions relative to the nitro group for this type of reaction to occur, fluorine-proton coupling constants were closely examined in order to establish which one acts as the leaving group. Firstly, a fluorine-decoupled ^{1}H-NMR spectrum was recorded to properly distinguish the proton-proton and proton-fluorine couplings from one another. A $^5J_{\mathrm{HF}}$ coupling (1.6 Hz) from Hc' and a $^4J_{\mathrm{HF}}$ coupling (8.9 Hz) from Hd' confirms the structure. If the reaction had occurred on the other fluorine atom, a $^3J_{\mathrm{HF}}$ coupling from Hc' would be seen, with a much higher value (around 9 Hz) than what is observed for the $^5J_{\mathrm{HF}}$ for compound **83**.[105] A similar reaction took place when photolysing compound **79**, as illustrated by the appearance of three new signals in the ^{19}F-NMR spectrum (Figure **23**, **D**), but the conversion was only 19% after 24 hr, possibly due to inductive effect from the fluorine atom in *para*-position on the aniline, which would make it less nucleophilic. An important

aspect of these transformations is that even though the starting material is consumed, photodecomposition into smaller fragments is not actually occuring. The tetrahydroquinoline products could still be antimicrobially active, and if not, they might possess another form of biological activity that is outside the scope of this thesis, making them a potential risk to non-target organisms.

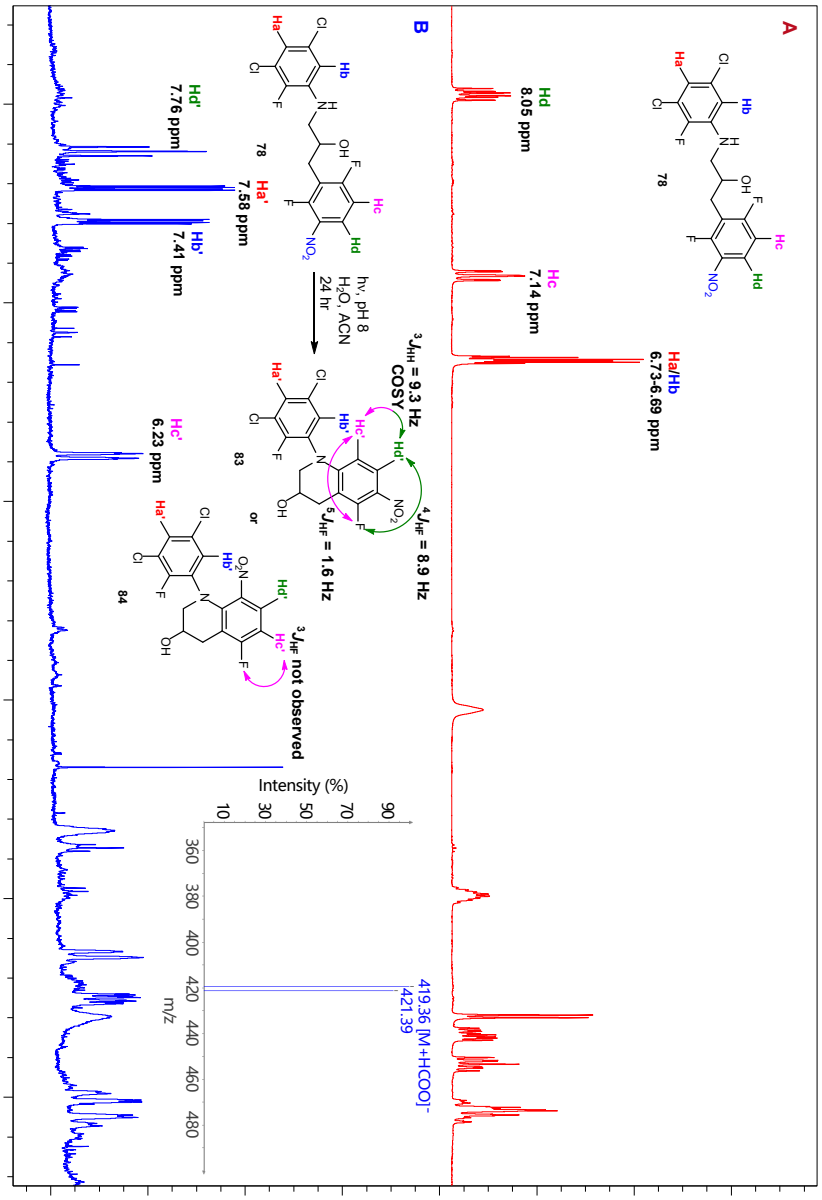


Figure 22. ^{1}H-NMR spectra of pure compound **78** before photolysis (A and C) and crude degradation mixture after 24 hr photolysis at pH 8 (B and D) in CD_3CN at 400.13 MHz.

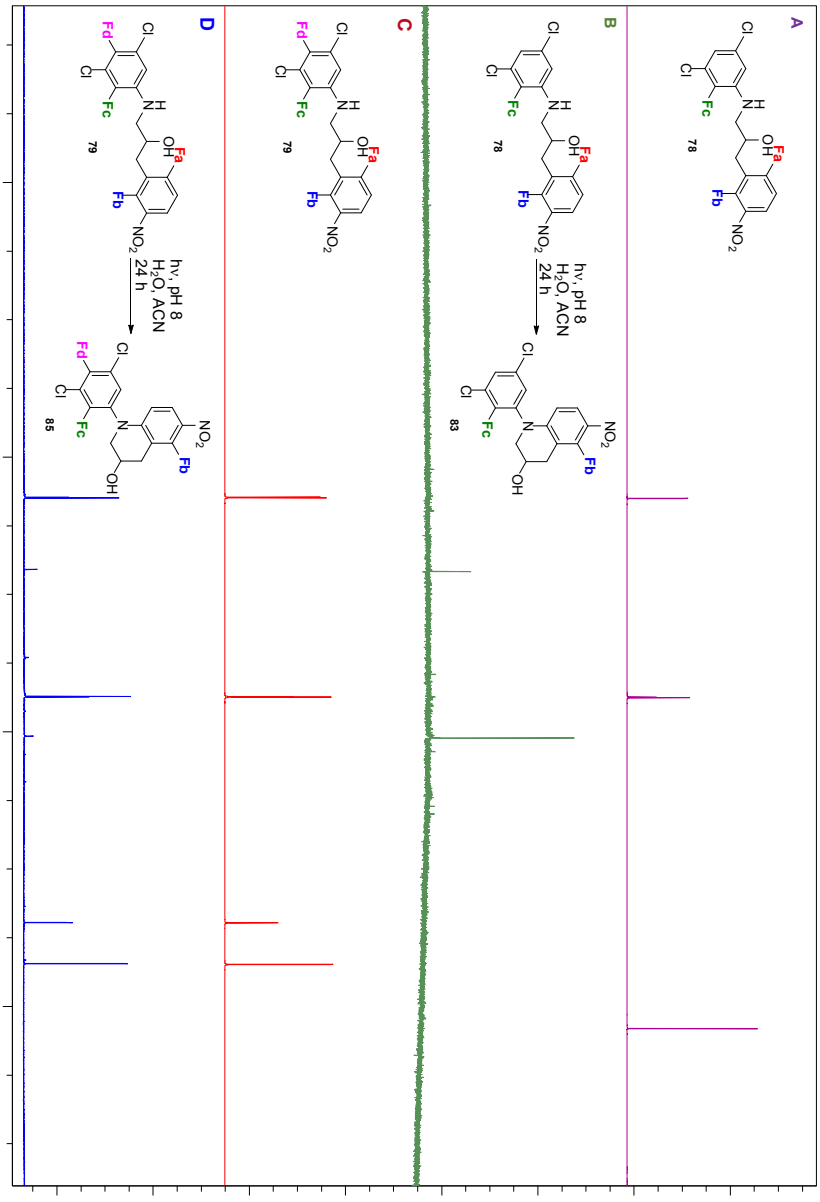


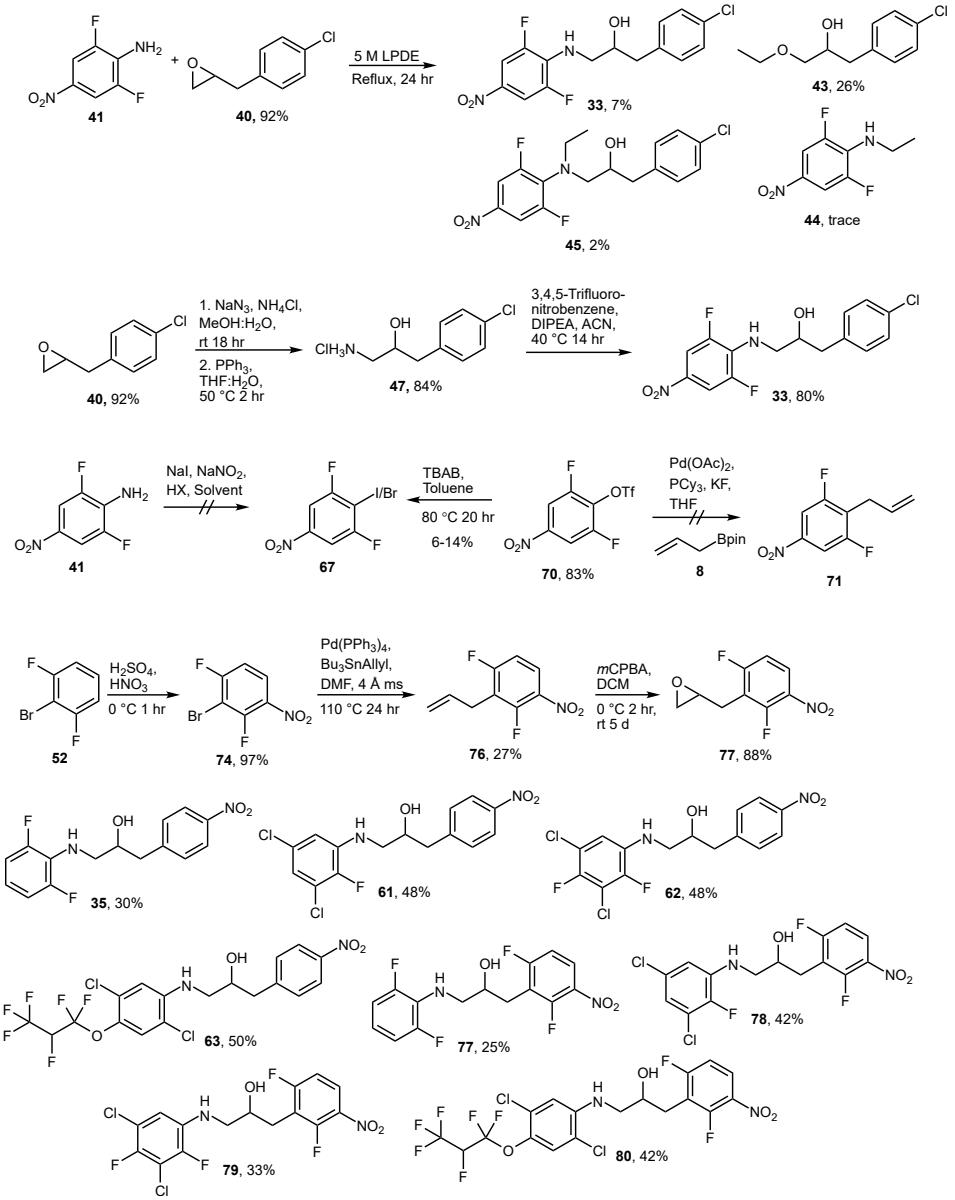
Figure 23. ^{19}F-NMR spectra of **78** and **79** before photolysis (A) and crude degradation mixture after 24 hr photolysis at pH 8 (B) in CD_3CN at 400.13 MHz.

For the degradation experiment at pH 8 with aminol **80**, the doublet of doublets at around δ 6.23 ppm did not appear, suggesting a different mechanism taking place. Unfortunately, the ^{1}H-NMR spectrum of the crude degradation mixture was quite complicated containing many unresolved multiplets and broad signals, making it difficult to assign any product. It is certain that a similar nucleophilic aromatic substitution that took place for **78** and **79** is not occuring, and neither is the photo-retro-aldol reaction due to the absence of signals from aniline **60**. However, the important part at this point is that complete photodecomposition occurred, giving several fragments that is likely to be biologically inactive.

III.3 Summary of synthetic outcomes

Ring opening of epoxide **40** with aniline **41** only gave 7% isolated yield of the target aminol **33**. Several other products were formed, such as ethylated aniline **44**, ethoxylated compound **43**, and ethylated product **45**. An alternative synthesis from epoxide **40** through ring opening with sodium azide followed by Staudinger reduction gave the desired aminol **33** in a satisfactory 80% yield (Scheme **38**). Similar side reactions were detected in the synthesis of aminol **34**. Using epoxide aminolysis, four analogues of the flubenzurons containing the 4-nitrophenyl group was synthesised.

A Sandmeyer reaction was attempted in order to obtain aryl iodide **67**. A few conditions gave the compoud, but the reaction was not reproducable and sufficient amounts of the product was not achieved. The aryl bromide was only isolated in 6-14% yields and scaling up the reaction resulted in trace amounts of the product. A Suzuki-Miyaura cross-coupling reaction with triflate **70** did not give any of the allylated product **71**. Nitration of aryl bromide **52** gave the *meta*-nitrated product with complete regioselectivity. A Stille cross coupling followed by epoxidation yielded epoxide **77** (Scheme **38**), which was used in ring-opening reactions with anilines **49** and **58-60** to give aminols **77-80**.



Scheme 38. Summary of synthetic outcomes in Part III.

III.4 Concluding remarks

In conclusion, Part III presents the successful synthesis of twelve target compounds based on the photodegradable scaffolds described in Part II. Four of these compounds, **63**, **78**, **79**, and **80**, displayed promising antimicrobial activity (described in Part IV) and were therefore selected for photodegradation studies at pH 13 and 8. Delightfully, **78** and **80** was converted completely at pH 8, and **63** and **79** displayed conversions of 25 and 19%, respectively. At pH 13, **63**, **78**, and **79** all degraded completely, however, stability studies revealed that only the former of these three compounds were stable under these conditions. At pH 8, all compounds were completely stable. The degradation patterns for all the compounds except **80** were investigated in detail, and for **63**, the photo-retro-aldol reaction described in Part II was the dominating process, and aniline **60** and *p*-nitrobenzoic acid (**81**) was formed. For **78** and **79**, a nucleophilic aromatic substitution reaction occurs, yielding tetrahydroquinolines **83** and **85**, and these products were confirmed by ^{1}H and ^{19}F NMR and LRMS. The photodegradation of **80** proceeds through a different mechanism, which at this point was not investigated further.

Part IV

Biological studies

With the flubenzuron analogues in hand, they were subjected to biological evaluation studies. Initially, they were designed to act as photodegradable chitin synthase inhibitors, but since bioactive compounds may possess several properties, it was envisioned that the flubenzuron analogues could also display antimicrobial activity. Therefore, the biological studies include growth inhibition studies against *L. salmonis*, a screening of the minimum inhibitory concentration against a range of Gram-positive and Gram-negative bacteria, and a cycotoxic evaluation.

IV.1 Growth inhibition of *L. salmonis*

The nauplius 1 stage of the parasite *L. salmonis* was used for a live-dead assay in order to evaluate the ability of the benzoyl urea analogues as anti-lice drugs. Around 10-30 individuals were exposed for one hour at 10 °C at concentrations 1.0, 0.1, and 0.001 ppt (parts per thousand), and was subsequently transferred to incubators with a constant flow of seawater at 9 °C. After 7 days the number of surviving nauplii or copepodids were counted as well as a subjective evaluation of their condition. If the compounds have no effect on the nauplii, they will develop naturally into copepodids through ecdysis,

i.e. shedding their chitin-rich exoskeleton, or exuvium. An active compound, such as diflubenzuron, would prevent formation of the exoskeleton when nauplii develop, resulting in observation of dead nauplii and no copepodids. The life cycle of *L. salmonis* has a total of six main stages until it reaches adulthood, with repeating ecdysis between each stage. A simplified illustration shows this development, as well as the target for chitin synthase inhibition (Figure **24**).[106]

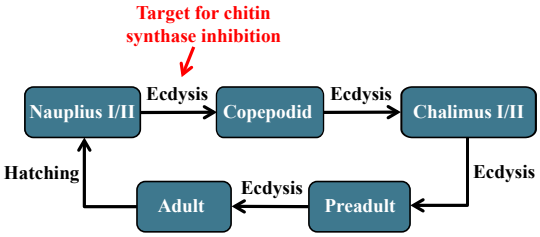


Figure 24. Simplified illustration of the life stages of *L. salmonis*.[106]

An initial screening was done with compounds **33** and **34**, and photodegraded mixtures **33d** and **34d**, at a concentration of 1.0 ppt. For **33**, the nauplii had developed into active copepodids and exuvia was observed, indicating no growth inhibition activity (Table **5**, entry 1). However, for **34** limp nauplii and no copepodids nor exuvia was observed, which means that growh inhibition is taking place (Table **5**, entry 3). For **34d**, a few copepodids were seen, along with limp nauplii, which indicates that post degradation, the activity decreases. The limp nauplii could also be caused by toxic effects from **34** or toxic byproducts from the photoreaction, since the concentration of 1.0 ppt is quite high. Since DMSO was used to

help solubilise the compounds, it is included as a reference, and at this concentration it displays no activity (Table **5**, entry 5). Further studies to validate the initial results are currently ongoing at the Sea Lice Research Centre at the University of Bergen.

Table 5. Initial screening of growth inhibition activity against *L. salmonis* at 1.0 ppt concentration.

Entry	Compound	Parallell 1	Parallell 2	Parallell 3	Comment
1	**33**	Cop.	Cop.	Cop.[a]	Exuvia
2	**33d**	Nauplii	Nauplii	Nauplii	Limp, no exuvia
3	**34**	Nauplii	Nauplii	Nauplii	Limp, no exuvia
4	**34d**	1 cop.	2 cop.	Nauplii	Limp, no exuvia
5	DMSO	Cop.	Cop.	Cop.	Exuvia

[a] A few nauplii seemed to be stuck in ecdysis.

A secondary screening of compounds **66**, **63**, **78**, **79**, and **80** gave promising results. At 1 ppt, all of the compounds were active except **63**, since mainly dead nauplii and no copepodids were seen (Table **6**, entries 1-5). This means that the nauplii were not able to develop naturally into copepodids through ecdysis. At 0.1 ppt, compound **80** is no longer active, since most of the nauplii have developed into copepodids with normal behaviour (Table **6**, entry 5), but **78** and **79** continues to effectively inhibit growth as there are no individuals at the copepodid stage (Table **6**, entries 3 and 4). Compound **66** is starting to lose its growth inhibition potential since a few copepodids are seen (Table **6**, entry 1), However, at 0.001 ppt most of the nauplii have developed into copepodids, which means that growth inhibition is no longer taking place. These results indicate that compound **78**

and **79** near rivals the activity of diflubenzuron (Table **6**, entry 6). Further studies on the degradation mixtures of these compounds are currently ongoing at the Sea Lice Research Centre at the University of Bergen, and initial results indicate that activity is decreasing post decomposition. Additionally, nauplii were exposed to compound **78** and **79** at 0.01 ppt, revealing that the former is no longer active at this concentration, since most of the nauplii developed into copepodids. For compound **79**, it seems that LC_{50} is around 0.01 ppt. However, in this screening diflubenzuron had no effect on the developing nauplii, and healthy copepodids were observed in all parallels at concentrations from 0.01-0.0001 ppt. Since activity was clearly seen in the previous screening (Table **6**, entry 6), this indicates a narrow time window in the nauplius life stage where efficient uptake of diflubenzuron occurs. When treating infestations, this drug is administered in feed to salmon, and lice are in turn exposed orally by feeding on their blood. This might not be easily transferable to bath treatment, explaining the varying observations.

Table 6. Secondary screening of growth inhibition activity against *L. salmonis*.

Entry	Compound	Conc. (ppt)	Parallel 1		Parallel 2	
			Naup.[a]	Cop.[b]	Naup.[a]	Cop.[b]
1	**66**	1	13	0	8	0
		0.1	11	4	28	2
		0.001	16	23	8	21
2	**63**	1	6	19	15	14
		0.1	12	17	1	10
		0.001	9	8[c]	6	25[c]
3	**78**	1	8	0	33	0
		0.1	22	0	37	0
		0.001	2	28[c]	5	18[c]
4	**79**	1	24	0	49	0
		0.1	28	0	18	0
		0.001	2	20[c]	1	18[c]
5	**80**	1	13	0	10	1
		0.1	6	13[c]	6	14
		0.001	13	22[c]	5	15[c]
6	Diflubenzuron	1	18	0	23	0
		0.1	9	0	15	1
		0.001	16	14	10	3
7	DMSO	1	5	20[c]	11	6[c]
		0.1	1	17[c]	8	6[c]
		0.001	1	27[c]	22	16[c]

[a] All nauplii observed were dead.
[b] Unless otherwise noted, all copepodids were either limp or paralysed.
[c] Copepodites with normal behaviour.

IV.2 Antibiotic acitivity and cytotoxicity

The minimum inhibitory concentration (MIC) of all the target compounds were screened against Gram-positive bacteria *S. aureus*, *S. epidermidis*, *E. faecalis*, and *S. agalactiae*, and Gram-negative bacteria *E. coli* and *P. aeruginosa*. Optical density at 600 nm (OD_{600})

was measured to determine the growth of bacteria in compound solutions ranging from 1.6-100 μM after incubation overnight at 37 °C. Experimental details of the testing procedure can be found in supplementary information in paper I.[107] The majority of the compounds displayed no bacterial growth inhibition, and are indicated with *I* (Figure **25**). The best results were obtained from the assays of compound **63** and **80** with 6.3 μM activity selectively against *S. agalactiae*, closely followed by **78** with 50 μM activity against *S. epidermidis*, *S. aureus*, and *S. agalactiae*, and lastly **79** with 50 μM activity selectively against *S. aureus*. The cytotoxic properties of the four active compounds were studied in concentrations from 1.6-100 μM using HepG2 (human liver cancer cell line) and MRC5 (human lung fibroplasts), and was found to be 25-50 and 50-75 μM, respectively (Figure **25**).

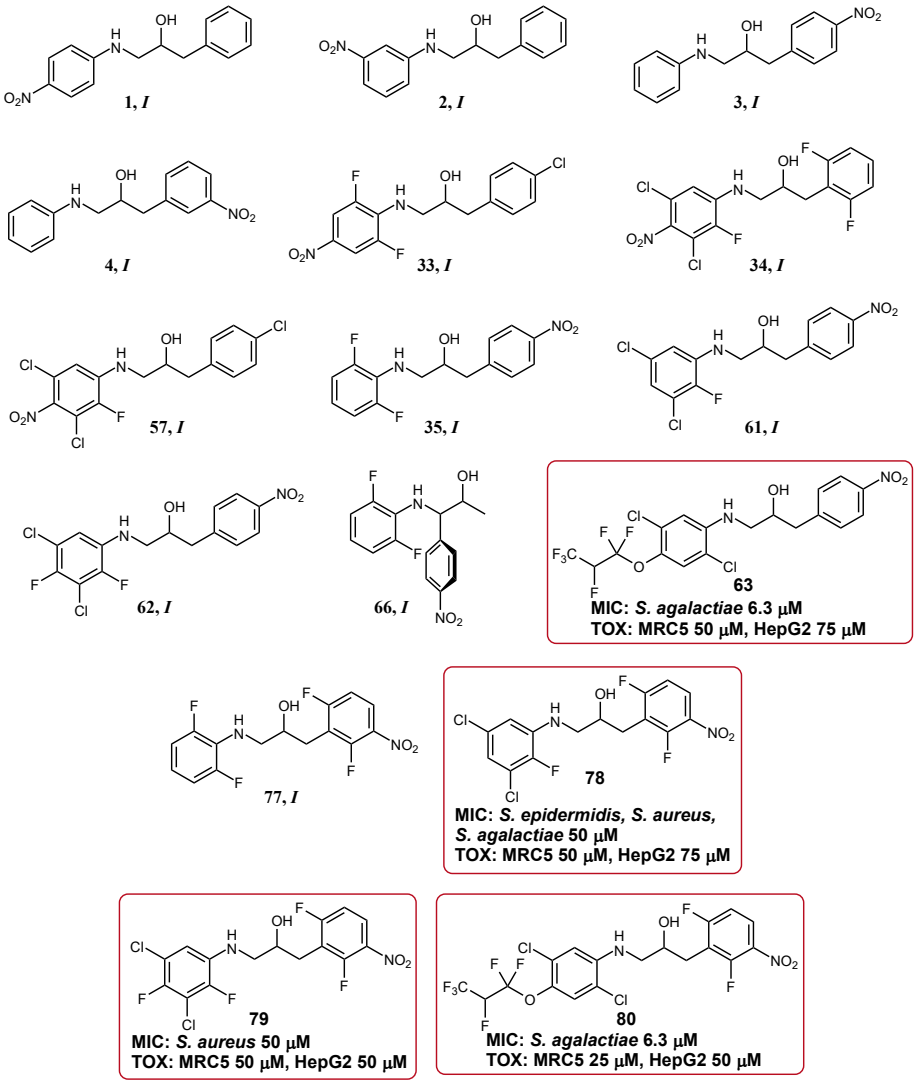


Figure 25. Minimum inhibitory concentration (MIC) and cytotoxicity (TOX) results for all target compounds. I = Inactive.

MIC of the crude degradation mixtures were screened against the same bacteria as in the previous assays, and no antibiotic activity was observed. Additionally, the cytotoxic properties against HepG2

and MRC5 cells were studied for the degradation products of the four active compounds, displaying no toxicity.

IV.3 Concluding remarks

For compounds **78** and **79**, promising activities against *L. salmonis* (0.1-0.001 ppt) were observed that near rivals the activity of the known anti-lice drug diflubenzuron. Both **66** and **80** were active at higher concentrations (1-0.1 ppt). Further studies on the degradation mixtures are currently ongoing, and initial results indicate that activity is decreasing post decomposition.

In the antimicrobial assay, the two most active compounds were **63** and **80**, displaying MIC of 6.3 μM, and **78** and **79** were active at the 50 μM level. All the compounds were moderately toxic against MRC5 and HepG2 cells at 25-75 μM. Delightfully, the photodegraded mixtures did not show any bacteria growth inhibition and no toxicity whatsoever. This means that some of these compounds are potent antimicrobial and anti-lice agents that degrades under UV light into inactive and non-toxic fragments, and they represent lead compounds for further development.

Part V

Chloramphenicol analogues

Out of all the antibiotics that have been detected in different parts of the biosphere in recent years, such as ciprofloxacin, sulfamethoxazole, amoxicillin, and chloramphenicol,[8–14] the latter of these is particularly interesting for designing some potentially active photodegradable analogues. This is due to the presence of the nitrophenyl group, which structurally resembles the model compounds described in Part II. To this end, two sets of analogues containing the structural elements of chloramphenicol while simultaneously maintaining the photodegradable properties were designed (Figure **26**). Once the compounds are obtained, their biological activity will be evaluated and, if active, their photodegradation will be investigated.

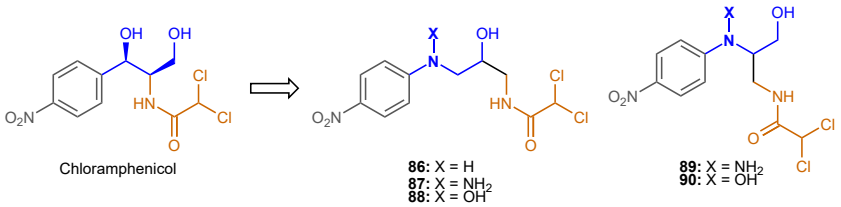
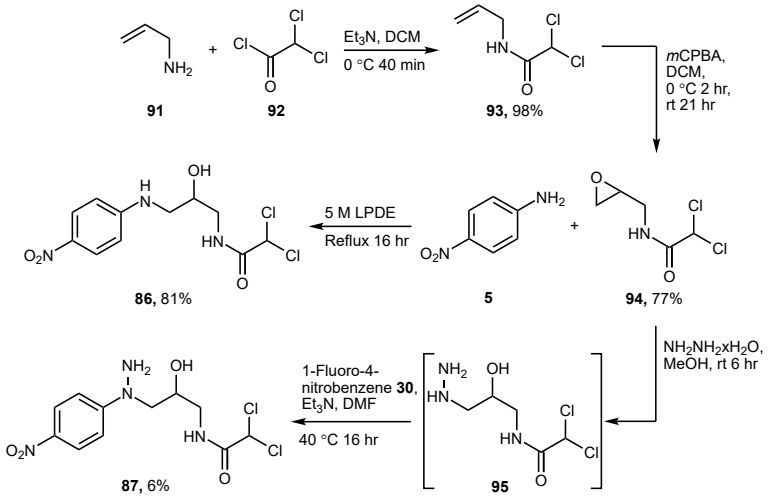


Figure 26. Two sets of analogues of chloramphenicol. X: H, OH, NH_2.

V.1 Synthesis of chloramphenicol analogues

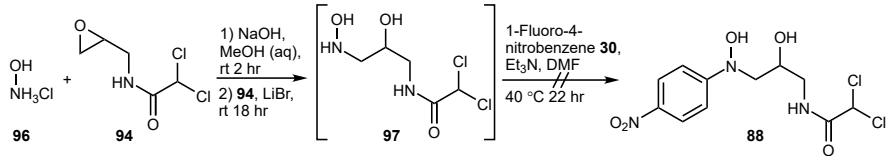
A simplified analogue of chloramphenicol was easily synthesised through three steps. Acylation with dichloroacetyl chloride, followed by epoxidation with *m*CPBA and subsequent Lewis acid-promoted epoxide ring opening with *p*-nitroaniline (**5**) yielded the target compound **86** in 61% overall yield (Scheme **39**). Additionally, epoxide **94** was opened with hydrazine hydrate, followed by a nucleophilic aromatic substitution reaction with 1-fluoro-4-nitrobenzene (**30**) to yield hydrazineyl **87** in a disappointingly low 6% yield. The low yield is due to an extremely difficult purification which required two consecutive attempts at column chromatography followed by two recrystallisations.



Scheme 39. Synthesis of chloramphenicol analogues **86** and **87**.

A similar approach was attempted in order to synthesise hydroxylamine analogue **88**. A methanolic solution of hydroxylamine was

prepared from hydroxylammonium chloride (**96**), which was then reacted with epoxide **94**. The resulting intermediate was used directly in a nucleophilic aromatic substitution with 1-fluoro-4-nitrobenzene (**30**), but the desired product was not detected in the reaction mixture (Scheme **40**).



Scheme 40. Attempted synthesis of chloramphenicol analogue **88**.

Preliminary analysis based on TLC revealed a complex reaction mixture with at least eight products. A mass spectrum extracted from one of the spots on the TLC plate suggests formation of *p*-nitrophenol (**98**), and bis-aryl compounds **99** and **100**, indicated by the detected m/z values 137.99, 273.97, and 457.03 (Figure **27**). There is also a possibility that the detection of *p*-nitrophenol (**98**) is the result of a fragmentation of the bis-aryl compounds, but this was not investigated further.

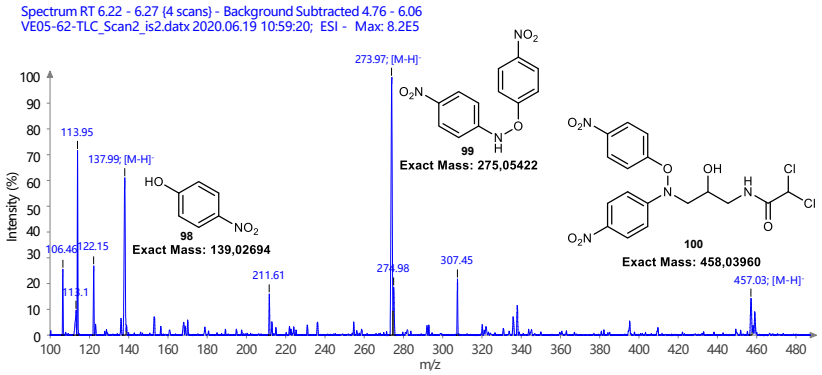
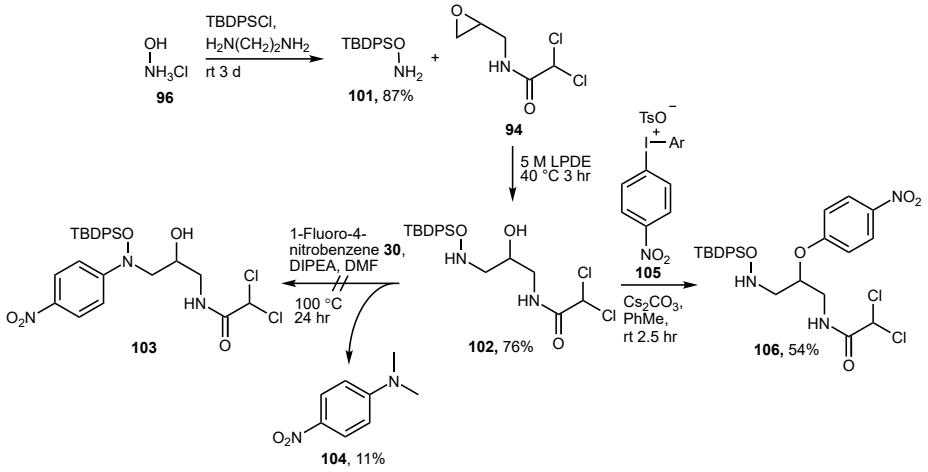


Figure 27. Mass spectrum extracted from the reaction mixture of the attempted synthesis of compound **88**.

To avoid any arylation taking place on the oxygen, hydroxylamine was protected as the *tert*-butyldiphenylsilyl (TBDPS) ether **101**.[108] Subsequent epoxide ring opening with **94** was uneventful and yielded compound **102** in 76% yield, but when attempting a nucleophilic aromatic substitution reaction with 1-fluoro-4-nitrobenzene (**30**), the target product was not detected (Scheme **41**). According to TLC-MS, unreacted starting materials were still present, and *N,N*-dimethyl-4-nitroaniline (**104**) was confirmed by NMR. This product is the result of base-assisted decomposition of DMF to form dimethylamine, which in turn reacts with 1-fluoro-4-nitrobenzene (**30**) to yield compound **104**. This reaction is known to occur with other bases, such as KOH, NaOH, KO^{t}Bu, and K_3PO_4.[109,110]



Scheme 41. Attempted synthesis of chloramphenicol analogue **88** through TBDPS-protected intermediate **103**.

Another attempt at arylating the hydroxylamine was done with diaryliodonium salt **105**, prepared according to a procedure by Guérard,[111] using cesium carbonate in anhydrous toluene, as described by Wang and coworkers for the arylation of silyl-protected hydroxylamine derivatives.[112] However, there was no reaction on the nitrogen, and arylation took place on the alcohol instead, yielding compound **106** in 54% yield, which could easily be distinguished from the target isomer by the presence of two NH signals displaying COSY cross peaks to the methylene protons (Figure **28**). Protecting the alcohol as the *tert*-butyldimethylsilyl (TBS) ether and then carrying out the arylation also did not yield any traces of the target product. After these unsuccessful attempts at synthesising compound **88**, it was decided to discard it and to carry on with the other more closely related analogues **89** and **90**.

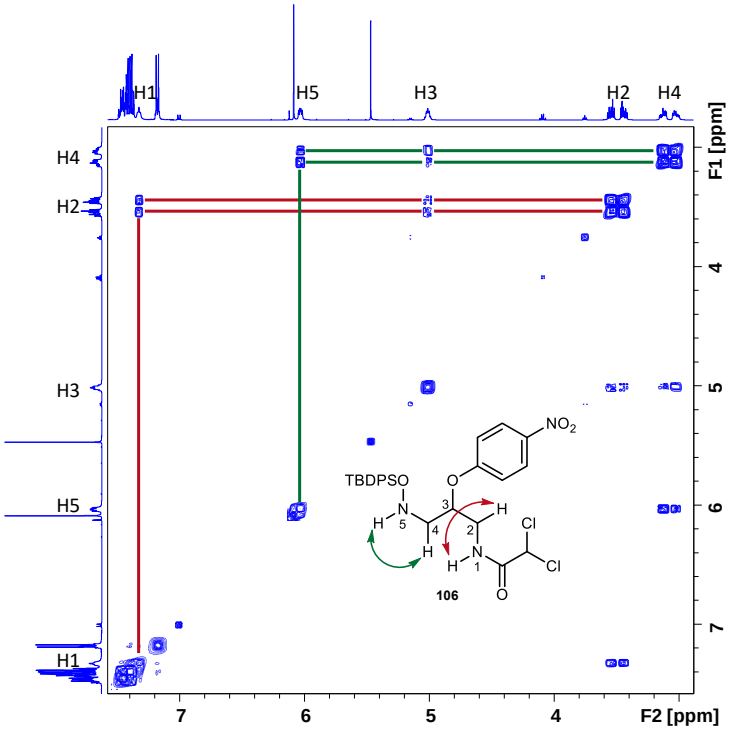
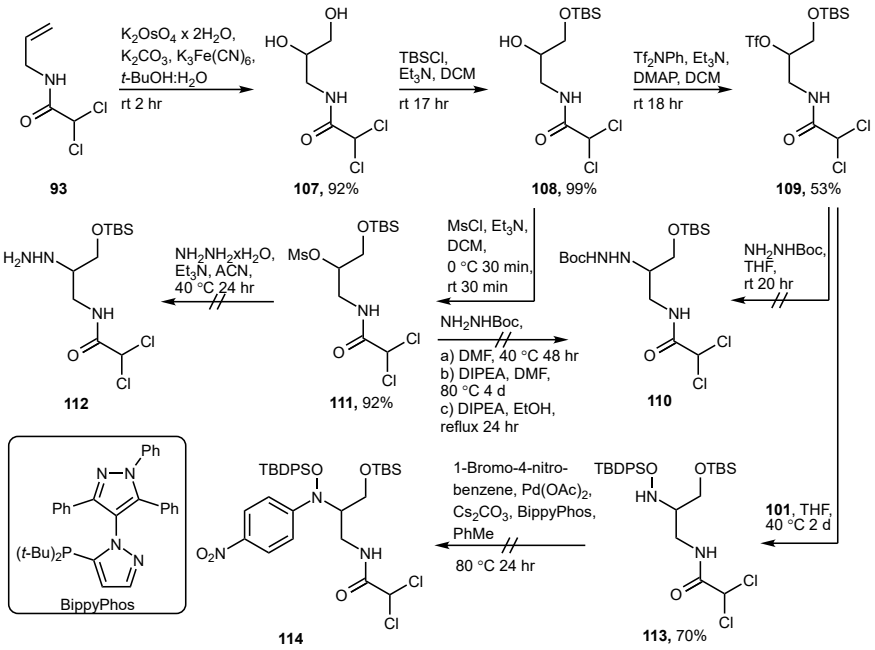


Figure 28. Selected COSY correlations confirming formation of compound **106**.

For hydrazine analogue **89**, the synthetic pathway started with allylamide **93**. Osmium-catalysed dihydroxylation with potassium ferricyanide as cooxidant,[113] followed by selective primary alcohol protection with TBS chloride yielded silyl ether **108** in excellent yield. The alcohol was triflated with *N*-phenyl-bis(trifluoromethanesulfonimide), but a nucleophilic substitution with *tert*-butyl carbazate did not yield compound **110** (Scheme **42**). Surprisingly, alcohol **108** was isolated in 15% yield from the reaction mixture. Mesylate **111** was used instead of the triflate, and reaction with hydrazine hydrate yielded an immensely complicated reaction mix-

ture with at least nine new products. This synthetic pathway was therefore discarded and *tert*-butyl carbazate was used instead, but after several attempts with various reaction conditions, mostly unreacted starting material remained. Traces of the target product **110** was detected by TLC-MS (Exact mass 429.16, detected m/z 428.11 $[M\text{-}H]^-$), but complete overlap with starting material on TLC did not allow for separation by column chromatography. Additionally, an unidentified compound with no chlorine atoms, indicated by the lack of isotopic distribution in the mass spectrum (Figure **29**), but containing the appropriate signals from both the Boc and the TBS group in NMR, was observed. This indicates that there are unwanted intramolecular reactions taking place, which greatly complicates the synthesis of **89**. The hydrazine analogue was therefore discarded, and focus was shifted to hydroxylamine **90** instead.



Scheme 42. Synthetic approach towards the chloramphenicol analogues **89** and **90**.

Nucleophilic substitution with triflate **109** and TBDPS-protected hydroxylamine **101** gave compound **113** in 70% yield (Scheme **42**). After the unsuccessful attempts at arylating **102** with nucleophilic aromatic substitution or by using the diaryliodonium salt, a new strategy involving palladium-catalysed arylation with 1-bromo-4-nitrobenzene was attempted, as described by Tomkinson and co-workers.[114] In their work, *N*-Boc-*O*-silyl hydroxylamines were coupled to a range of aryl halides using $Pd(OAc)_2$, Cs_2CO_3, and BippyPhos in toluene at 80 °C. Interestingly, out of 11 different phosphine ligands they tested, only BippyPhos yielded the *N*-arylated product. However, when those reaction conditions were applied to arylate hy-

droxylamine **113**, only traces of the target product was detected by TLC-MS. Most of the starting material remained unreacted, but this was not investigated further.

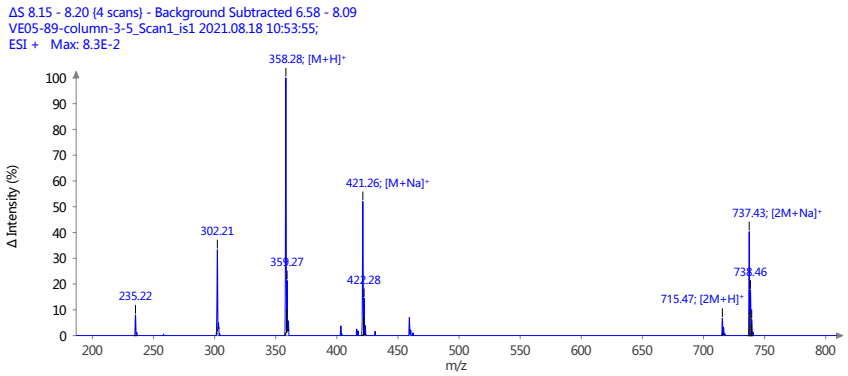


Figure 29. Mass spectrum of the unidentified compound.

Following these results, it was decided to continue with the biological evaluation of the two compounds that were successfully synthesised, **86** and **87**. The antimicrobial activity was screened against the same bacterial cultures and at identical concentration levels as for the results introduced in Part IV. Unfortunately, no antibiotic activity was observed (Figure **30**), and therefore the cytotoxicity and photodegradation studies of these compounds were not investigated, with the exception of an initial degradation experiment of **86** at pH 11, which resulted in complete degradation and formation of *p*-nitroaniline.

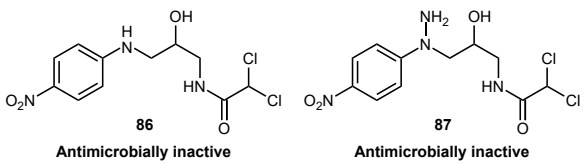
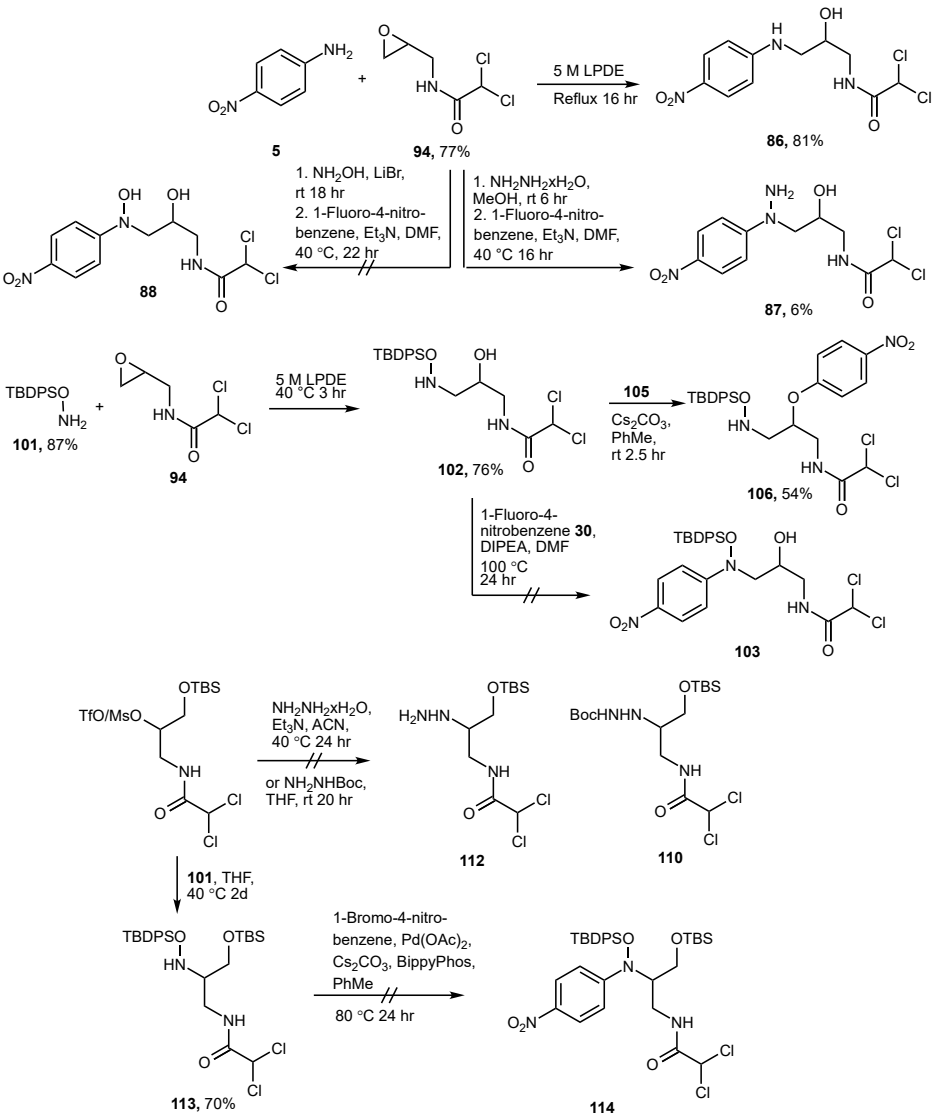


Figure 30. Chloramphenicol analogues **86** and **87**.

V.2 Summary of synthetic outcomes

The simplest analogue, compound **86**, was readily formed in 61% overall yield in a simple three-step procedure. However, from this point on the synthetic procedures proved to be challenging and different side reactions were dominating. Hydrazine **87** was isolated in a disappointing 6% yield after a challenging purification. When attempting to synthesise hydroxylamine analogue **88** by a similar strategy to the one used for **87**, overarylation was a problem and the reaction mixture was complex with at least eight products. Protecting the hydroxylamine as the TBDPS ether, compound **102**, resulted in an unreactive nitrogen, and arylation took place on the alcohol instead, which formed **106**. Reaction of mesylate **111** with hydrazine hydrate or *tert*-butyl carbazate did not give the desired products **112** and **110**. However, triflate **109** reacted with TBDPS-protected hydroxylamine in a nucleophilic substitution reaction to form compound **113**, but the subsequent arylation was not successful (Scheme **43**).



Scheme 43. Summary of synthetic outcomes in Part V.

V.3 Concluding remarks

Part V presents the design and synthesis of analogues of the well-known antibiotic chloramphenicol. These endeavors ended up in the

successful synthesis of two analogues of chloramphenicol, compounds **86** and **87**, but unfortunately no antimicrobial activity was observed. The former of these compounds was subjected to photodegradation and complete decomposition at pH 11 was observed.

Part VI

Towards a photodegradable penicillin

Inspired by the photodegradation of a cephalosporanic acid antibiotic presented by Mobashery and co-workers,[40] a photodegradable penicillin was envisioned. A similar strategy to the one used for the chloramphenicol analogues, discussed in Part V, was applied in order to design a photodegradable analogue of the penicillin class of antibiotics. Exchanging the phenylacetic moiety of penicillin G with part of the photodegradable scaffold gives the suggested compound **115**, which could possibly act as an active antibiotic agent that degrades under light.

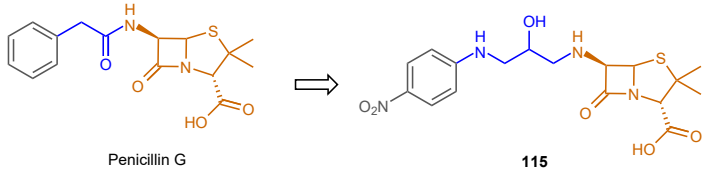
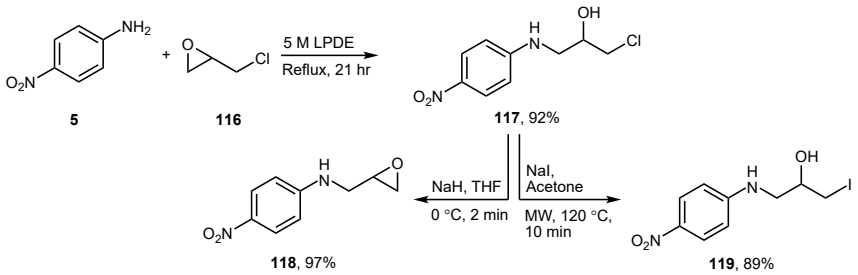


Figure 31. A photodegradable analogue of penicillin G.

VI.1 Synthesis of the penicillin analogue

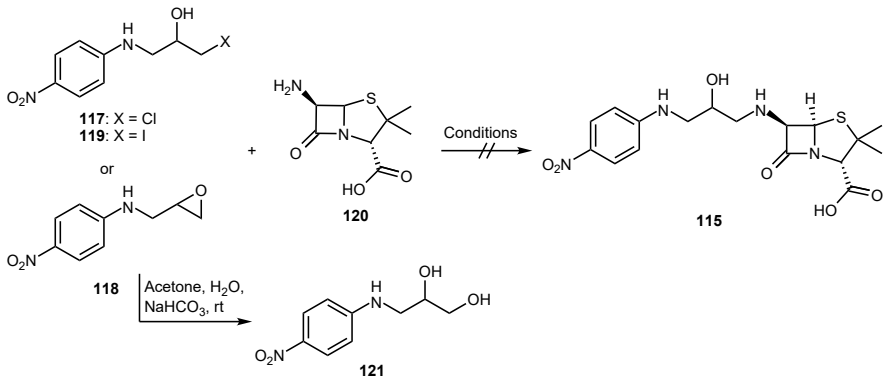
In order to synthesise penicillin analogue **115**, a potent electrophile was needed to react with 6-aminopenicillanic acid (6-APA, **120**). Stereochemistry was not a concern at this point, which is why a reasonably priced racemic mixture of epichlorohydrin (**116**) was used. The epoxide was easily opened with *p*-nitroaniline (**5**) in 5 M LPDE

to yield chlorohydrin **117**, and subsequent treatment with NaH in THF yielded epoxide **118** in 89% overall yield. Iodohydrin **119** was obtained by performing a microwave-assisted Finckelstein reaction with sodium iodide in acetone in 82% total yield (Scheme **44**).



Scheme 44. Synthesis of epoxide **118** and iodohydrin **119**.

It was then attempted to synthesise penicillin **115** in either an alkylation or epoxide ring-opening reaction with 6-APA (**120**) (Scheme **45**). This would give a highly efficient protecting group-free synthesis of a possible antibiotic agent that should decompose when exposed to light. However, preliminary experiments revealed that the amino group in 6-APA did not react at all at rt and due to stability issues with the β-lactam, the temperature could not be increased much higher.



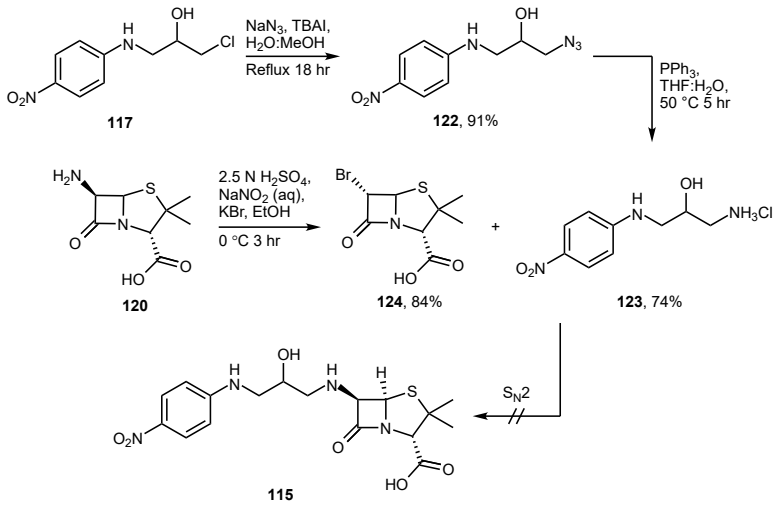
Scheme 45. Attempted synthesis of β-lactam **115** by an S_N2 or an epoxide ring-opening reaction.

In most experiments with iodohydrin **119** the alkaline conditions simply gave epoxide **118** (Table **7**, entry 2, 4, 5, and 7), which was completely stable under anhydrous conditions. However, when water was present as solvent, the corresponding diol **121**, arising from ring opening with water, was detected (Table **7**, entry 5). 5 M LPDE conditions were attempted, but no product formation was observed (Table **7**, entry 6). A range of different bases and additives were attempted, such as sodium iodide, copper(II) triflate, and silver nitrate, but the desired product was never seen in any of the experiments (Table **7**).

Table 7. Conditions for the attempted S_N2-/epoxide ring-opening reaction to form β-lactam **115**.

Entry	Substrate	Reagent	Base	Solvent	Temp. (°C)	Time	Product
1	117	NaI	Cs_2CO_3	EtOH	rt - 70	4 d	-
2	119	-	Et_3N	DMF	rt	8 d	118
3	118	4 Å MS, $Cu(OTF)_2$	CsOH	DMF	rt	4 d	-
4	118	$AgNO_3$	Et_3N	DMF	rt	3 d	118
5	119	-	$NaHCO_3$	Acetone, H_2O	rt	2 d	118, 121
6	118	5 M LPDE	-	Et_2O	rt - 60	2 d	-
7	119	$AgNO_3$	Et_3N	ACN	rt	1 hr	118

A possible explanation is that the amino group in 6-APA (**120**) simply is not nucleophilic enough, due to inductive effect from the adjacent carbonyl group, and the epoxide/iodohydrin is not sufficiently electrophilic to react under these mild conditions. It was then decided to reverse the reactivity, and in doing so taking advantage of the deactivating effect from the carbonyl. An azide was introduced in chlorohydrin **117** to form **122**, followed by a Staudinger reduction to yield amine **123** in 67% yield over two steps (Scheme **46**). The amino group in 6-APA (**120**) was easily exchanged with a bromide in a Sandmeyer-type reaction to give β-lactam **124** with complete inversion of stereochemistry. A nucleophilic substitution was attempted, but sadly the desired product was once again not achieved (Scheme **46**).



Scheme 46. Attempted synthesis of β-lactam **115** by reversing electrophile and nucleophile.

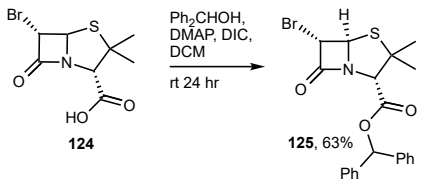
A few different conditions were screened (Table **8**), but none gave any formation of penicillin **115**. Since the substitution occurs under basic conditions, the carboxylic acid is immediately deprotonated in the reaction mixture. Nucleophilic substitution on a negatively charged species is, of course, unlikely to happen, but before introducing protecting groups to prevent this from happening, a few experiments had to be tested in case protection was unnecessary.

Table 8. Conditions for the attempted S_N2 reaction to form β-lactam **115** with reversed nucleophile/electrophile.

Entry	Reagent	Base	Solvent	Temp. (°C)	Time
1	-	K_2CO_3	DMF	rt	2 d
2	-	Cs_2CO_3	DMF	40	2 d
3	18-Crown-6[a]	K_2CO_3	ACN	40	8 hr
4	4 Å MS	CsOH	DMF	rt	24 hr

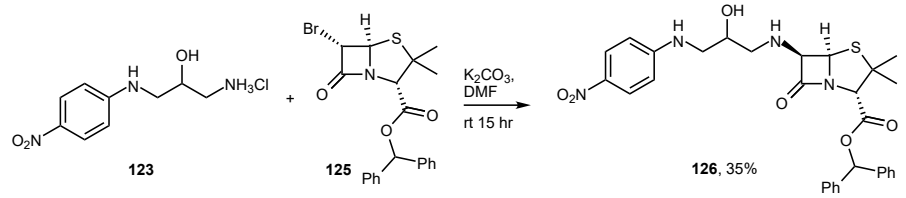
[a] 18-Crown-6 was added after 5 hr.

In order to prevent the β-lactam from hydrolysing, a carboxylic protecting group that could be removed without the use of strong acid or base at elevated temperatures was needed. The diphenylmethyl ester has previously been applied on other β-lactam compounds and removal occurs under mild heating in phenol or *m*-cresol with or without addition of a catalytic amount of acid, such as TFA or *p*-TSA.[40,115] The protecting group was easily introduced under Steglich conditions,[116] with diphenylmethanol, diisopropylcarbodiimide (DIC), and DMAP to yield ester **125** (Scheme **47**). DIC was used instead of the traditional dicyclohexylcarbodiimide (DCC), since the resulting dialkylurea byproduct is significantly easier to remove from the reaction mixture by simply washing the organic layer with water.



Scheme 47. Protection of 6-bromopenicillanic acid **124** as diphenylmethyl ester **125**.

The substitution reaction with protected β-lactam **125** did indeed result in the desired product **126** (Scheme **48**), which was isolated as a pair of diastereomers after four consecutive purifications by autoflash column chromatography. This explains the modest yield of 35%, as overlapping compounds were a major problem.



Scheme 48. Synthesis of β-lactam **126** by nucleophilic substitution.

The two carbonyl groups were easily identified by their correlations in ^{1}H-^{13}C-HMBC NMR (Figure **32**). Couplings shown in **red** (H6 → C3) and **green** (H4 → C3) confirms formation of the desired compound. Remaining signals were identified in combination with ^{1}H-^{13}C-HSQC NMR. H4 has an unusually high chemical shift value (δ 7.59 ppm) compared to the corresponding signal in the ^{1}H-NMR spectrum of compound **125** (δ 4.82 ppm), probably due to its proximity to the deshielding zone of one or more aromatic rings.

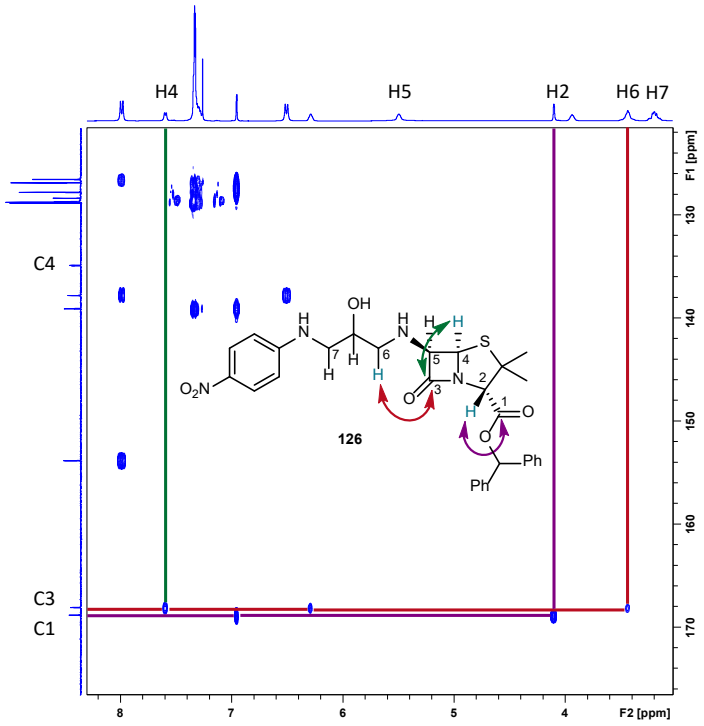
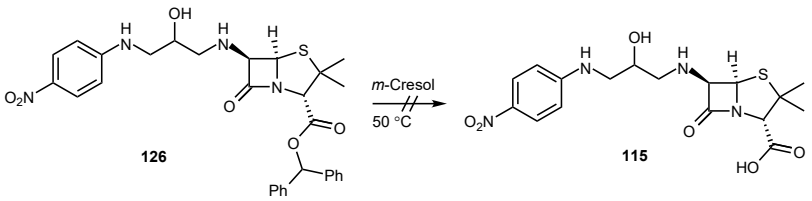


Figure 32. ^{1}H-^{13}C-HMBC cross peaks confirming formation of β-lactam **126**.

With compound **126** in hand, a simple deprotection would yield the desired β-lactam **115**. Following the procedure described by Torii and coworkers,[115] deprotection of β-lactam **126** was attempted in *m*-cresol at 50 °C (Scheme **49**). The reaction progressed extremely slowly as indicated by TLC, and thermal decomposition products were a major concern. In order to speed up the reaction, TFA was added, and after three hours the starting material was completely consumed.



Scheme 49. Attempted deprotection of the diphenylmethyl ester in β-lactam **126**.

During aqueous workup, a small amount of a yellow precipitated solid was obtained and analysed by NMR. At first glance, it seemed that the desired product was finally successfully synthesised. However, by taking a closer look at the ^{1}H-NMR spectrum, it became clear that this was not the case (Figure **33**). The two doublets at δ 6-8 ppm corresponding to the aromatic protons in the *p*-nitroaniline moiety is present. So are all the signals from the alkyl chain around δ 3-4 ppm, however two of the signals are overlapping with the solvent residual signal for methanol. When comparing the integrals of these signals with the singlets at δ 7.58 ppm, 3.98 ppm, 1.52 ppm, and 1.27 ppm, all of which belong to the β-lactam part of the molecule, it was obvious that the reaction had been unsuccessful. The integrals of the singlets are simply too low, meaning that the spectrum displays two separate compounds, instead of the target molecule **115**. The reaction was repeated many times with lower temperatures, different amounts of TFA, and phenol instead of *m*-cresol, but the desired product was never detected.

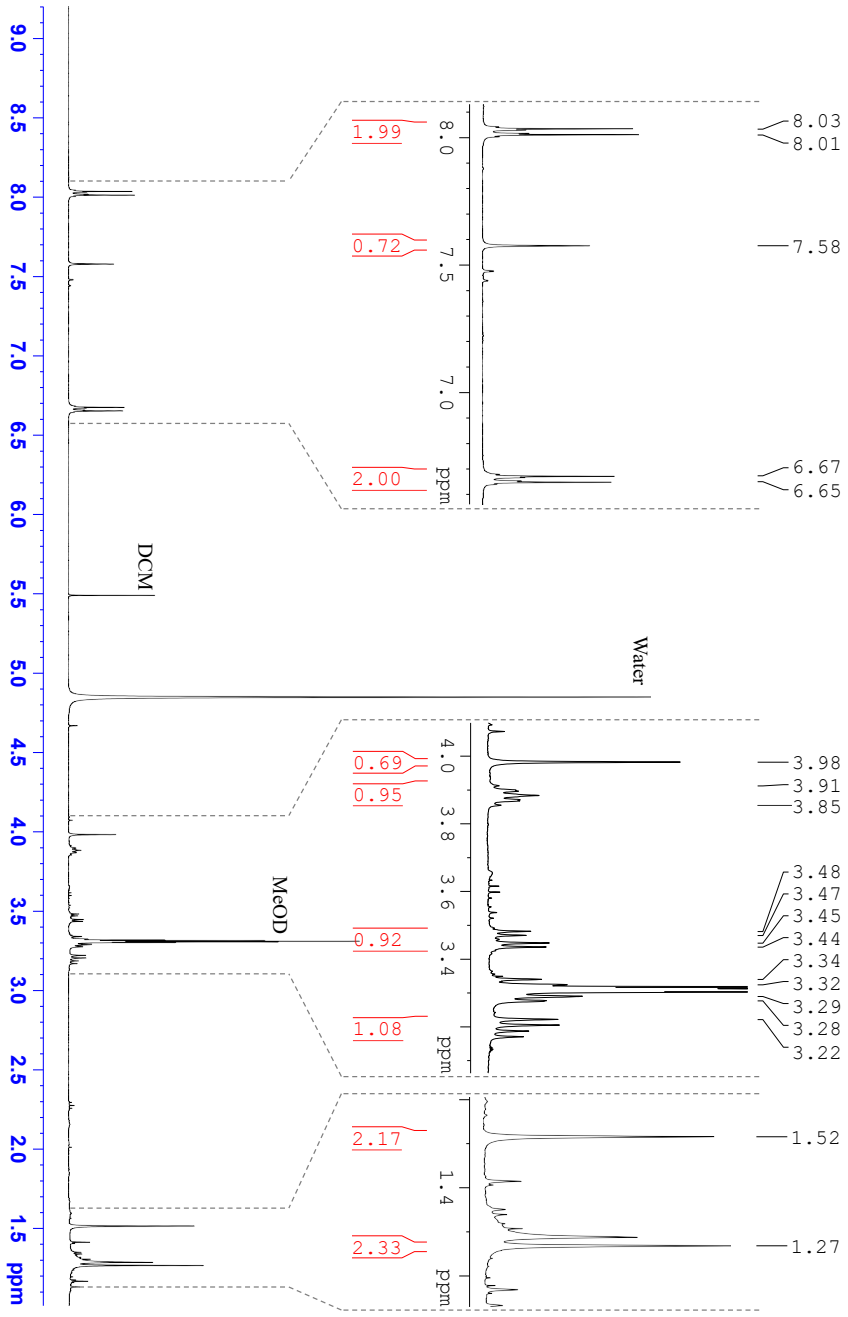
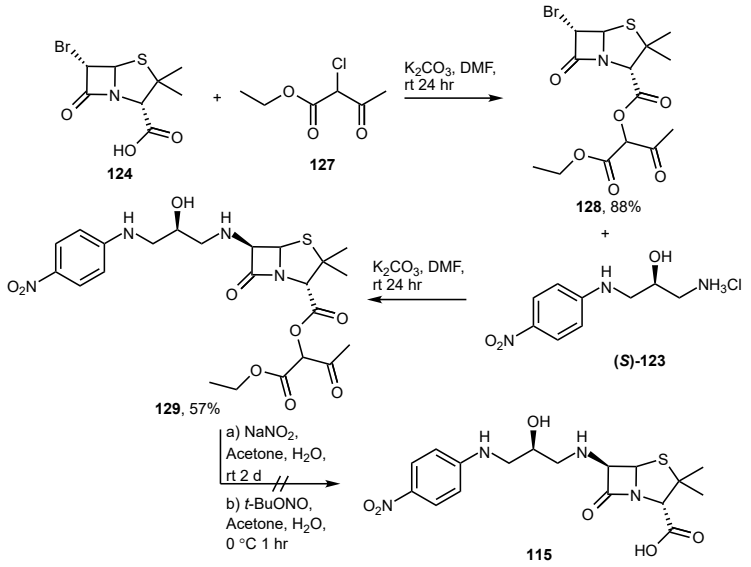


Figure 33. ^{1}H-NMR spectrum of the yellow precipitate from the deprotection of compound **126**.

After the unsuccessful attemps at deprotecting compound **126**, it was decided to apply a different protecting group. A dicarbonyl ester has previously been used to protect and deprotect penicillin G without harming the β-lactam ring.[117] Cleavage occurs with sodium nitrite in aqueous acetone or with alkyl nitrites. To this end, penicillanic acid **124** was protected using ethyl 2-chloroacetoacetate (**127**) with potassium carbonate in DMF to form compound **128** in 88% yield (Scheme **50**), which initially was reacted with the racemic mixture of aminol **123**. However, the purification and ^{1}H-NMR spectrum of the product was very complicated because of the presence of four diastereomers. Therefore, optically pure **(*S*)-123** was prepared using the same procedure as for the racemate, with the exception of using (*R*)-(-)-epichlorohydrin instead of the racemic mixture. A nucleophilic substitution reaction with penicillinate **128** gave the desired product **129** as a pair of diastereomers in 57% yield (Scheme **50**), which was easier to identify by NMR. Deprotection with sodium nitrite in acetone and water did not give any noticable reaction, even after two days. The reaction was repeated with *tert*-butyl nitrite at 0 °C, and after 1 hr, there were at least seven new products according to TLC analysis. A few attempts at column chromatography did not give any identifiable products. A myriad of unknown and overlapping signals in the ^{1}H-NMR spectra made it clear that the reaction was unsuccessful. Analysis of the crude reaction mixture

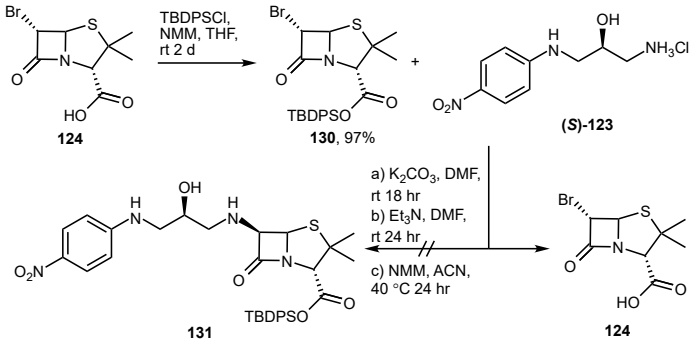
with TLC-MS also did not give any signals that could be attributed to the desired product.



Scheme 50. Attempted synthesis of β-lactam **115** through dicarbonyl-protected compound **129**.

This led to using a third and less stable protecting group in the hopes that removal could be done under extremely mild conditions. The TBDPS group, more commonly used for the protection of alcohols, was introduced uneventfully in penicillanic acid **124** with TBDPSCl and *N*-methylmorpholine (NMM) in anhydrous THF to yield compound **130** in 97% yield. A few different attempts at a nucleophilic substitution reaction with aminol **(*S*)-123** was performed, but the desired product was never detected (Scheme **51**). Instead, the silyl ether cleaved off giving penicillanic acid **124**, which was confirmed by TLC-MS (Exact mass 278.96, detected m/z 277.91 and 279.88

[M-H]$^-$) by the appearance of two peaks of identical intensity, displaying the characteristic isotopic distribution of bromine.

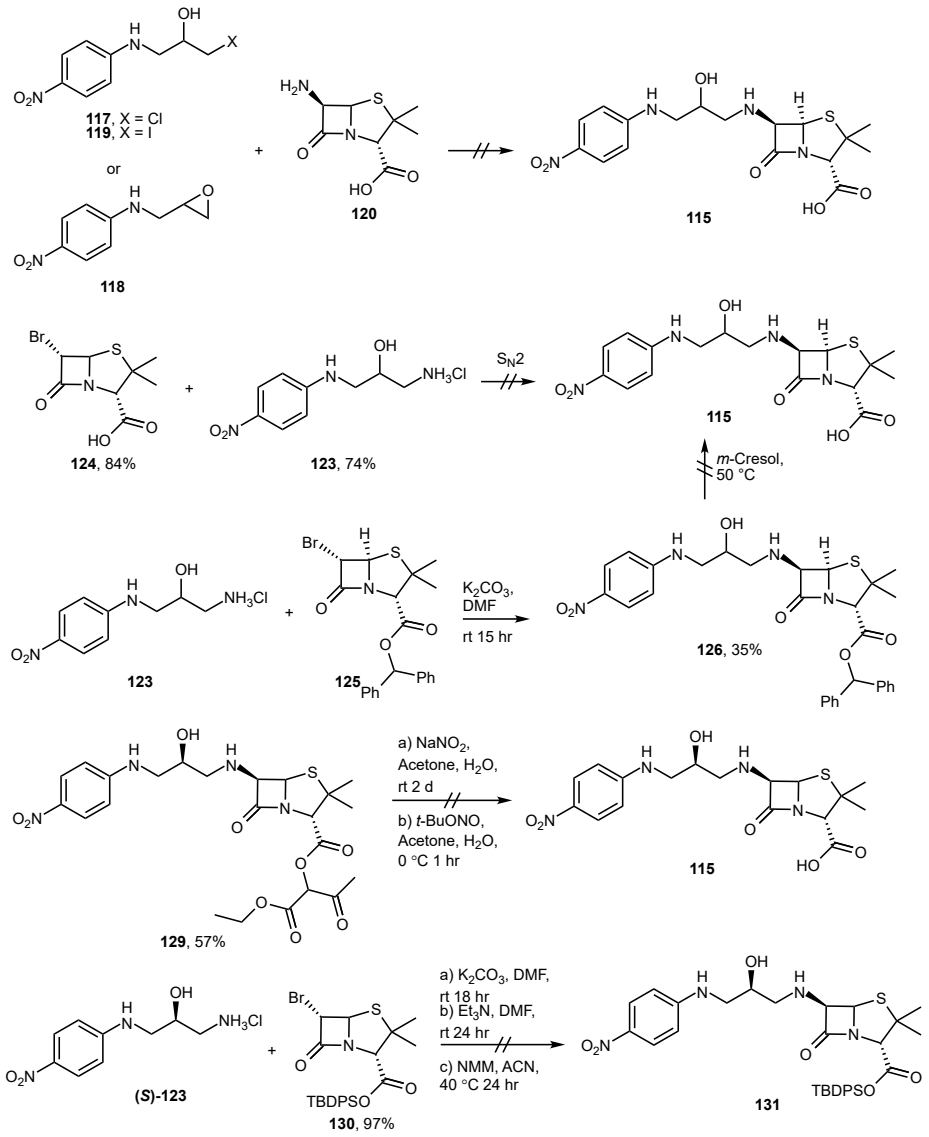


Scheme 51. Attempted synthesis of β-lactam **115** through TBDPS-protected compound **131**.

VI.2 Summary of synthetic outcomes

At first, reaction of 6-APA (**120**) was attempted with halohydrins **117** and **119** and epoxide **118** in an S_N2 or a ring-opening reaction, respectively. However, no reactivity was observed, and the halohydrins simply collapsed to give epoxide **118**, which did not react with 6-APA (**120**). Reversing the reactivity, and reacting 6-bromopenicillanic acid (**124**) with amine **123** also failed to give the desired product. Protecting the carboxylic acid in compound **124** as the diphenylmethyl ester and subsequent S_N2 reaction with amine **123** gave the protected β-lactam **126**. However, after several attempts, the deprotection did not yield the target compound **115**. Instead, a fragmentation seems to occur, since the aromatic signals from the nitroaniline and the signals corresponding to the β-lactam

part clearly belongs to two separate compounds, indicated by integral values in the ^{1}H-NMR spectrum. Another protecting group, dicarbonyl ester **129**, was attempted, but deprotection gave a complex reaction mixture with at least seven products, none of which was the desired compound **115**. A TBDPS ester was introduced, hoping that this protecting group could be removed under extremely mild reaction conditions. During the nucleophilic substitutiton with amine **123**, the silyl ester was cleaved, and β-lactam **131** was not formed.



Scheme 52. Summary of synthetic outcomes in Part VI.

VI.3 Concluding remarks

Part VI presents efforts towards the synthesis of a photodegradable β-lactam antibiotic. These endeavours did not give any formation of the target compound **115**, presumably due to the lability of the β-lactam ring. Perhaps using 7-aminocephalosporanic acid instead of 6-APA (**120**) would result in a more stable compound that would survive the deprotection process under acidic condidtions. This is clearly indicated by the successful cleavage of a diphenylmethyl ester of a cephalosporanic-acid antibiotic reported by Mobashery and co-workers.[40]

Part VII

Experimental

VII.1 Experimental equipment

All chemicals were purchased from VWR Avantor, Merck, or Enamine Ltd and used without further purification. Anhydrous THF was prepared by distillation from sodium benzophenone ketyl over a nitrogen atmosphere and stored over 4 Å MS.

IR spectra were recorded on an Agilent Cary 630 FT-IR spectrophotometer equipped with an attenuated total reflectance (ATR) attachment. Samples were analysed neat and the absorption frequencies are given in wave numbers (cm^{-1}) and are assigned as broad (br) when relevant.

UV-vis spectra were obtained on an Agilent 8453 single-beam UV-vis spectrophotometer with a deuterium-discharge lamp for the UV range and a tungsten lamp for the visible wavelength range. Samples were analysed in an Agilent open-top UV quartz cell (10 mm, 3.0 mL) with ethanol as solvent. Wavelengths are reported in nm and extinction coefficients in $M^{-1}cm^{-1}$.

NMR spectra were recorded on a Bruker Ascend™ 400 spectrometer (400.13 MHz for ^{1}H, 100.61 MHz for ^{13}C, 376.46 MHz for ^{19}F, and 161.98 MHz for ^{31}P), a Bruker Biospin AV500 spectrometer (500.13 MHz for ^{1}H and 125.76 MHz for ^{13}C), or a Bruker Ascend™ 850 spectrometer (850.13 MHz for ^{1}H and 213.77 MHz for ^{13}C). Coupling constants (J) are given in Hz and the multiplicity is reported as singlet (s), doublet (d), triplet (t), quartet (q), sextet (sxt), multiplet (m), and broad singlet (bs). The chemical shift values are reported upfield to downfield in ppm and calibration is done using the residual solvent signals for chloroform-d (^{1}H 7.26 ppm; ^{13}C 77.16 ppm), methanol-d_4 (^{1}H 3.31 ppm; ^{13}C 49.00 ppm), acetonitrile-d_3 (^{1}H 1.94 ppm; ^{13}C 1.32 ppm), water-d_2 (^{1}H 4.79 ppm), and DMSO-d_6 (^{1}H 2.50 ppm; ^{13}C 39.52 ppm).[118] Calibration for ^{19}F NMR is done using α,α,α-trifluorotoluene as internal standard in chloroform-d (-62.61 ppm) and acetonitrile-d_3 (-63.10 ppm).[119]

High-resolution mass spectra were obtained on a JEOL AccuTOF™ T100GC mass spectrometer operated in ESI mode. Low-resolution mass spectra were recorded on an Advion expression^{S} compact mass spectrometer (CMS) operated in ESI mode equipped with a Plate Express® TLC plate reader for sample injection. A solution of ammonium acetate (3.0 mM) and formic acid (0.05%) in acetonitrile and water (95:5) was used as mobile phase for both positive and

negative ESI modes.

Thin-layer chromatography (TLC) was carried out with silica gel (60 F_{254}) on aluminium sheets with solvent systems consisting of various mixtures of petroleum ether, ethyl acetate, methanol, DCM, and 20% aqueous ammonia. Visualisation was achieved with either exposure to UV light (254 and/or 365 nm) or a potassium permanganate stain. Flash chromatography was performed with a hand pump and 230-400 mesh silica gel. Dry vacuum flash chromatography was performed with water aspirator vacuum and 400-600 mesh silica gel. Autoflash silica gel chromatography was performed on either an Interchim Puriflash 215 or a Biotage Selekt system with Biotage Snap Ultra HP-Sphere^{TM} 25 μm silica-gel columns.

VII.2 Experimental procedures

General procedure for lithium perchlorate-promoted epoxide ring opening

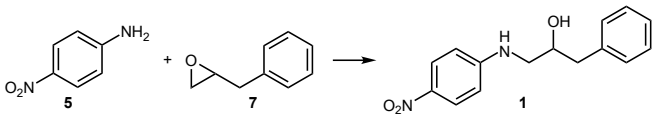
Anhydrous lithium perchlorate, prepared by drying on high vacuum at rt for 1 hr, was dissolved in anhydrous diethyl ether to a 5 M solution. Aniline (1.0-1.3 equiv., ≈0.2 M) and epoxide (1.0 equiv., ≈0.2 M) were added and the reaction mixture was stirred overnight at 40 °C under an Ar atmosphere. DCM and water was added (equal amount to reaction volume), the phases were separated, and the aqueous phase was extracted with DCM (3 x 10 mL). The com-

bined organic layers were concentrated onto celite and the target compound was isolated by silica-gel flash column chromatography (pet. ether:DCM, 3:7).

General procedure for photochemical reactions

A solution of the appropriate compound (≈0.10 mmol) in acetonitrile was added to a photochemical reactor containing distilled water at the appropriate pH to a concentration of ≈0.7 mM and a total volume of either 75 or 150 mL (7:3, water:ACN), depending on the reactor size. The reaction vessel was either purged with N_2 during the reaction or left open to air. The reaction mixture was photolysed with either a 6 W low-pressure (mainly 254 nm irradiation) or a 125 W medium-pressure (300-600 nm irradiation) mercury-vapour lamp depending on the substrate. After completion, the reaction mixture was transferred to a separatory funnel, saturated with NaCl, and extracted with either DCM or EtOAc (3 x 50 mL). The pH was adjusted to ≈2 and the aqueous layer was extracted again with either DCM or EtOAc (3 x 50 mL). The combined organic layers were dried ($MgSO_4$), filtered and concentrated on a rotary evaporator to yield a residue which was analysed by ^{1}H NMR.

1-Phenyl-3-(4-nitrophenylamino)propan-2-ol (1)



A sealed reactor tube was charged with *p*-nitroaniline (**5**) (0.28 g, 2.0 mmol), (2,3-epoxypropyl)benzene (**7**) (0.29 mL, 2.2 mmol), and methanol (0.50 mL). The reaction mixture was irradiated in the microwave reactor at 160-170 °C for 3 hr. The mixture was evaporated onto celite and the crude product was isolated by silica-gel flash chromatography (DCM:MeOH, 98:2). Concentration of the relevant fractions yielded **1** as a yellow solid (0.26 g, 47%, mp. 111-112 °C).

TLC: R_f 0.43, DCM:MeOH, 99:1.

IR (neat): ν_{max} 3392 (NH and OH, overlapping), 3029, 2921, 1600.

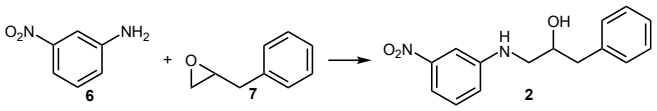
UV-vis: λ_{max} (EtOH) 384 nm (ϵ 1740 $M^{-1}cm^{-1}$).

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.08 (d, J = 9.2 Hz, 2H), 7.38-7.33 (m, 2H), 7.31-7.27 (m, 1H), 7.24-7.22 (m, 2H), 6.53 (d, J = 9.2 Hz, 2H), 4.15-4.09 (m, 1H), 3.41 (dd, J = 13.0 Hz, 3.4 Hz, 1H), 3.20 (dd, J = 13.0 Hz, 7.8 Hz, 1H), 2.92 (dd, J = 13.6 Hz, 5.1 Hz, 1H), 2.83 (dd, J = 13.6 Hz, 8.1 Hz, 1H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 153.4, 138.3, 137.1, 129.4, 129.0, 127.2, 126.5, 111.5, 71.1, 48.4, 41.9.

HRMS (ESI/TOF): Calcd for $C_{15}H_{15}N_2O_3$ $[M-H]^-$ 271.10827, found 271.10866.

1-Phenyl-3-(3-nitrophenylamino)propan-2-ol (2)



A sealed reactor tube was charged with *m*-nitroaniline (**6**) (0.28 g, 2.0 mmol), (2,3-epoxypropyl)benzene (**7**) (0.27 mL, 2.0 mmol), and methanol (0.50 mL). The reaction mixture was irradiated in the microwave reactor at 170-180 °C for 90 min. The mixture was evaporated onto celite and the crude product was isolated by flash column chromatography (DCM:pet. ether, 70:30). Concentration of the relevant fractions yielded **2** as a yellow solid (0.39 g, 72%, mp. 74-77 °C).

TLC: R_f 0.45, DCM:MeOH, 99:1.

IR (neat): ν_{max} 3545, 3302, 3081, 2915, 1618.

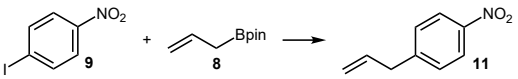
UV-vis: λ_{max} (EtOH) 400 nm (ϵ 1331 $M^{-1}cm^{-1}$).

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.53 (dd, J = 8.0 Hz, 1.9 Hz, 1H), 7.40-7.33 (m, 3H), 7.30-7.23 (m, 4H), 6.88 (dd, J = 8.0 Hz, 1.5 Hz, 1H), 4.15-4.09 (m, 1H), 3.36 (dd, J = 12.6 Hz, 2.7 Hz, 1H), 3.15 (dd, J = 12.6 Hz, 7.8 Hz, 1H), 2.92 (dd, J = 13.6 Hz, 4.9 Hz, 1H), 2.87 (dd, J = 13.6 Hz, 8.0 Hz, 1H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 149.5, 149.0, 137.3, 129.9, 129.5, 129.0, 127.1, 119.4, 112.5, 106.8, 71.1, 49.1, 41.8.

HRMS (ESI/TOF): Calcd for $C_{15}H_{15}N_2O_3$ $[M+H]^+$ 273.1234, found 273.1236.

1-Allyl-4-nitrobenzene (11)



A 25 mL round-bottom flask fitted with a condenser was charged with 1-iodo-4-nitrobenzene (**9**) (1.00 g, 4.00 mmol), CsF (1.52 g, 10.0 mmol), $Pd(PPh_3)_4$ (0.70 g, 15 mol%), THF (20 mL), and water (2 mL). The mixture was stirred at rt under Ar for 30 min, followed by addition of allylboronic acid pinacol ester (**8**) (1.36 mL, 7.20 mmol) in THF (8 mL). The reaction mixture was refluxed (oil bath, 95 °C) for 22 hr. After cooling to rt the product mixture was evaporated onto celite and purified by silica-gel column chromatography (pet. ether). Concentration of the relevant fractions yielded **11** (0.35 g, 52%) as a slightly yellow liquid. Spectroscopic data are in accordance with previously reported literature.[120]

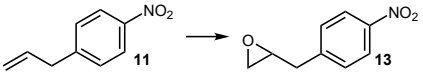
TLC: R_f 0.47, pet. ether:EtOAc, 8:2.

IR (neat): ν_{max} 3091, 3017, 2918, 1593, 1502.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.15 (d, J = 8.8 Hz, 2H), 7.34 (d, J = 8.8 Hz, 2H), 5.99-5.89 (m, 1H), 5.18-5.09 (m, 2H), 3.49 (d, J = 6.7 Hz, 2H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 147.9, 146.7, 135.6, 129.5, 123.8, 117.5, 40.0.

2-(4-Nitrobenzyl)oxirane (13)



An oven-dried 25 mL round-bottom flask charged with 1-allyl-4-nitrobenzene (**11**) (0.35 g, 2.16 mmol) in anhydrous DCM (12 mL) was cooled (ice/water bath) and stirred for 5 min under Ar followed by addition of *m*CPBA (0.59 g, 2.63 mmol). The reaction mixture was stirred at ambient temperature for 2 hr, then at rt for 15 hr. More *m*CPBA (0.12 g, 0.54 mmol) was added and stirring continued for another 31 hr before quenching the reaction with 1M aq NaOH solution (10 mL). The phases were separated and the aq phase was extracted with DCM (3 x 15 mL). The combined organic phases were washed with water (20 mL), brine (20 mL), dried ($MgSO_4$), filtered, and concentrated *in vacuo*. The product was isolated by silica-gel autoflash chromatography (pet. ether:EtOAc:DCM, 93:2:5 $\rightarrow$ 40:55:5) and concentration of the relevant fractions yielded **13** as a yellow oily liquid (0.20 g, 52%).

TLC: R_f 0.26, pet. ether:DCM, 5:5.

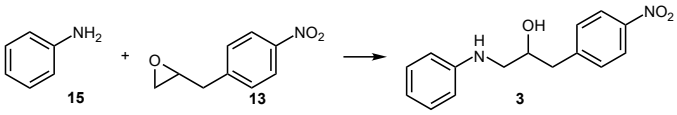
IR (neat): ν_{max} 2957, 2923, 2853, 1599, 1516, 1345.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.18 (d, J = 8.5 Hz, 2H), 7.43 (d, J = 8.5 Hz, 2H), 3.20-3.16 (m, 1H), 3.06 (dd, J = 14.8 Hz, 4.2 Hz, 1H), 2.90 (dd, J = 14.8 Hz, 6.4 Hz, 1H), 2.84 (dd, J = 4.7 Hz, 4.0 Hz, 1H), 2.55 (dd, J = 4.7 Hz, 2.6 Hz, 1H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 147.1, 145.0, 130.0, 123.9,

51.7, 46.8, 38.6.

1-(4-Nitrophenyl)-3-(phenylamino)propan-2-ol (3)



A sealed reactor tube was charged with aniline (**15**) (0.15 mL, 1.67 mmol), 2-(4-nitrobenzyl)oxirane (**13**) (0.30 g, 1.67 mmol), and methanol (0.5 mL). The reaction mixture was irradiated in the microwave reactor (170-180 °C, 9.5 bar, 300 W, 5 min ramping) for 5 min. The mixture was evaporated onto celite and the crude product was isolated by silica-gel autoflash column chromatography (pet. ether: EtOAc:DCM, 90:5:5 → 45:50:5). Concentration of the relevant fractions yielded **3** as a yellow solid (0.29 g, 64%, mp. 90-93 °C) and small amounts of a product resulting from the amino group in **3** reacting with a second unit of epoxide **13** (20 mg, 3%).

TLC: R_f 0.20, DCM:MeOH, 99:1.

IR (neat): ν_{max} 3326, 3054, 2919, 1598, 1506.

UV-vis: λ_{max} (EtOH) 248 nm (ϵ 15067 $M^{-1}cm^{-1}$).

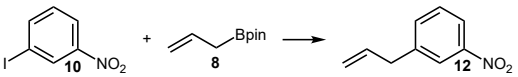
^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.17 (d, J = 8.6 Hz, 2H), 7.42 (d, J = 8.6 Hz, 2H), 7.19 (dd, J = 8.5 Hz, 7.4 Hz, 2H), 6.77 (t, J = 7.4 Hz, 1H), 6.66 (d, J = 8.5 Hz, 2H), 4.16-4.10 (m, 1H), 3.32 (dd, J = 13.1 Hz, 3.4 Hz, 1H), 3.12 (dd, J = 13.1 Hz, 8.1 Hz, 1H),

2.99 (dd, J = 13.8 Hz, 4.7 Hz, 1H), 2.92 (dd, J = 13.8 Hz, 8.0 Hz, 1H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 147.9, 146.9, 146.1, 130.3, 129.5, 123.8, 118.5, 113.5, 70.7, 49.9, 41.2.

HRMS (ESI/TOF): Calcd for $C_{15}H_{15}N_2O_3$ $[M+H]^+$ 273.1234, found 273.1236.

1-Allyl-3-nitrobenzene (12)



An oven-dried 25 mL round-bottom flask fitted with a condenser was charged with 1-iodo-3-nitrobenzene (**10**) (1.99 g, 8.00 mmol), CsF (3.65 g, 24.0 mmol), $Pd(PPh_3)_4$ (1.38 g, 15 mol%), THF (55 mL), and water (15 mL). The mixture was stirred at rt under Ar for 30 min, followed by addition of allylboronic acid pinacol ester (**8**) (2.72 mL, 14.4 mmol). The reaction mixture was refluxed (oil bath, 95 °C) for 23 hr. $Pd(PPh_3)_4$ (0.46 g, 5 mol%), CsF (1.22 g, 8.00 mmol), and allylboronic acid pinacol ester (**8**) (0.75 mL, 4.00 mmol) was added. THF was evaporated off and replaced with dioxane (55 mL) followed by reflux (oil bath, 135 °C) for 28 hr. After cooling to rt the product mixture was evaporated onto celite and purified by silica-gel autoflash chromatography (pet. ether:DCM, 95:5). Concentration of the relevant fractions yielded **12** (0.50 g, 35%) as a slightly yellow liquid. Spectroscopic data are in accordance with previously

reported literature.[120]

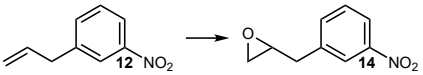
TLC: R_f 0.53, pet. ether:EtOAc, 8:2.

IR (neat): ν_{max} 3072, 2922, 1639, 1522.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.09-8.06 (m, 2H), 7.54-7.51 (m, 1H), 7.48-7.44 (m, 1H), 6.01-5.91 (m, 1H), 5.19-5.11 (m, 2H), 3.50 (d, J = 6.7 Hz, 2H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 148.6, 142.2, 135.8, 135.0, 129.4, 123.6, 121.5, 117.5, 39.8.

2-(3-Nitrobenzyl)oxirane (14)



An oven-dried 25 mL round-bottom flask charged with 1-allyl-3-nitrobenzene (**12**) (0.49 g, 3.00 mmol) in anhydrous DCM (15 mL) was cooled (ice/water bath) and stirred for 5 min under Ar followed by addition of *m*CPBA (1.35 g, 6.02 mmol). The reaction mixture was stirred at ambient temperature for 2 hr, then at rt for 26 hr before quenching with 1M aq NaOH solution (20 mL). The phases were separated and the aq phase was extracted with DCM (3 x 15 mL). The combined organic phases were washed with water (20 mL), brine (20 mL), dried ($MgSO_4$), filtered, and concentrated *in vacuo*. The product was isolated by silica-gel autoflash chromatography (pet. ether:EtOAc:DCM, 90:5:5 $\rightarrow$ 35:60:5) and concentration of the rele-

vant fractions yielded **14** as a yellow oily liquid (0.31 g, 58%).

TLC: R_f 0.28, pet. ether:EtOAc:DCM, 80:15:5.

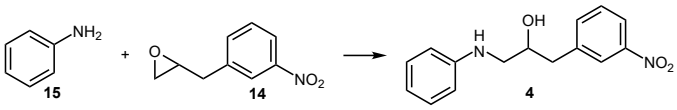
IR (neat): ν_{max} 3060, 2989, 2924, 1522, 1348.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.13-8.11 (m, 2H), 7.62-7.60 (m, 1H), 7.51-7.48 (m, 1H), 3.21-3.17 (m, 1H), 3.06 (dd, J = 14.8 Hz, 4.4 Hz, 1H), 2.92 (dd, J = 14.8 Hz, 6.3 Hz, 1H), 2.85-2.83 (m, 1H), 2,55 (dd, J = 4.8 Hz, 2.6 Hz, 1H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 148.5, 139.3, 135.4, 129.6, 124.0, 122.0, 51.8, 46.8, 38.3.

HRMS (ESI/TOF): Sample did not give signal.

1-(3-Nitrophenyl)-3-(phenylamino)propan-2-ol (4)



15 + 14 → 4

A sealed reactor tube was charged with aniline (**15**) (0.09 mL, 1.00 mmol), 2-(3-nitrobenzyl)oxirane (**14**) (0.18 g, 1.00 mmol), and methanol (0.5 mL). The reaction mixture was irradiated in the microwave reactor (170-180 °C, 9.5 bar, 300 W, 5 min ramping) for 5 min. The mixture was evaporated onto celite and the product was isolated by silica-gel autoflash column chromatography (pet. ether:EtOAc:DCM, 90:5:5 → 45:50:5). Concentration of the relevant fractions yielded **4** as a brown oily wax (0.25 g, 91%) and small amounts of a product resulting from the amino group in **4** reacting with a second unit of

epoxide **14** (30 mg, 6%).

TLC: R_f 0.52, DCM:MeOH, 95:5.

IR (neat): ν_{max} 3393, 3352, 3053, 3024, 2920, 1602.

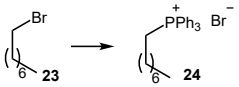
UV-vis: λ_{max} (EtOH) 248 nm (ϵ 17019 $M^{-1}cm^{-1}$).

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.14-8.13 (m, 1H), 8.10 (ddd, J = 8.2 Hz, 2.2 Hz, 1.0 Hz, 1H), 7.60-7.58 (m, 1H), 7.50-7.46 (m, 1H), 7.19 (dd, J = 8.5 Hz, 7.4 Hz, 2H), 6.76 (tt, J = 7.4 Hz, 1.0 Hz, 1H), 6.64 (dd, J = 8.5 Hz, 1.0 Hz, 2H), 4.13-4.07 (m, 1H), 3.32 (dd, J = 13.1 Hz, 3.4 Hz, 1H), 3.11 (dd, J = 13.1 Hz, 8.2 Hz, 1H), 2.98 (dd, J = 14.0 Hz, 4.6 Hz, 1H), 2.90 (dd, J = 14.0 Hz, 8.2 Hz, 1H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 148.5, 148.0, 140.3, 135.8, 129.49, 129.48, 124.3, 121.8, 118.4, 113.5, 70.7, 49.9, 40.9.

HRMS (ESI/TOF): Calcd for $C_{15}H_{15}N_2O_3$ $[M+H]^+$ 272.1234, found 273.1233.

Octyltriphenylphosphonium bromide (24)



A solution of triphenylphosphine (1574 mg, 6.00 mmol) and octyl bromide (**23**) (1.14 mL, 6.60 mmol) in toluene (20 mL) was refluxed (oil bath, 135 °C) for 4 d. Toluene was decanted off and the residue was rinsed with toluene (3 x 10 mL) to remove excess octyl bromide, to yield **24** as a colourless syrup (2695 mg, 99%).

IR (neat): ν_{max} 3390, 3051, 2923, 2853, 1586, 1436.

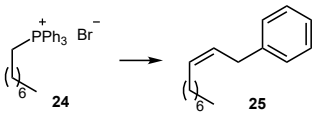
^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.90-7.84 (m, 6H), 7.81-7.76 (m, 3H), 7.72-7.67 (m, 6H), 3.90-3.83 (m, 2H), 1.64-1.61 (m, 4H), 1.25-1.19 (m, 10H), 0.83 (t,J = 6.9 Hz, 3H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 135.0 (d, J = 3.0 Hz), 133.9 (d, J = 10.0 Hz), 130.6 (d, J = 12.5 Hz), 118.7 (d, J = 85.7 Hz), 31.8, 30.5 (d, J = 15.4 Hz), 29.4, 29.0, 23.2, 22.8 (d, J = 4.5 Hz), 22.7, 14.2.

^{31}P NMR (161.98 MHz, $CDCl_3$): δ 24.6.

HRMS (ESI/TOF): Calcd for $C_{26}H_{32}P$ $[M]^+$ 375.22361, found 375.22380.

(*Z*)-Dec-2-en-1-ylbenzene (25)



To a stirred solution of octyltriphenylphosphonium bromide (**24**) (2695 mg, 5.92 mmol) in anhydr. THF (30 mL) was added a 60% dispersion of sodium hydride in mineral oil (237 mg, 5.92 mmol). After 2 hr of stirring at rt, the solution was cooled (ice/water bath), phenylacetaldehyde (0.69 mL, 5.92 mmol) was added and the reaction mixture was stirred for 1 hr at 0 °C, then 48 hr at rt. THF was removed under reduced pressure, water (20 mL) was added, followed by extraction with DCM (3 x 15 mL). The combined organic layers were concentrated onto celite and the title compound was

isolated by silica-gel flash chromatography (pet. ether). Concentration of the relevant fractions yielded a 94:6 mixture of Z:E isomers **25** as a colourless liquid (617 mg, 48%). Ratio was estimated by integrating benzylic methylene protons. Overlapping multiplet for alkene protons makes it difficult to assign stereochemistry based on coupling constant. ^{1}H NMR shift values and splitting pattern corresponds perfectly with that of (*Z*)-non-2-en-1-ylbenzene, as reported by Lipshutz and co-workers.[121] *Z*-configuration is further confirmed by coupling constant values of epoxide **26**.

TLC: R_f 0.56, pet. ether.

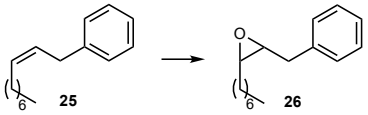
IR (neat): ν_{max} 3011, 2923, 2853, 1602, 1453.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.31-7.27 (m, 2H), 7.21-7.17 (m, 3H), 5.59-5.49 (m, 2H), 3.41 (d, J = 6.0 Hz, 2H), 2.18-2.13 (m, 2H), 1.42-1.28 (m, 10H), 0.89 (t, J = 6.9 Hz, 3H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 141.4, 131.2, 128.52, 128.49, 128.1, 125.9, 33.6, 32.0, 29.9, 29.5, 29.4, 27.4, 22.8, 14.3.

HRMS (EI/TOF): Calcd for $C_{16}H_{24}$ $[M]^+$ 216.18725, found 216.18699.

cis-2-Benzyl-3-heptyloxirane (26)



A stirred solution of (*Z*)-dec-2-en-1-ylbenzene (**25**) (616 mg, 2.85 mmol) in DCM (10 mL) under Ar was cooled (ice/water bath), fol-

lowed by addition of *m*CPBA (766 mg, 3.42 mmol). The reaction mixture was stirred at 0 °C for 2 hr and rt for 22 hr, before quenching with 1:1 sat. aq. $NaHCO_3$:10% $Na_2S_2O_3$ (20 mL). The phases were separated and the aq. layer was extracted with DCM (3 x 15 mL). The combined organic phases were washed with 1:1 sat. aq. $NaHCO_3$:10% $Na_2S_2O_3$ (20 mL), sat. aq. $NaHCO_3$ (2 x 20 mL), water (20 mL), brine (20 mL), dried ($MgSO_4$), filtered, and concentrated *in vacuo* to yield a 96:4 mixture of *cis*:*trans* isomers **26** as a colourless liquid (456 mg, 92%). Due to overlapping signals, only data for the *cis* isomer is included. Therefore, the ratio 70:30 is assumed to be identical to the ratio for compound **25**. *cis*-Configuration is confirmed by large coupling constant values (≈5 for *cis* and ≈2 for *trans*).[122]

TLC: R_f 0.33, pet. ether:EtOAc, 95:5.

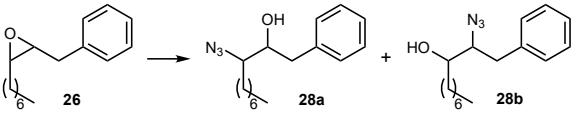
IR (neat): ν_{max} 3028, 2955, 2923, 2854, 1604.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.35-7.30 (m, 2H), 7.28-7.22 (m, 3H), 3.17 (td, J = 6.2 Hz, 4.2 Hz, 1H), 3.00 (ddd, J = 6.7 Hz, 5.5 Hz, 4.2 Hz, 1H), 2.92 (dd, J = 14.7 Hz, 6.4 Hz, 1H), 2.81 (dd, J = 14.7 Hz, 6.2 Hz, 1H), 1.70-1.60 (m, 2H), 1.58-1.23 (m, 10H), 0.89 (t, J = 7.0 Hz, 3H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 138.2, 128.9, 128.7, 126.7, 57.6, 57.5, 34.5, 31.9, 29.7, 29.4, 28.2, 26.8, 22.8, 14.2.

HRMS (ESI/TOF): Calcd for $C_{26}H_{24}ONa$ $[M+Na]^+$ 255.17194, found 255.17201.

3-Azido-1-phenyldecan-2-ol (28a)



To a stirred solution of 2-benzyl-3-heptyloxirane (**26**) (654 mg, 2.81 mmol) in MeOH (6.3 mL) and water (0.7 mL), was added NaN_3 (548 mg, 8.43 mmol) and NH_4Cl (301 mg, 5.62 mmol) at rt. The reaction mixture was stirred at 50 °C for 48 hr. MeOH and water was removed under reduced pressure and the residue was purified by silica-gel flash chromatography (pet. ether:EtOAc, 95:5 → 90:10) to yield a 70:30 mixture of regioisomers **28a**:**28b** as a colourless oily liquid (624 mg, 81%).

TLC: R_f 0.34, pet. ether:EtOAc, 9:1.

IR (neat): ν_{max} 3438 (OH), 2039, 2925, 2856, 2101 (N_3), 1604.

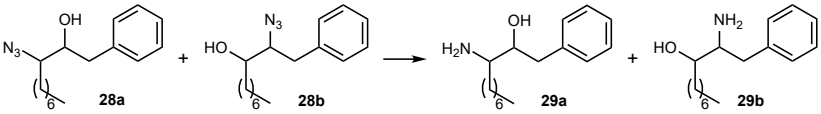
^{1}H NMR (400.13 MHz, $CDCl_3$): δ .7.35-7.31 (m, 2H, a/b), 7.28-7.22 (m, 3H, a/b), 3.84-3.79 (m, 1H, a), 3.54-3.50 (m, 1H, b), 3.49-3.45 (m, 1H, b), 3.23-3.19 (m, 1H, a), 3.03 (dd, J = 13.8 Hz, 6.1 Hz, 1H, b), 2.94 (dd, J = 13.8 Hz, 8.3 Hz, 1H, b), 2.90-2.81 (m, 2H, a), 1.77-1.61 (m, 2H, a/b), 1.60-1.23 (m, 10H, a/b), 0.90-0.86 (m, 3H, a/b).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 137.75 (a), 137.67 (b),

129.49 (a), 129.47 (b), 128.9 (a), 128.8 (b), 127.0 (b), 126.9 (a), 74.6 (a), 72.7 (b), 68.1 (b), 65.6 (a), 41.0 (a), 37.5 (b), 34.7 (b), 31.90 (b), 31.88 (a), 31.0 (a), 29.6 (b), 29.5 (a), 29.31 (b), 29.26 (a), 26.4 (a), 25.8 (b), 22.8 (a/b), 14.2 (a/b).

HRMS (ESI/TOF): Calcd for $C_{16}H_{25}N_3ONa$ $[M+Na]^+$ 298.18898, found 298.18898.

3-Amino-1-phenyldecan-2-ol (29a)



To a mixture of regioisomers **28a**:**28b** (620 mg, 2.25 mmol) dissolved in EtOAc (7 mL) was added 10% Pd/C (38 mg, 10 mol%). The reaction mixture was purged with hydrogen gas (1 atm, balloon) for 10 min before the flask was sealed and left stirring under a hydrogen atmosphere for 24 hr. Pd/C was removed by filtering through a 0.45 μm PP syringe filter and concentration of the filtrate yielded a 70:30 mixture of regioisomers **29a**:**29b** (543 mg, 97%).

TLC: R_f 0.09, DCM:MeOH, 99:1.

IR (neat): ν_{max} 3287, 3112, 3029, 2919 2852, 1602.

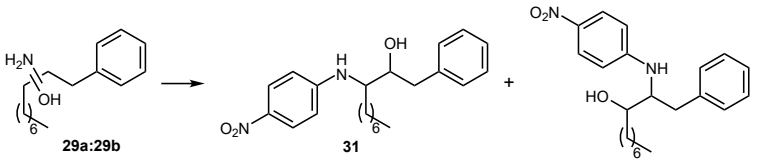
^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.35-7.29 (m, 2H, a/b), 7.25-7.18 (m, 3H, a/b), 3.58-3.53 (m, 1H, a), 3.37-3.33 (m, 1H, b), 2.95-2.86 (m, 2H, b), 2.84 (dd, J = 13.7 Hz, 4.2 Hz, 1H, a), 2.72 (dd, J = 13.7 Hz, 8.1 Hz, 1H, a), 2.64-2.60 (m, 1H, a), 2.48 (dd, J = 13.0

Hz, 9.1 Hz, 1H, b), 1.60-1.46 (m, 2H, a/b), 1.43-1.27 (m, 10H, a/b), 0.88 (t, J = 6.8 Hz, 3H, a/b).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 139.3 (b), 139.0 (a), 129.5 (a), 129.4 (b), 128.7 (b), 128.6 (a), 126.5 (b), 126.4 (a), 74.8 (a), 73.6 (b), 56.8 (b), 54.7 (a), 41.3 (b), 41.1 (a), 34.83 (b), 34.81 (a), 31.99 (b), 31.96 (a), 29.9 (b), 29.8 (a), 29.44 (b), 29.40 (a), 26.4 (a), 26.0 (b), 22.81 (b), 22.79 (a), 14.25 (b), 14.24 (a).

HRMS (ESI/TOF): Calcd for $C_{16}H_{28}NO$ $[M+H]^+$ 250.21654, found 250.21677.

3-((4-Nitrophenyl)amino)-1-phenyldecan-2-ol (31)



A solution of amines **29** (249 mg, 1.00 mmol), 1-fluoro-4-nitrobenzene (**30**) (155 mg, 1.10 mmol), DIPEA (0.52 mL, 3.00 mmol) in DMF (2 mL) was stirred at 80 °C under Ar for 24 hr. The product was isolated from a 70:30 mixture of regioisomers by two consecutive purifications by silica-gel column chromatography (pet. ether:DCM, 5:5 → 0:1 and pet. ether:DCM, 3:7) and concentration of the relevant fractions yielded **31** as a yellow waxy solid (44 mg, 12%). The undesired regioisomer was not isolated.

TLC: R_f 0.11, pet. ether:DCM, 3:7.

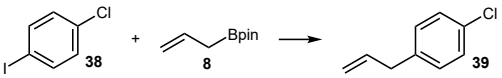
IR (neat): ν_{max} 3500 (OH), 3398 (NH), 3061, 2925, 2855, 1597.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.08 (d, J = 9.2 Hz, 2H), 7.34-7.30 (m, 2H), 7.28-7.24 (m, 1H), 7.16-7.14 (m, 2H), 6.51 (d, J = 9.2 Hz, 2H), 4.91 (d, J = 9.6 Hz, NH), 4.02 (t, J = 6.6 Hz, 1H), 3.49-3.43 (m, 1H), 2.82 (d, J = 6.9 Hz, 2H), 1.78 (bs, OH), 1.72-1.57 (m, 2H), 1.35-1.24 (m, 10H), 0.86 (t, J = 6.9 Hz, 3H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 153.7, 137.8, 137.5, 129.5, 129.0, 127.1, 126.8, 111.3, 73.5, 55.6, 41.2, 32.8, 31.9, 29.7, 29.3, 26.4, 22.7, 14.2.

HRMS (ESI/TOF): Calcd for $C_{22}H_{30}N_2O_3Na$ $[M+Na]^+$ 393.21486, found 393.21499.

1-Allyl-4-chlorobenzene (39)



A mixture of 4-chloro-1-iodobenzene (**38**) (954 mg, 4.00 mmol), $Pd(PPh_3)_4$ (46 mg, 1 mol%), CsF (2127 mg, 14.0 mmol), allyl boronate pinacol ester (**8**) (1.51 mL, 8.00 mmol) in anhydr. THF (50 mL) was refluxed (oil bath, 70 °C) under Ar for 16 hr. The reaction mixture was cooled to rt and pet. ether (20 mL) and water (20 mL) was added. The phases were separated and the aq. layer was extracted with pet. ether (3 x 15 mL). The combined organic phases were concentrated under reduced pressure onto celite and the product was isolated by silica-gel column chromatography (pet. ether). Concen-

tration of the relevant fractions yielded **39** as a colorless liquid (471 mg, 77%). Spectroscopic data are in accordance with previously reported literature.[120]

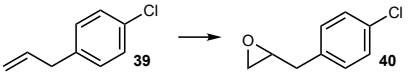
TLC: R_f 0.51, pet. ether.

IR (neat): ν_{max} 3080, 2979, 2904, 1490.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.28 (d, J = 8.5 Hz, 2H), 7.14 (d, J = 8.6 Hz, 2H), 6.01-5.91 (m, 1H), 5.13-5.11 (m, 1H), 5.10-5.07 (m, 1H), 3.38 (d, J = 6.7 Hz, 2H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 138.6, 137.0, 132.0, 130.1, 128.6, 116.4, 39.6.

2-(4-Chlorobenzyl)oxirane (40)



A stirred solution of 1-allyl-4-chlorobenzene (**39**) (450 mg, 2.95 mmol) in anhydrous DCM (8 mL) under Ar was cooled (ice/water bath), followed by addition of *m*CPBA (804 mg, 3.59 mmol). The reaction mixture was stirred at ambient temperature for 2 hr and rt for 22 hr, before quenching with 1:1 sat. aq. $NaHCO_3$:$Na_2S_2O_3$ (20 mL). The phases were separated and the aq. layer was extracted with DCM (3 x 15 mL). The combined organic phases were washed with 1:1 sat. aq. $NaHCO_3$:10% $Na_2S_2O_3$ (20 mL), sat. aq. $NaHCO_3$ (2 x 20 mL), water (20 mL), brine (20 mL), dried ($MgSO_4$), filtered, and concentrated *in vacuo* to yield **40** as a colorless liquid (456 mg,

92%). Spectroscopic data are in accordance with previously reported literature.[123]

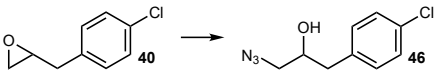
TLC: R_f 0.60, pet. ether:EtOAc 6:4.

IR (neat): ν_{max} 3051, 2988, 2918, 1490.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.28 (d, J = 8.5 Hz, 2H), 7.19 (d, J = 8.6 Hz, 2H), 3.15-3.10 (m, 1H), 2.84 (d, J = 5.4 Hz, 2H), 2.79 (dd, J = 4.9 Hz, 3.9 Hz, 1H), 2.52 (dd, J = 4.9 Hz, 2.7 Hz, 1H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 135.7, 132.7, 130.5, 128.8, 52.3, 46.8, 38.1.

1-Azido-3-(4-chlorophenyl)propan-2-ol (46)



To a stirred solution of 2-(4-chlorobenzyl)oxirane (**40**) (214 mg, 1.27 mmol) in MeOH (2.7 mL) and water (0.3 mL), was added NaN_3 (248 mg, 3.81 mmol) and NH_4Cl (136 mg, 2.54 mmol) at rt. The reaction mixture was stirred at rt for 18 hr. MeOH was removed under reduced pressure and water (5 mL) and EtOAc (5 mL) was added. The phases were separated and the aq. layer was extracted with EtOAc (3 x 10 mL). The combined organic layers were dried ($MgSO_4$), filtered, and concentrated under reduced pressure to yield **46** as a colorless oily liquid (250 mg, 93%), which was essentially

pure based on ^{1}H NMR.

TLC: R_f 0.59, pet. ether:EtOAc 5:5.

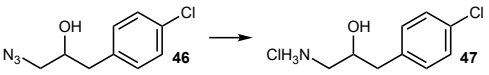
IR (neat): ν_{max} 3415 (OH), 2922, 2096 (N_3), 1491.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.30 (d, J = 8.5 Hz, 2H), 7.15 (d, J = 8.6 Hz, 2H), 4.00-3.94 (m, 1H), 3.39 (dd, J = 12.5 Hz, 3.7 Hz, 1H), 3.29 (dd, J = 12.5 Hz, 6.8 Hz, 1H), 2.79-2.77 (m, 2H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 135.7, 132.9, 130.8, 129.0, 71.6, 56.1, 40.2.

HRMS (ESI/TOF): Sample gave a signal that could not be attributed to the compound, possibly due to azide fragmentation.

1-Amino-3-(4-chlorophenyl)propan-2-ol hydrochloride (47)



A solution of azide **46** (520 mg, 2.46 mmol) and PPh_3 (708 mg, 2.70 mmol) in THF (9 mL) and water (1 mL) was stirred at 50 °C under Ar for 2 hr. THF was removed under reduced pressure, and EtOAc (20 mL) and 6 M aq. hydrochloric acid (20 mL) was added. The phases were separated and the organic layer was extracted with water (2 x 10 mL). The combined aq. phases were washed with Et_2O (40 mL) and concentrated under reduced pressure. Traces of water was azeotropically removed with toluene (3 x 5 mL) to yield **47** as sharp white needles (459 mg, 84%, mp. 188-190 °C).

TLC: R_f 0.17 as freebase, DCM:EtOH:20% NH_3 89:10:1.

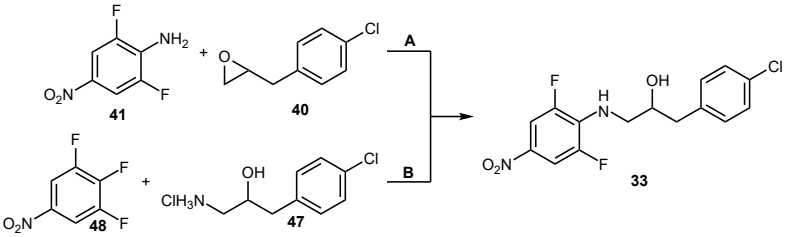
IR (neat): ν_{max} 3452 (OH), 3209, 2913 (br), 1600.

^{1}H NMR (400.13 MHz, D_2O) δ 7.30 (d, J = 8.4 Hz, 2H), 7.21 (d, J = 8.4 Hz, 2H), 4.10-4.04 (m, 1H), 3.17 (dd, J = 13.1 Hz, 2.5 Hz, 1H), 2.93 (dd, J = 13.0 Hz, 10.2 Hz, 1H), 2.84 (dd, J = 14.0 Hz, 4.9 Hz, 1H), 2.72 (dd, J = 14.0 Hz, 8.4 Hz, 1H).

^{13}C NMR (100.61 MHz, D_2O) δ 135.8, 131.9, 130.9, 128.5, 68.6, 44.1, 39.7.

HRMS (ESI/TOF): Calcd for $C_9H_{13}NOCl$ $[M+H]^+$ 186.06802, found 186.06890.

1-(4-Chlorophenyl)-3-((2,6-difluoro-4-nitrophenyl)amino)-propan-2-ol (33)



Method A: Lithium perchlorate-promoted epoxide ring opening

2,6-Difluoro-4-nitroaniline (**41**) (272 mg, 1.56 mmol) and 2-(4-chlorobenzyl)oxirane (**40**) (220 mg, 1.30 mmol) were reacted according to the general procedure for 24 hr, and isolation by silica-gel flash column chromatography (pet. ether:DCM, 4:6) yielded the target compound **33** (31 mg, 7%, R_f 0.17, DCM), ethylated compound **45** (10 mg, 2%, R_f 0.31, DCM), ethoxylated epoxide **43** (72 mg, 26%, R_f

0.33, pet. ether:DCM, 4:6), and ethylated aniline **44** (trace amounts, R_f 0.54, pet. ether:DCM, 4:6).

Method B: Nucleophilic aromatic substitution

A solution of amine hydrochloride **47** (222 mg, 1.00 mmol), 3,4,5-trifluoronitrobenzene (**48**) (128 µL, 1.10 mmol), DIPEA (700 µL, 4.00 mmol) in ACN (6 mL) was stirred at 40 °C under Ar for 14 hr. The product was isolated by silica-gel column chromatography (DCM) and concentration of the relevant fractions yielded **33** as a yellow crystalline solid (273 mg, 80%, mp. 130-132 °C).

TLC: R_f 0.18, DCM.

IR (neat): ν_{max} 3491 (NH), 3294 (OH), 3095, 3023, 2897, 1610.

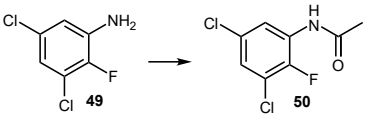
^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.78 (dd, J = 8.1 Hz, 2.1 Hz, 2H), 7.31 (d, J = 8.4 Hz, 2H), 7.16 (d, J 8.4 Hz, 2H), 4.80 (bs, NH), 4.07-4.01 (m, 1H), 3.75 (d, J = 13.4 Hz, 1H), 3.43 (dd, J = 13.4 Hz, 8.2 Hz, 1H), 2.87 (dd, J = 13.7 Hz, 4.5 Hz, 1H), 2.74 (dd, J = 13.7 Hz, 8.5 Hz, 1H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 150.1 (dd, J = 243.6 Hz, 9.0 Hz), 135.9 (t, J = 10.7 Hz), 135.5, 133.1, 132.7 (t, J = 12.6 Hz), 130.8, 129.1, 109.0 (dd, J = 18.2 Hz, 9.6 Hz), 71.8, 50.1 (t, J = 4.5 Hz), 40.9.

^{19}F NMR (376.46 MHz, $CDCl_3$): δ -128.8.

HRMS (ESI/TOF): Calcd for $C_{15}H_{12}N_2O_3ClF_2$ $[M\text{-}H]^-$ 341.05100, found 341.05100.

N-(3,5-Dichloro-2-fluorophenyl)acetamide (50)



A solution of 3,5-dichloro-2-fluoroaniline (**49**) (216 mg, 1.20 mmol) in anhydr. DCM (4 mL) was cooled (ice/water bath), followed by dropwise addition of acetyl chloride (140 µL, 1.92 mmol) and Et_3N (270 µL, 1.92 mmol) over a period of 5 min. The reaction mixture was stirred at ambient temperature for 30 min, then at rt. for another 30 min, before quenching with water (10 mL) and sat. aq. $NaHCO_3$ solution (10 mL). The phases were separated, the aq. layer was extracted with DCM (3 x 10 mL), the combined organic layers were concentrated, and the product was isolated by silica-gel column chromatography (pet. ether:EtOAc, 8:2). Concentration of the relevant fractions yielded **50** as a white solid (242 mg, 91%, mp. 168-169 °C).

TLC: R_f 0.24, pet. ether:EtOAc, 8:2.

IR (neat): ν_{max} 3293, 3252, 3116, 3046, 2990, 2923, 1678.

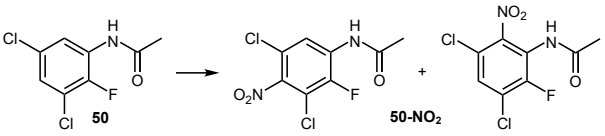
^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.35 (dd, J = 5.9 Hz, 2.0 Hz, 1H), 7.33 (bs, NH), 7.11 (dd, J = 6.2 Hz, 2.6 Hz, 1H), 2.24 (s, 3H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 168.3, 145.7, 130.1 (d, J = 4.6 Hz), 128.4 (d, J = 11.0 Hz), 124.5, 121.3 (d, J = 17.7 Hz), 119.9, 24.9.

^{19}F NMR (376.46 MHz, $CDCl_3$): δ -135.2.

HRMS (ESI/TOF): Calcd for $C_8H_5NOCl_2F$ $[M\text{-}H]^-$ 219.97377, found 219.97361.

N-(3,5-Dichloro-2-fluoronitrophenyl)acetamide (50-NO_2)



A stirred solution of *N*-(3,5-Dichloro-2-fluorophenyl)acetamide (**50**) (520 mg, 2.34 mmol) in conc. sulfuric acid (6.5 mL) was cooled to -10 °C (ice/salt bath), followed by dropwise addition of an ice cold mixture of conc. sulfuric acid (6.5 mL) and 65% nitric acid (8.4 mL) over a period of 15 min. The reaction mixture was stirred at ambient temperature for 1 hr and then poured into a beaker with ice. DCM (20 mL) was added, the phases were separated, and the aq layer was extracted with DCM (3 x 15 mL). The combined organic layers were washed with water (20 mL), dried ($MgSO_4$), filtered, and concentrated under reduced pressure to yield a 62:38 mixture of *p*- and *o*-nitrated products **50-NO_2**, as evident from ^{1}H-NMR analysis, as an off-white solid (500 mg, 80%). The next step was performed without further purification.

TLC: R_f 0.47 (*p*-NO_2), 0.51 (*o*-NO_2), pet. ether:EtOAc, 6:4.

IR (neat): ν_{max} 3263 (NH), 3194, 3112, 3073, 1706, 1682.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.63 (d, J = 6.7 Hz, 1H,

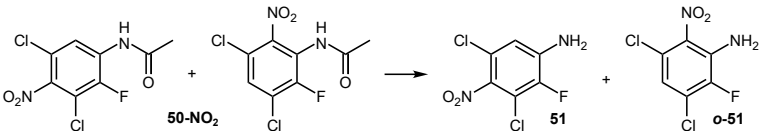
p-NO_2), 7.60 (bs, NH, *p*-NO_2), 2.28 (s, 3H, *p*-NO_2), 7.52 (d, J = 6.3 Hz, 1H, *o*-NO_2), 7.42 (bs, NH, *o*-NO_2), 2.21 (s, 3H, *o*-NO_2).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 168.9 (C=O, *p*-NO_2), 168.7 (C=O, *o*-NO_2), 152.4 (d, J = 256.7 Hz), 146.4 (d, J = 248.3 Hz), 143.5, 129.8, 129.5 (d, J = 10.8 Hz), 125.9 (d, J = 18.5 Hz), 122.4 (d, J = 4.6 Hz), 122.1 (d, J = 5.1 Hz), 121.1 (d, J = 17.8 Hz), 120.2, 115.1 (d, J = 21.8 Hz), 24.9 (CH_3, *p*-NO_2), 23.2 (CH_3, *o*-NO_2).

^{19}F NMR (376.46 MHz, $CDCl_3$): δ -114.7 (*o*-NO_2), -129.7 (*p*-NO_2).

HRMS (ESI/TOF): Calcd for $C_8H_4N_2O_3Cl_2F$ $[M\text{-}H]^-$ 264.95885, found 264.95792.

3,5-Dichloro-2-fluoro-4-nitroaniline (51)



A mixture of *o*- and *p*-nitro isomers of acetamide **50-NO_2** (500 mg, 1.87 mmol) was dissolved in methanol (25 mL) and 37% hydrochloric acid (3 mL) followed by stirring at 60 °C for 6 hr. The volatiles were removed under reduced pressure and the product was isolated by silica-gel autoflash chromatography (pet. ether:EtOAc, 9:1 → 7:3). Concentration of the relevant fractions yielded *p*-nitro isomer **51** (284 mg, 67%, mp. 142-144 °C) and *o*-nitro isomer ***o*-51** (138 mg, 33%, mp. 68-69 °C) as yellow crystalline solids.

***p*-51**:

TLC: R_f 0.17, pet. ether:EtOAc, 8:2.

IR (neat): ν_{max} 3498 (NH_2), 3394 (NH_2), 3205, 3055, 1623.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 6.75 (d, J = 7.5 Hz, 1H), 4.27 (bs, NH_2).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 145.1 (d, J = 244.9 Hz), 139.2, 137.8 (d, J = 13.4 Hz), 122.6 (d, J = 4.2 Hz), 116.0 (d, J = 20.5 Hz), 114.1 (d, J = 3.9 Hz).

^{19}F NMR (376.46 MHz, $CDCl_3$): δ -135.1.

HRMS (ESI/TOF): Calcd for $C_8H_{12}Cl_2FN_2O_4$ $[M+2CH_3OH+H]^+$ 289.01527, found 289.01543.

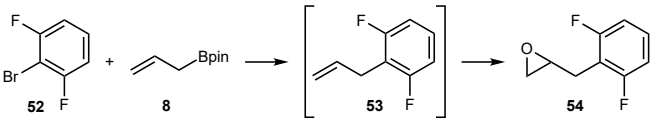
***o*-51**:

TLC: R_f 0.42, pet. ether:EtOAc, 8:2.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 6.85 (dd, J = 6.6 Hz, 1.2 Hz, 1H), 5.28 (bs, NH_2)

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 146.3 (d, J = 244.4 Hz), 134.3 (d, J = 16.2 Hz), 133.5, 124.6 (d, J = 16.4 Hz), 124.1 (d, J = 4.8 Hz), 118.7.

2-(2,6-Difluorobenzyl)oxirane (54)



A 35 mL reactor tube charged with 2-bromo-1,3-difluorobenzene

(**52**) (965 mg, 5.00 mmol), allylboronic acid pinacol ester (**8**) (1008 mg, 6.00 mmol), $Pd(PPh_3)_4$ (289 mg, 5 mol%), cesium fluoride (2658 mg, 17.5 mmol), and anhydr. THF (15 mL) was purged with argon gas and irradiated at 120 °C for 1 hr in a microwave reactor. The resulting slurry was filtered through paper with the aid of DCM (100 mL) and the filtrate was evaporated onto celite. Allylated product **53** was isolated by silica-gel flash chromatography (pet. ether) and the relevant fractions (R_f 0.65, pet. ether) were concentrated until 5 mL pet. ether remained. Anhydr. DCM (15 mL) was added and the solution was cooled to 0 °C (ice/water) followed by addition of *m*CPBA (1345 mg, 6.00 mmol). The reaction mixture was stirred at ambient temperature for 2 hr, then rt. for 26 hr before quenching with 1:1 sat. aq. $NaHCO_3$:10% $Na_2S_2O_3$ solution (30 mL). The phases were separated and the aq. layer was extracted with DCM (3 x 15 mL). The combined organic layers were washed with 1:1 sat. aq. $NaHCO_3$:10% $Na_2S_2O_3$ solution (30 mL), sat. aq. $NaHCO_3$ solution (30 mL), water (30 mL), dried ($MgSO_4$, filtered, and concentrated to yield **54** as a colorless liquid (340 mg, 40% over two steps).

TLC: R_f 0.43, pet. ether:DCM, 5:5.

IR (neat): ν_{max} 3056, 2998, 2928, 1626, 1589, 1468.

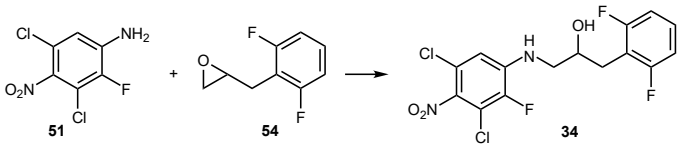
^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.24-7.16 (m, 1H), 6.90-6.86 (m, 2H), 3.19-3.10 (m, 2H), 2.82 (dd, J = 14.0 Hz, 5.8 Hz, 1H), 2.77-2.75 (m, 1H), 2.56 (dd, J = 4.9 Hz, 2.4 Hz).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 161.9 (dd, J = 247.4 Hz, 8.5 Hz), 128.6 (t, J = 10.2 Hz), 112.6 (t, J = 20.5 Hz), 111.3 (dd, J = 19.0 Hz, 6.9 Hz), 50.8, 47.1, 25.6 (t, J = 2.0 Hz).

^{19}F NMR (376.46 MHz, $CDCl_3$): δ -114.8.

HRMS: Sample sent for analysis.

1-((3,5-Dichloro-2-fluoro-4-nitrophenyl)amino)-3-(2,6-difluorophenyl)propan-2-ol (34)

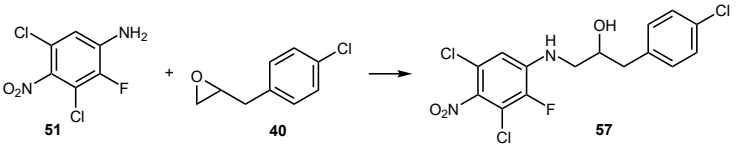


An oven-dried 25 mL round-bottom flask was charged with lithium perchlorate (2.52 g, 23.7 mmol) and diethyl ether (5 mL). The solution was stirred for 30 min followed by addition of 3,5-dichloro-2-fluoro-4-nitroaniline (**51**) (260 mg, 1.16 mmol) and 2-(2,6-difluorobenzyl)oxirane (**54**) (198 mg, 1.16 mmol). The reaction mixture was stirred at reflux (oil bath, 60 °C) for 3 d before DCM (10 mL) was added followed by dropwise addition of water (10 mL). The phases were separated and the organic layer was extracted with DCM (3 x 10 mL). Isolation by silica-gel flash chromatography (pet. ether:DCM, 5:5) and concentration of the relevant fractions yielded **34** (91 mg, 20%, mp. 158-159 °C), recovered 3,5-dichloro-2-fluoro-4-nitroaniline (**51**) (89 mg, 34%), 3,5-dichloro-*N*-ethyl-2-fluoro-4-nitroaniline (**55**) (81 mg, 28%, mp. 77-79 °C, R_f 0.60, pet. ether:DCM, 3:7) as yellow crystalline solids, and 1-(2,6-difluorophenyl)-3-ethoxy-propan-2-

ol (**56**) (2 mg, <2%, R_f 0.26, pet. ether:DCM, 3:7) as an oily liquid.

TLC: R_f 0.16, pet. ether:DCM, 3:7.
IR (neat): ν_{max} 3402 (OH), 3369 (NH), 2968, 2928, 1604.
^{1}H NMR (400.13 MHz, CD_3CN): δ 7.32-7.24 (m, 1H), 7.00-6.93 (m, 1H), 6.83 (d, J = 7.5 Hz, 1H), 5.45 (bs, NH), 4.05-3.98 (m, 1H), 3.37-3.31 (m, 1H), 3.31 (d, J = 5.3 Hz, OH), 2.87-2.85 (m, 2H).
^{13}C NMR (100.61 MHz, CD_3CN): δ 162.8 (dd, J = 245 Hz, 9 Hz), 116.0 (d, J = 244 Hz), 141.3 (d, J = 12 Hz), 137.3, 129.5 (t, J = 10 Hz), 123.4 (d, J = 4 Hz), 115.1 (d, J = 20 Hz), 115.1 (t, J = 20 Hz), 112.1 (d, J = 26 Hz), 111.0 (J = 4 Hz), 69.8, 49.1, 28.7.
^{19}F NMR (376.46 MHz, CD_3CN): δ -115.7, -136.8.
HRMS (ESI/TOF): Calcd for $C_{15}H_{10}N_2O_3Cl_2F_3$ $[M-H]^-$ 393.00261, found 393.00245.

3-(4-Chlorophenyl)-1-((3,5-dichloro-2-fluoro-4-nitrophenyl)-amino)propan-2-ol (57)



3,5-Dichloro-2-fluoro-4-nitroaniline (**51**) (77 mg, 0.34 mmol) and 2-(4-chlorobenzyl)oxirane (**40**) (57 mg, 0.34 mmol) was reacted according to the general procedure for 20 hr. The target compound was isolated by two consecutive purifications by silica-gel flash chromatography (pet. ether:DCM, 2:8 and pet. ether:EtOAc, 7:3). The

relevant fractions were concentrated and pet. ether (2 mL) and EtOAc (3 drops) was added to the residue. The mixture was heated to 50 °C and the liquid was decanted off, leaving a yellow solid containing 80% pure product. This mixture was washed with a solution of pet. ether (10 mL) and EtOAc (15 drops), followed by a final rinse with EtOAc (10 mL) to yield aminol **57** as a yellow crystalline solid (20 mg, 15%, mp. 148-149 °C).

TLC: R_f 0.46, pet. ether:EtOAc, 6:4.

IR (neat): ν_{max} 3380 (OH, NH overlapping), 3091, 2918, 2859.

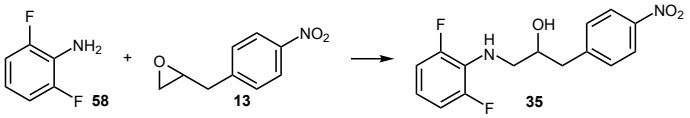
^{1}H NMR (400.13 MHz, CD_3CN): δ 7.31 (d, J = 8.5 Hz, 2H), 7.25 (d, J = 8.5 Hz, 2H), 6.79 (d, J = 7.5 Hz, 1H), 5,44 (bs, NH), 3.99-3.91 (m, 1H), 3.29 (ddd, J = 13.7 Hz, 6.3 Hz, 3.9 Hz, 1H), 3.19 (d, J = 5.1 Hz, OH), 3.17-3.11 (m, 1H), 2.82 (dd, J = 13.8 Hz, 4.9 Hz, 1H), 2.70 (dd, J = 13.8 Hz, 8.0 Hz, 1H).

^{13}C NMR (100.61 MHz, CD_3CN): δ 146.0 (d, J = 244 Hz), 141.3 (d, J = 12 Hz), 138.7, 137.3, 132.5, 132.2, 129.2, 123.5 (d, J = 4 Hz), 115.2 (d, J = 21 Hz), 111.0 (d, J = 4 Hz), 71.2, 49.1, 41.1.

^{19}F NMR (376.46 MHz, CD_3CN): δ -136.7.

HRMS (ESI/TOF): Calcd for $C_{15}H_{11}Cl_3FN_2O_3$ $[M-H]^-$ 390.98248, found 390.98148.

1-((2,6-Difluorophenyl)amino)-3-(4-nitrophenyl)propan-2-ol (35)



2,6-Difluoroaniline (**58**) (43 mg, 0.33 mmol) and 2-(4-nitrobenzyl)-oxirane (**13**) (59 mg, 0.33 mmol) was reacted according to the general procedure for 6 hr to yield aminol **35** as a white solid (31 mg, 30%, mp. 86-87 °C) along with 43% recovery of epoxide.

TLC: R_f 0.15, DCM.

IR (neat): ν_{max} 3308 (NH, OH), 3080, 2946, 2886, 1600.

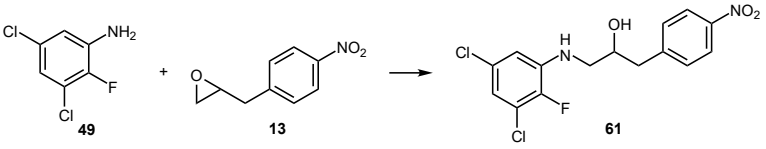
^{1}H NMR (400.13 MHz, CD_3CN): δ 8.13 (d, J = 8.7 Hz, 2H), 7.43 (d, J = 8.7 Hz, 2H), 6.91-6.81 (m, 2H), 6.73-6.65 (m, 1H), 4.27 (bs, NH), 3.98-3.90 (m, 1H), 3.46-3.40 (m, 1H), 3.20-3.14 (m, 1H, overlapping with OH), 3.17 (d, J = 5.3 Hz, OH), 2.95 (dd, J = 13.7 Hz, 4.4 Hz, 1H), 2.80 (dd, J = 13.7 Hz, 8.4 Hz, 1H).

^{13}C NMR (100.61 MHz, CD_3CN): δ 154.3 (dd, J = 240 Hz, 8 Hz), 148.4, 147.6, 131.5, 127.0 (t, J = 14 Hz), 124.2, 118.5 (t, J = 10 Hz), 112.5 (dd, J = 16 Hz, 7 Hz), 71.9, 52.4 (t, J = 4 Hz), 41.7.

^{19}F NMR (376.46 MHz, CD_3CN): δ -130.0.

HRMS (ESI/TOF): Calcd for $C_{15}H_{15}F_2N_2O_3$ $[M+H]^+$ 309.10453, found 309.10467.

1-((3,5-Dichloro-2-fluorophenyl)amino)-3-(4-nitrophenyl)-propan-2-ol (61)



3,5-Dichloro-2-fluoroaniline (**49**) (59 mg, 0.33 mmol) and 2-(4-nitrobenzyl)oxirane (**13**) (59 mg, 0.33 mmol) was reacted according to the general procedure for 20 hr to yield aminol **61** as a white solid (57 mg, 48%, mp. 113-114 °C) along with 32% recovery of epoxide.

TLC: R_f 0.31, DCM.

IR (neat): ν_{max} 3447 (NH), 3351 (OH), 3263, 3117, 3085, 2950.

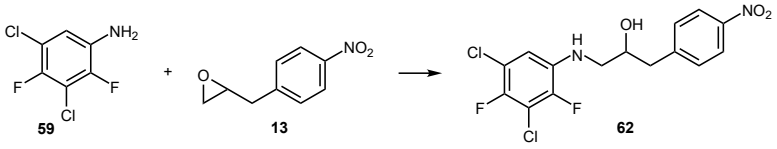
^{1}H NMR (400.13 MHz, CD_3CN): δ 8.13 (d, J = 8.8 Hz, 2H), 7.47 (d, J = 8.8 Hz, 2H), 6.68-6.64 (m, 2H), 4.92 (bs, NH), 4.03-3.96 (m, 1H), 3.28-3.22 (m, 1H, overlapping with OH), 3.24 (d, J = 5.1 Hz, OH), 3.12-3.05 (m, 1H), 2.97 (dd, J = 13.7 Hz, 4.4 Hz, 1H), 2.82 (dd, J = 13.7 Hz, 8.4 Hz, 1H).

^{13}C NMR (100.61 MHz, CD_3CN): δ 148.3, 147.6, 146.8 (d, J = 239 Hz), 139.9 (d, J = 12 Hz), 131.5, 130.5 (d, J = 4 Hz), 124.2, 121.3 (d, J = 16 Hz), 116.4 (d, J = 2 Hz), 111.5 (d, J = 3 Hz), 70.9, 49.5, 41.6.

^{19}F NMR (376.46 MHz, CD_3CN): δ -141.6.

HRMS (ESI/TOF): Calcd for $C_{15}H_{14}Cl_2FN_2O_3$ $[M+H]^+$ 359.03600, found 359.03598.

1-((3,5-Dichloro-2,4-difluorophenyl)amino)-3-(4-nitrophenyl)propan-2-ol (62)



3,5-Dichloro-2,4-difluoroaniline (**59**) (65 mg, 0.33 mmol) and 2-(4-nitro-benzyl)oxirane (59 mg, 0.33 mmol) was reacted according to the general procedure for 18 hr to yield aminol **62** as a white solid (59 mg, 48%, mp. 124-125 °C) along with 24% recovery of epoxide.

TLC: R_f 0.29, DCM.

IR (neat): ν_{max} 3386 (NH), 3293 (OH), 3116, 3080, 2927, 1601.

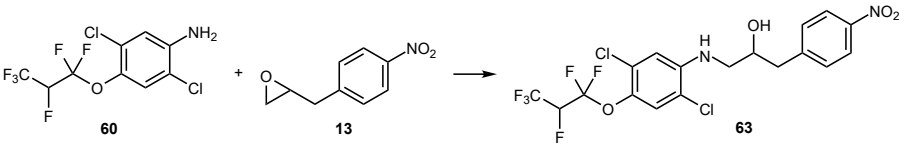
^{1}H NMR (400.13 MHz, CD_3CN): δ 8.14 (d, J = 8.8 Hz, 2H), 7.48 (d, J = 8.8 Hz, 2H), 6.76 (dd, J = 8.5 Hz, 7.2 Hz, 1H), 4.72 (bs, NH), 4.04-3.96 (m, 1H), 3.26-3.21 (m, 1H, overlapping with OH), 3.21 (d, J = 5.0 Hz, OH), 3.10-3.04 (m, 1H), 2.97 (dd, J = 13.7 Hz, 4.4 Hz, 1H), 2.82 (dd, J = 13.7 Hz, 8.4 Hz, 1H).

^{13}C NMR (100.61 MHz, CD_3CN): δ 148.3, 147.7, 146.9 (dd, J = 242 Hz, 2 Hz), 146.3 (dd, J = 237 Hz, 2 Hz), 135.7 (dd, J = 12 Hz, 3 Hz), 131.5, 124.2, 117.3 (dd, J = 18 Hz, 4 Hz), 111.2 (dd, J = 22 Hz, 20 Hz), 111.1 (d, J = 4 Hz), 70.9, 49.8, 41.6.

^{19}F NMR (376.46 MHz, CD_3CN): δ -134.2 (d, J = 4.4 Hz), -136.9 (d, J = 4.4 Hz).

HRMS (ESI/TOF): $C_{15}H_{13}Cl_2F_2N_2O_3$ $[M+H]^+$ 377.02658, found 377.02648.

1-((2,5-Dichloro-4-(1,1,2,3,3,3-hexafluoropropoxy)phenyl)-amino)-3-(4-nitrophenyl)propan-2-ol (63)



2,5-Dichloro-4-(1,1,2,3,3,3-hexafluoropropoxy)aniline (**60**) (111 mg, 0.34 mmol) and 2-(4-nitrobenzyl)oxirane (**13**) (62 mg, 0.34 mmol) was reacted according to the general procedure for 19 hr to yield aminol **63** as a white solid (87 mg, 50%, mp. 121-123 °C) along with 40% recovery of epoxide.

TLC: R_f 0.36, DCM.

IR (neat): ν_{max} 3507 (OH), 3420, 3376 (NH), 2980, 2918, 1599.

UV-vis: λ_{max} (EtOH) 255 nm (ϵ 18339 $M^{-1}cm^{-1}$).

^{1}H NMR (850.13 MHz, CD_3CN): δ 8.14 (d, J = 8.7 Hz, 2H), 7.49 (d, J = 8.7 Hz, 2H), 7.33 (s, 1H), 6.81 (s, 1H), 5.55 (dsxt, J = 42.8 Hz, 5.8 Hz, 1H), 5.03 (t, J = 5.6 Hz, NH), 4.06-4.02 (m, 1H), 3.30 (ddd, J = 13.3 Hz, 6.4 Hz, 3.9 Hz, 1H), 3.27 (d, J = 4.4 Hz, OH), 3.15-3.11 (m, 1H), 2.98 (dd, J = 13.8 Hz, 4.5 Hz, 1H), 2.85 (dd, J = 13.8 Hz, 8.4 Hz, 1H).

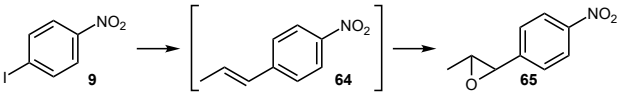
^{13}C NMR (213.77 MHz, CD_3CN): δ 148.2, 147.7, 145.2, 134.5, 131.5, 127.9, 125.6, 124.3, 121.3 (qd, J = 281 Hz, 25 Hz), 119.1 (td,

J = 271 Hz, 23 Hz), 117.7, 112.6, 85.4 (dsxt, J = 198 Hz, 35 Hz), 70.8, 49.7, 41.7

^{19}F NMR (376.46 MHz, CD_3CN): δ -75.6--75.7 (m, 3F), -78.4--80.4 (m, 2F), -213.3 (sxt, J = 12 Hz, 1F).

HRMS (ESI/TOF): Calcd for $C_{18}H_{14}Cl_2F_6N_2O_4Na$ $[M+Na]^+$ 529.01270, found 529.01286.

2-Methyl-3-(4-nitrophenyl)oxirane (65)



An oven-dried 100 mL round-bottom flask fitted with a condenser was charged with 1-iodo-4-nitrobenzene (**9**) (996 mg, 4.00 mmol), cesium fluoride (2127 mg, 14.0 mmol), Pd(PPh3)4 (231 mg, 5 mol%), allylboronic acid pinacol ester (**8**) (1344 mg, 8.00 mmol), and THF (50 mL). The reaction mixture was refluxed (oil bath, 80 °C) for 18 hr. After cooling to rt the product mixture was evaporated onto celite and purified by silica-gel flash chromatography (pet. ether). Concentration of the relevant fractions yielded a slightly yellow residue, which was dissolved in DCM (15 mL) and cooled (ice/water bath). *m*CPBA (632 mg, 2.82 mmol) was added and the reaction mixture was stirred at 0 °C for 2 hr and rt for 22 hr before quenching with 1:1 sat. $NaHCO_3$:10% $Na_2S_2O_3$ (20 mL). The phases were separated and the aq. layer was extracted with DCM (3 x 15 mL). The combined organic phases were washed with 1:1 sat. aq. $NaHCO_3$:10% $Na_2S_2O_3$ (20 mL), sat. aq. $NaHCO_3$ (2 x 20 mL), water (20 mL),

brine (20 mL), dried ($MgSO_4$), filtered, and concentrated in vacuo to yield **65** as a white solid (348 mg, 49% over two steps, mp. 79-81 °C, lit. 87-88 °C[124]). NMR data is in accordance with previously reported literature.[125]

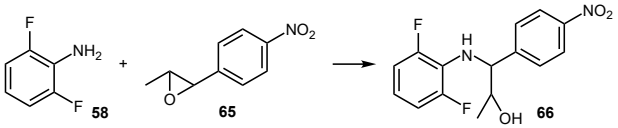
TLC: R_f 0.60, pet. ether:EtOAc 6:4.

IR (neat): ν_{max} 3109, 3073, 2977, 2933, 2855, 1601, 1513.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.20 (d, J = 8.8 Hz, 2H), 7.42 (d, J = 8.8 Hz, 2H), 3.67 (d, J = 1.9 Hz, 1H), 3.02 (qd, J = 5.1 Hz, 1.9 Hz, 1H), 1.50 (d, J = 5.1 Hz, 3H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 147.9, 145.5, 126.4, 123.9, 60.0, 58.5, 18.0.

1-((2,6-Difluorophenyl)amino)-1-(4-nitrophenyl)propan-2-ol (66)



2,6-Difluoroaniline (**58**) (0.21 mL, 2.03 mmol) and 2-methyl-3-(4-nitrophenyl)oxirane (**65**) (364 mg, 2.03 mmol) was reacted according to the general procedure for 22 hr to yield aminol **66** as a slightly yellow oily liquid (305 mg, 49%) along with 43% recovery of epoxide.

TLC: R_f 0.24, DCM.

IR (neat): ν_{max} 3526 (OH), 3382 (NH), 3077, 2973, 2932, 1623.

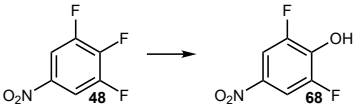
^{1}H NMR (850.13 MHz, $CDCl_3$): δ 8.15 (d, J = 8.8 Hz, 2H), 7.47 (d, J = 8.8 Hz, 2H), 6.74-6.70 (m, 2H), 6.64-6.60 (m, 1H), 4.81 (d, J = 10.1 Hz, NH), 4.73 (dd, J = 10.1 Hz, 3.7 Hz, 1H), 4.36-4.33 (m, 1H), 1.51 (d, J = 6.5 Hz, OH), 1.13 (d, J = 6.5 Hz, 3H).

^{13}C NMR (213.77 MHz, $CDCl_3$): δ 153.7 (dd, J = 242 Hz, 7 Hz), 147.5, 146.9, 129.1, 123.9 (t, J = 14 Hz), 123.4, 118.9 (t, J = 10 Hz), 111.6 (dd, J = 19 Hz, 4 Hz), 69.8, 64.1 (t, J = 4 Hz), 20.8.

^{19}F NMR (376.46 MHz, $CDCl_3$): δ -127.7.

HRMS (ESI/TOF): Calcd for $C_{15}H_{15}F_2N_2O_3$ $[M+H]^+$ 309.10453, found 309.10451.

2,6-Difluoro-4-nitrophenol (68)



A suspension of 3,4,5-trifluoronitrobenzene (**48**) (1197 mg, 6.76 mmol) in 5 M NaOH solution (15 mL) was stirred at 60 °C for 22 hr before adjusting the pH to 2 with 6 M HCl solution. The aqueous solution was extracted with DCM (4 x 15 mL), dried ($MgSO_4$), filtered, and concentrated under reduced pressure to yield phenol **68** as a white crystalline solid (1136 mg, 96%).

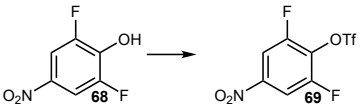
^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.90 (dd, J = 6.7 Hz, 1.0 Hz, 2H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 150.7 (dd, J = 247.9 Hz,

6.0 Hz), 139.6 (t, J = 15.9 Hz), 139.5 (t, J = 9.5 Hz), 108.9 (dd, J = 17.5 Hz, 8.9 Hz).

^{19}F NMR (376.46 MHz, $CDCl_3$): δ -131.1.

2,6-Difluoro-4-nitrophenyl trifluoromethanesulfonate (69)



2,6-Difluoro-4-nitrophenol (**68**) (533 mg, 3.00 mmol) was dissolved in anhydrous DCM (30 mL) and cooled (ice/water bath). Triethylamine (1.25 mL, 9.00 mmol) was added followed by dropwise addition of triflic anhydride (0.56 mL, 3.30 mmol) over 30 min. The resulting reaction mixture was stirred at 0 °C for 1 hr before quenching with water (20 mL). The phases were separated, the aqueous layer was extracted with DCM (3 x 15 mL), and the combined organic phases were concentrated. The residue was filtered through a short plug of silica with the aid of DCM (50 mL) and the filtrate was concentrated to yield the target compound **69** (760 mg, 83%) as a light brown transparent liquid.

TLC: R_f 0.28, pet. ether:EtOAc, 95:5.

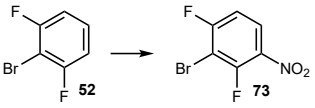
^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.04 (d, J = 6.7 Hz, 2H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 154.8 (dd, J = 260.6 Hz, 2.6 Hz), 146.8 (t, J = 8.6 Hz), 130.9 (t, J = 16.2 Hz), 118.6 (q, J = 321.1 Hz), 109.4 (dd, J = 24.1 Hz, 3.4 Hz).

^{19}F NMR (376.46 MHz, $CDCl_3$): δ -72.5 (t, J = 6.6 Hz, 3F), -117.3 (q, J = 6.6 Hz, 2F).

HRMS (ESI/TOF): Calcd for $C_6H_2F_2NO_3$ $[M\text{-}Tf]^-$ 174.00027, found 174.00065.

2-Bromo-1,3-difluoro-4-nitrobenzene (73)



2-Bromo-1,3-difluorobenzene (**52**) (579 mg, 3.00 mmol) was dissolved in 95-97% sulfuric acid (4 mL) and cooled to 0 °C. To this was added an ice-cold mixture of 95-97% sulfuric acid (4 mL) and 65% nitric acid (5.2 mL) dropwise over 15 min. The reaction mixture was stirred at 0 °C for 1 hr, poured over ice, and extracted with DCM (3 x 15 mL). The combined organic layers were washed with sat. aq. $NaHCO_3$ solution (20 mL), dried ($MgSO_4$), filtered, and concentrated under reduced pressure to yield 2-bromo-1,3-difluoro-4-nitrobenzene (**73**) as a white crystalline solid (691 mg, 97%, mp. 52-53 °C).

TLC: R_f 0.24, pet. ether:EtOAc, 95:5.

IR (neat): ν_{max} 3099, 1916, 1591, 1530.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.12 (ddd, J = 9.4 Hz, 8.0 Hz, 5.5 Hz, 1H), 7.14 (ddd, J = 9.4 Hz, 7.0 Hz, 2.0 Hz, 1H).

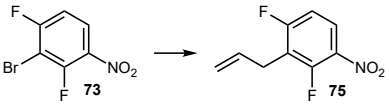
^{13}C NMR (100.61 MHz, $CDCl_3$): δ 163.1 (dd, J = 260 Hz, 3

Hz), 154.3 (dd, J = 267 Hz, 5 Hz), 134.8, 126.2 (dd, J = 10 Hz, 2 Hz), 112.1 (dd, J = 24 Hz, 4 Hz), 101.3 (dd, J = 25 Hz, 24 Hz).

^{19}F NMR (376.46 MHz, $CDCl_3$): δ -92.0 (d, J = 9.5 Hz), -104.4 (d, J = 9.5 Hz).

HRMS (EI/TOF): Calcd for $C_6H_2BrF_2NO_2$ $[M]^+$ 236.92315, found 236.92306.

2-Allyl-1,3-difluoro-4-nitrobenzene (75)



Bromobenzene **73** (870 mg, 3.66 mmol), $Pd(PPh_3)_4$ (634 mg, 15 mol%), and $Bu_3SnAllyl$ (1.53 mL, 4.39 mmol) was dissolved in anhydrous DMF (8 mL). 4 Å molecular sieves were added and the reaction mixture was stirred at 100 °C under Ar for 24 hr. The solids were filtered off through cotton with the aid of DCM (50 mL), and the volatiles were removed under reduced pressure. The title compound was isolated by silica-gel flash chromatography (pet. ether:EtOAc, 99:1) and concentration of the relevant fractions yielded 2-allyl-1,3-difluoro-4-nitrobenzene (**75**) as a colourless liquid (194 mg, 27%).

TLC: R_f 0.34, pet. ether:EtOAc, 95:5.

IR (neat): ν_{max} 3087, 2957, 2925, 2855, 1623, 1596.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.01 (ddd, J = 9.0 Hz, 8.5 Hz, 5.7 Hz, 1H), 7.04-6.99 (m, 1H), 5.95-5.85 (m, 1H), 5.13-5.08 (m,

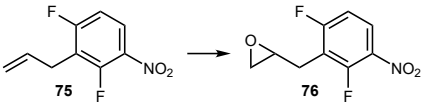
2H), 3.51-3.48 (m, 2H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 164.2 (dd, J = 258 Hz, 8 Hz), 155.2 (dd, J = 266 Hz, 10 Hz), 134.5 (dd, J = 12 Hz, 4 Hz), 132.9, 125.4 (dd, J = 11 Hz, 2 Hz), 118.8 (dd, J = 22 Hz, 19 Hz), 117.4, 111.7 (dd, J = 25 Hz, 4 Hz), 26.8 (t, J = 3 Hz).

^{19}F NMR (376.46 MHz, $CDCl_3$): δ -102.5 (d, J = 14.1 Hz), -116.6 (d, J = 14.1 Hz).

HRMS (EI/TOF): Calcd for $C_9H_7F_2NO_2$ $[M]^+$ 199.04394, found 199.04399.

2-(2,6-Difluoro-3-nitrobenzyl)oxirane (76)



To a stirred solution of 2-allyl-1,3-difluoro-4-nitrobenzene (**75**) (194 mg, 0.97 mmol) at 0 °C was added mCPBA (425 mg, 1.94 mmol). The reaction mixture was stirred for 2 hr at 0 °C and 5 d at rt, followed by addition of a 1:1 sat. aq. $NaHCO_3$:10% $Na_2S_2O_3$ solution (30 mL). The phases were separated and the aqueous layer was extracted with DCM (3 x 15 mL). The combined organic phases were washed with a 1:1 sat. aq. $NaHCO_3$:10% $Na_2S_2O_3$ solution (30 mL), sat. aq. $NaHCO_3$ (30 mL), water (30 mL), dried ($MgSO_4$), filtered and concentrated under reduced pressure to yield 2-(2,6-difluoro-3-nitrobenzyl)oxirane (**76**) as a slightly yellow oily liquid (183 mg,

88%).

TLC: R_f 0.53, DCM.

IR (neat): ν_{max} 3104, 3000, 2926, 1728, 1624.

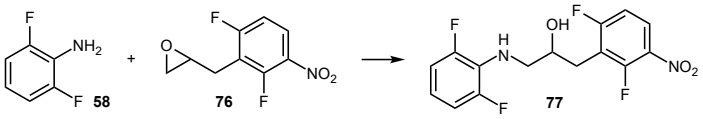
^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.05 (ddd, J = 9.2 Hz, 8.5 Hz, 5.7 Hz, 1H), 7.07-7.02 (m, 1H), 3.22-3.17 (m, 1H), 3.16-3.11 (m, 1H), 3.03-2.97 (m, 1H), 2.81-2.79 (m, 1H), 2.56 (dd, J = 4.8 Hz, 2.5 Hz, 1H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 164.5 (dd, J = 259 Hz, 8 Hz), 155.5 (dd, J = 266 Hz, 9 Hz), 134.5, 126.1 (dd, J = 11 Hz, 1 Hz), 115.8 (dd, J = 22 Hz, 20 Hz), 111.8 (dd, J = 25 Hz, 4 Hz), 50.1, 46.9, 25.8 (t, J = 2 Hz).

^{19}F NMR (376.46 MHz, $CDCl_3$): δ -101.5 (d, J = 13.6 Hz), -115.7 (d, J = 13.6 Hz).

HRMS (EI/TOF): Calcd for $C_7H_4F_2NO_2$ $[M\text{-}C_2H_2O]^+$ 172.02046, found 172.02080.

3-(2,6-Difluoro-3-nitrophenyl)-1-((2,6-difluorophenyl)-amino)propan-2-ol (77)



2,6-Difluoroaniline (**58**) (39 mg, 0.30 mmol) and 2-(2,6-difluoro-3-nitrobenzyl)oxirane (**76**) (65 mg, 0.30 mmol) was reacted according to the general procedure for 19 hr to yield aminol **77** as an off-white

solid (26 mg, 25%, mp. 74-76 °C) along with 26% recovery of epoxide.

TLC: R_f 0.23, DCM.

IR (neat): ν_{max} 3346 (NH), 3256 (OH), 3100, 2933, 1621.

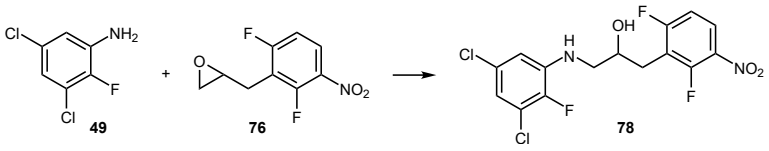
^{1}H NMR (400.13 MHz, CD_3CN): δ 8.04 (ddd, J = 9.2 Hz, 8.6 Hz, 5.7 Hz, 1H), 7.12 (ddd, J = 9.2 Hz, 8.6 Hz, 1.8 Hz, 1H), 6.91-6.81 (m, 2H), 6.72-6.66 (m, 1H), 4.31 (bs, NH), 3.98-3.90 (m, 1H), 3.50-3.44 (m, 1H), 3.31-3.20 (m, 2H, overlapping OH), 2.95-2.84 (m, 2H).

^{13}C NMR (100.61 MHz, CD_3CN): δ 165.5 (dd, J = 256 Hz, 8 Hz), 156.2 (dd, J = 263 Hz, 10 Hz), 154.3 (dd, J = 239 Hz, 8 Hz), 135.4, 126.9 (t, J = 14 Hz), 126.6 (dd, J = 12 Hz, 1 Hz), 118.7 (dd, J = 21 Hz, 19 Hz), 118.5 (t, J = 10 Hz), 112.7-112.5 (m), 112.5 (dd, J = 16 Hz, 7 Hz), 70.3, 52.3 (t, J = 4 Hz), 29.1.

^{19}F NMR (376.46 MHz, CD_3CN): δ -103.2 (d, J = 14.0 Hz, 1F), -117.7 (d, J = 14.0 Hz, 1F), -130.1 (s, 2F).

HRMS (ESI/TOF): Calcd for $C_{15}H_{13}F_4N_2O_3$ $[M+H]^+$ 345.08568, found 345.08577.

1-((3,5-Dichloro-2-fluorophenyl)amino)-3-(2,6-difluoro-3-nitrophenyl)propan-2-ol (78)



3,5-Dichloro-2-fluoroaniline (**49**) (82 mg, 0.46 mmol) and 2-(2,6-difluoro-3-nitrobenzyl)oxirane (**76**) (77 mg, 0.36 mmol) was reacted according to the general procedure for 20 hr to yield aminol **78** as a sticky colourless oil (60 mg, 42%) along with 7% recovery of epoxide.

TLC: R_f 0.35, DCM.

IR (neat): ν_{max} 3565 (OH), 3425 (NH), 3101, 2926, 2857, 1597.

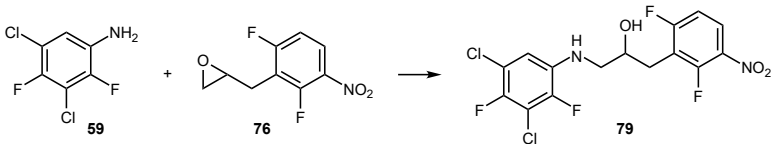
UV-vis: λ_{max} (EtOH) 254 nm (ϵ 16575 $M^{-1}cm^{-1}$).

^{1}H NMR (400.13 MHz, CD_3CN): δ 8.05 (ddd, J = 9.3 Hz, 8.5 Hz, 5.7 Hz, 1H), 7.14 (ddd, J = 9.3 Hz, 8.5 Hz, 1.9 Hz, 1H), 6.73-6.69 (m, 2H), 4.93 (bs, NH), 4.06-3.98 (m, 1H), 3.34 (d, J = 5.3 Hz, OH), 3.32 (ddd, J = 13.5 Hz, 6.7 Hz, 4.0 Hz, 1H), 3.20-3.13 (m, 1H), 2.99-2.87 (m, 2H).

^{13}C NMR (100.61 MHz, CD_3CN): δ 165.5 (dd, J = 256 Hz, 8 Hz), 156.2 (dd, J = 263 Hz, 10 Hz), 146.8 (d, J = 239 Hz), 139.9 (d, J = 12 Hz), 135.5, 130.5 (d, J = 4 Hz), 126.7 (dd, J = 12 Hz, 2 Hz), 121.4 (d, J = 16 Hz), 118.6 (dd, J = 22 Hz, 19 Hz), 116.5 (d, J = 1 Hz), 112.6 (dd, J = 25 Hz, 4 Hz), 111.6 (d, J = 4 Hz), 69.3, 49.5, 29.1.

^{19}F NMR (376.46 MHz, CD_3CN): δ -103.0 (d, J = 13.6 Hz, 1F), -117.5 (d, J = 13.6 Hz, 1F), -141.7 (s, 1F).
HRMS (ESI/TOF): Calcd for $C_{15}H_{12}Cl_2F_3N_2O_3$ $[M+H]^+$ 395.01716, found 395.01724.

1-((3,5-Dichloro-2,4-difluorophenyl)amino)-3-(2,6-difluoro-3-nitrophenyl)propan-2-ol (79)



3,5-Dichloro-2,4-difluoroaniline (**59**) (65 mg, 0.33 mmol) and 2-(2,6-difluoro-3-nitrobenzyl)oxirane (**76**) (71 mg, 0.33 mmol) was reacted according to the general procedure for 22 hr to yield aminol **79** as a white solid (45 mg, 33%, mp. 110-112 °C) along with 46% recovery of epoxide.

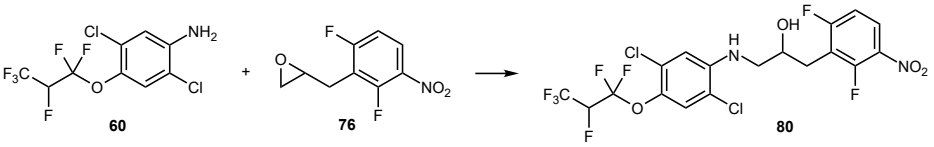
TLC: R_f 0.33, DCM.
IR (neat): ν_{max} 3433 (OH), 3382 (NH), 3103, 2935, 2857, 1625.
UV-vis: λ_{max} (EtOH) 242 nm (ϵ 16289 $M^{-1}cm^{-1}$).
^{1}H NMR (400.13 MHz, CD_3CN): δ 8.04 (ddd, J = 9.2 Hz, 8.6 Hz, 5.7 Hz, 1H), 7.13 (ddd, J = 9.2 Hz, 8.5 Hz, 1.8 Hz, 1H), 6.79 (dd, J = 8.6 Hz, 7.1 Hz, 1H), 4.72 (bs, NH), 4.05-3.97 (m, 1H), 3.34 (d, J = 5.3 Hz, OH), 3.92 (ddd, J = 13.4 Hz, 6.6 Hz, 4.1 Hz, 1H), 3.16-3.10 (m, 1H), 2.98-2.87 (m, 2H).
^{13}C NMR (100.61 MHz, CD_3CN): δ 165.5 (dd, J = 256 Hz, 8

Hz), 156.2 (dd, J = 263 Hz, 10 Hz), 146.9 (dd, J = 242 Hz, 1 Hz), 146.3 (dd, J = 237 Hz, 2 Hz), 135.7 (dd, J = 12 Hz, 3 Hz), 135.4 (dd, J = 8 Hz, 3 Hz), 126.7 (d, J = 12 Hz), 118.5 (dd, J = 22 Hz, 20 Hz), 117.3 (dd, J = 18 Hz, 4 Hz), 112.6 (dd, J = 25 Hz, 4 Hz), 111.4-111.0 (m, 1C), 111.2 (d, J = 4 Hz), 69.4, 49.7, 29.1.

^{19}F NMR (376.46 MHz, CD_3CN): δ -103.0 (d, J = 14.0 Hz, 1F), -117.5 (d, J = 14.0 Hz, 1F), -133.9 (d, J = 4.0 Hz, 1F), -137.0 (d, J = 4.0 Hz, 1F).

HRMS (ESI/TOF): Calcd for $C_{15}H_{11}Cl_2F_4N_2O_3$ $[M+H]^+$ 413.00774, found 413.00790.

1-((2,5-Dichloro-4-(1,1,2,3,3,3-hexafluoropropoxy)phenyl)-amino)-3-(2,6-difluoro-3-nitrophenyl)propan-2-ol (80)



2,5-Dichloro-4-(1,1,2,3,3,3-hexafluoropropoxy)aniline (**60**) (151 mg, 0.46 mmol) and 2-(2,6-difluoro-3-nitrobenzyl)oxirane (**76**) (100 mg, 0.46 mmol) was reacted according to the general procedure for 18 hr to yield aminol **80** as a sticky colourless oil (104 mg, 42%) along with 16% recovery of epoxide.

TLC: R_f 0.36, DCM.

IR (neat): ν_{max} 3550 (OH), 3411 (NH), 3103, 2937, 1624.

UV-vis: λ_{max} (EtOH) 254 nm (ϵ 25248 $M^{-1}cm^{-1}$).

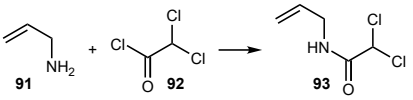
^{1}H NMR (850.13 MHz, CD_3CN): δ 8.06-8.04 (m, 1H), 7.33 (s, 1H), 7.15-7.13 (m, 1H), 6.86 (s, 1H), 5.55 (dsxt, J = 42.7 Hz, 5.8 Hz, 1H), 5.04 (t, J = 5.7 Hz, NH), 4.07-4.04 (m, 1H), 3.39 (d, J = 5.2 Hz, OH), 3.36 (ddd, J = 13.3 Hz, 6.4 Hz, 4.1 Hz, 1H), 3.22-3.19 (m, 1H), 2.97 (dd, J = 13.9 Hz, 4.8 Hz, 1H), 2.93 (dd, J = 13.9 Hz, 8.3 Hz, 1H).

^{13}C NMR (213.77 MHz, CD_3CN): δ 165.5 (dd, J = 256 Hz, 8 Hz), 156.2 (dd, J = 263 Hz, 10 Hz), 145.2, 135.5 (d, J = 5 Hz), 134.5, 127.9, 126.7 (d, J = 12 Hz), 125.6, 121.3 (qd, J = 281 Hz, 25 Hz), 119.1 (td, J = 271 Hz, 23 Hz), 118.5 (dd, J = 22 Hz, 19 Hz), 117.7, 112.73-112.60 (m, 1C), 112.71, 85.4 (dsxt, J = 198 Hz, 35 Hz), 69.3, 49.6, 29.2.

^{19}F NMR (376.46 MHz, CD_3CN): δ -75.6--75.7 (m, 3F), -78.5--80.4 (m, 2F), -102.9 (d, J = 14.0 Hz, 1F), -117.4 (d, J = 14.0 Hz, 1F), -213.3 (sxt, J = 11.2 Hz, 1F).

HRMS (ESI/TOF): $C_{18}H_{13}Cl_2F_8N_2O_4$ $[M+H]^+$ 543.01191, found 543.01196.

N-Allyl-2,2-dichloroacetamide (93)



A stirred solution of allyl amine (**91**) (1.90 mL, 25.4 mmol) in anhydrous DCM (20 mL) under Ar was cooled (ice/water bath) followed by simultaneous dropwise addition of dichloroacetyl chloride (**92**) (2.70 mL, 28.0 mmol) in anhydrous DCM (5 mL) and Et_3N (3.91

mL, 28.0 mmol) in anhydrous DCM (5 mL) over a period of 40 min. The reaction mixture was stirred at ambient temperature for 1 hr and diluted with DCM (30 mL). The phases were separated and the organic layer was washed with water (25 mL), 2% aq. HCl solution (2 x 25 mL), sat. aq. $NaHCO_3$ solution (2 x 25 mL), water (25 mL), dried ($MgSO_4$), filtered and concentrated *in vacuo* to yield compound **93** as transparent oily liquid (4.20 g, 98%), which was essentially pure based on ^{1}H NMR. Spectroscopic data are in accordance with data previously reported in the literature.[126]

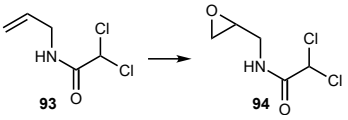
TLC: R_f 0.38, pet. ether:EtOAc, 7:3.

IR (neat): ν_{max} 3282, 3082, 3009, 2925, 1669, 1523.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 6.63 (bs, NH), 5.95 (s, 1H), 5.91-5.81 (m, 1H), 5.29-5.20 (m, 2H), 3.97-3.94 (m, 2H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 164.1, 132.7, 117.5, 66.5, 42.6.

2,2-Dichloro-*N*-(oxiran-2-ylmethyl)acetamide (94)



A stirred solution of *N*-allyl-2,2-dichloroacetamide (**93**) (3.73 g, 22.2 mmol) in anhydrous DCM (15 mL) under Ar was cooled (ice/water bath), followed by addition of *m*CPBA (6.00 g, 26.8 mmol). The reaction mixture was stirred at ambient temperature for 2 hr and rt

for 19 hr, before quenching with sat. aq. $NaHCO_3$ (20 mL) and 10% aq. $Na_2S_2O_3$ solution (20 mL). The phases were separated and the organic layer was extracted with DCM (3 x 20 mL) and washed with 1:1 sat. aq. $NaHCO_3$:$Na_2S_2O_3$ (40 mL) and sat. aq. $NaHCO_3$ (2 x 30 mL). The combined aq layers were back-extracted with DCM (3 x 20 mL) and the combined organic layers were washed with brine (30 mL), dried ($MgSO_4$), filtered, and concentrated *in vacuo* to yield compound **94** as an off-white solid (3.16 g, 77%, mp. 54-55 °C), which was essentially pure based on ^{1}H NMR.

TLC: R_f 0.15, pet. ether:EtOAc, 7:3.

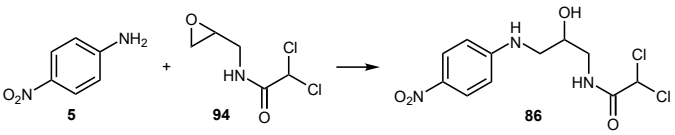
IR (neat): ν_{max} 3278, 3095, 3006, 2933, 1671, 1555.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 6.72 (bs, NH), 5.94 (s, 1H), 3.78 (ddd, J = 14.6 Hz, 5.8 Hz, 2.9 Hz, 1H), 3.44 (ddd, J = 14.6 Hz, 6 1H), 3.19-3.15 (m, 1H), 2.83 (m, 1H), 2.64 (dd, J = 4.5 Hz, 2.6 Hz, 1H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 164.7, 66.3, 50.1, 45.0, 41.1.

HRMS (ESI/TOF): Calcd for $C_5H_7NO_2Cl_2Na$ $[M+Na]^+$ 205.9746, found 205.9747.

2,2-Dichloro-*N*-(2-hydroxy-3-((4-nitrophenyl)amino)-propyl)acetamide (86)



An oven-dried 25 mL round-bottom flask was charged with lithium perchlorate (2.13 g, 20.0 mmol) and diethyl ether (4 mL). The mixture was stirred for 30 min followed by addition of 2,2-dichloro-*N*-(oxiran-2-ylmethyl)acetamide (**94**) (0.37 g, 2.00 mmol). After stirring for 10 min, *p*-nitroaniline (**5**) (0.30 g, 2.20 mmol) was added and the reaction mixture was stirred at reflux (oil bath, 60 °C) for 16 hr. Diethyl ether (5 mL) was added followed by dropwise addition of water (15 mL). The phases were separated and the aq. layer was extracted with DCM (3 x 10 mL) and EtOAc (3 x 10 mL). The combined organic phases were dried ($MgSO_4$), filtered, and evaporated on celite. The product was isolated by autoflash chromatography (pet. ether:EtOAc:DCM, 7:1:2 → 1:7:2) and concentration of the relevant fractions yielded **86** as a yellow solid (0.52 g, 81%, mp. 107-110 °C).

TLC: R_f 0.27, pet. ether:EtOAc:DCM, 2:6:2.

IR (neat): ν_{max} 3500, 3362, 3284, 3086, 2933, 1679.

UV-vis: λ_{max} (EtOH) 382 nm (ϵ 17264 $M^{-1}cm^{-1}$).

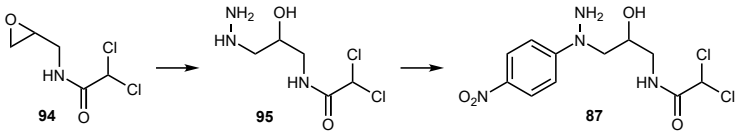
^{1}H NMR (400.13 MHz, CD_3CN): δ 8.02 (d, J = 9.2 Hz, 2H), 7.22 (bs, NH), 6.65 (d, J = 9.2 Hz, 2H), 6.16 (s, 1H), 5.70 (bs, NH),

3.93-3.86 (m, 1H), 3.48 (d, J = 5.1 Hz, OH), 3.40 (ddd, J = 13.8 Hz, 6.0 Hz, 4.7 Hz, 1H), 3.32-3.26 (m, 2H), 3.17 (ddd, J = 13.8 Hz, 7.1 Hz, 5.9 Hz, 1H).

^{13}C NMR (100.61 MHz, CD_3CN): δ 165.5, 155.3, 138.3, 127.1, 112.1, 69.0, 67.8, 47.3, 44.5.

HRMS (ESI/TOF): Calcd for $C_{11}H_{13}N_3O_4Cl_2Na$ $[M+Na]^+$ 344.0175, found 344.0179.

2,2-Dichloro-*N*-(2-hydroxy-3-(1-(4-nitrophenyl)hydrazineyl)-propyl)acetamide (87)



To a solution of 2,2-dichloro-*N*-(oxiran-2-ylmethyl)acetamide (**94**) (552 mg, 3.00 mmol) in MeOH (3 mL) was added hydrazine monohydrate (0.15 mL, 3.15 mmol) and the resulting reaction mixture was stirred at rt. under Ar for 6 hr. Following concentration under reduced pressure, excess hydrazine was azeotropically removed with water (2 mL), which in turn was azeotropically removed with toluene (2 x 5 mL). The residue was dissolved in anhydrous DMF (5 mL), followed by addition of 1-fluoro-4-nitrobenzene (**30**) (381 mg, 2.70 mmol) and Et_3N (0.42 mL, 3.00 mmol), and stirred at 40 °C under Ar for 18 hr. The reaction mixture was evaporated on celite and compound **87** was isolated by column chromatography and subsequent recrystallization in EtOAc (5 mL) as a yellow crystalline solid

(73 mg, 6%, mp. 178-179 °C).

TLC: R_f 0.07, DCM:MeOH, 99:1.

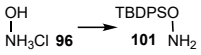
IR (neat): ν_{max} 3407, 3278, 3155, 3110, 2938, 2872, 1675.

^{1}H NMR (400.13 MHz, CD_3CN): δ 8.05 (d, J = 9.5 Hz, 2H), 7.26 (bs, NH), 7.01 (d, J = 9.5 Hz, 2H), 6.17 (s, 1H), 4.33 (s, NH_2), 4.16-4.08 (m, 1H), 3.66 (d, J = 5.1 Hz, OH), 3.63 (dd, J = 14.8 Hz, 3.8 Hz, 1H), 3.55 (dd, J = 14.8 Hz, 8.3 Hz, 1H), 3.40 (ddd, J = 13.8 Hz, 6.0 Hz, 4.6 Hz, 1H), 3.30 (ddd, J = 13.8 Hz, 6.5 Hz, 6.0 Hz, 1H).

^{13}C NMR (100.61 MHz, CD_3CN): δ 165.4, 157.2, 138.1, 126.6, 111.6, 68.7, 67.8, 58.3, 44.6.

HRMS (ESI/TOF): Calcd for $C_{11}H_{14}N_4O_4Cl_2Na$ $[M+Na]^+$ 359.02843, found 359.02861.

O-(*tert*-Butyldiphenylsilyl)hydroxylamine (101)

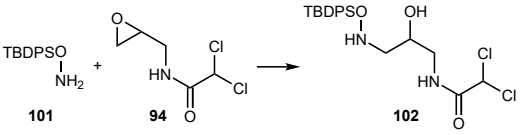


A suspension of hydroxylammonium chloride (**96**) (2.00 g, 29.0 mmol) and ethylenediamine (1.74 mL, 29.0 mmol) in anhydr. DCM (25 mL) was stirred at rt. under Ar for 4 hr. TDBPSCl (8.00 g, 29.0 mmol) was added dropwise over a period of 5 min, and the reaction mixture was stirred at rt for 64 hr. Solid ethylenediamine dihydrochloride was filtered off, the filtrate was concentrated under reduced pressure, re-dissolved in Et_2O (50 mL), and filtered again. The etheral solution

was dried ($MgSO_4$, filtered, and concentrated under reduced pressure to yield compound **101** as a white solid (6.83 g, 87%). Spectroscopic data are in accordance with previously reported literature.[127]

IR (neat): ν_{max} 3328, 3072, 2928, 2856, 1426.
^{1}H NMR (400.13 MHz, $(CD_3)_2SO$): δ 7.70-7.68 (m, 4H), 7.43-7.37 (m, 6H), 6.14 (s, 2H), 1.00 (s, 9H).
^{13}C NMR (100.61 MHz, $(CD_3)_2SO$): δ 135.1, 134.2, 129.4, 127.5, 27.2, 18.8.

N-(3-(((*tert*-Butyldiphenylsilyl)oxy)amino)-2-hydroxypropyl)-2,2-dichloroacetamide (102)



An oven-dried 10 mL round-bottom flask was charged with lithium perchlorate (1.64 g, 10.0 mmol) and diethyl ether (4 mL). The mixture was stirred for 30 min followed by addition of 2,2-dichloro-*N*-(oxiran-2-ylmethyl)acetamide (**94**) (184 mg, 1.00 mmol) and TBDPS hydroxylamine (**101**) (271 mg, 1.00 mmol), and the reaction mixture was stirred at 40 °C) for 3 hr. DCM (5 mL) was added followed by dropwise addition of water (15 mL). The phases were separated and the aq. layer was extracted with DCM (3 x 10 mL) and concentrated on celite. The product was isolated by column chromatography (pet. ether:EtOAc, 8:2 → 7:3) and concentration of the relevant fractions

yielded **102** as a colorless oil (347 mg g, 76%).

TLC: R_f 0.20, pet. ether:EtOAc, 7:3.

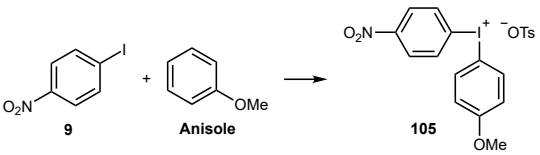
IR (neat): ν_{max} 3406 (NH), 3312 (OH), 3071, 3051, 2954, 2931, 2892, 2857, 1676 (C=O), 1528.

^{1}H NMR (500.13 MHz, $CDCl_3$): δ 7.73-7.69 (m, 4H, ArH), 7.48-7.39 (m, 6H, ArH), 6.85 (bs, NH), 5.87 (s, 1H), 3.90-3.86 (m, 1H), 3.42 (ddd, J = 13.9 Hz, 6.2 Hz, 3.9 Hz, 1H), 3.18 (ddd, J = 13.9 Hz, 6.2 Hz, 5.7 Hz, 1H), 3.02 (dd, J = 13.6 Hz, 3.0 Hz, 1H), 2.77 (dd, J = 13.6 Hz, 8.2 Hz, 1H), 1.10 (s, 9H).

^{13}C NMR (125.76 MHz, $CDCl_3$): δ 164.8, 135.9, 135.8, 134.9, 133.1, 133.0, 130.2, 130.1, 128.0, 127.9, 127.8, 68.1, 66.4, 56.8, 43.6, 27.4, 19.2.

HRMS (ESI/TOF): Calcd for $C_{21}H_{28}N_2O_3Cl_2SiNa$ $[M+Na]^+$ 477.11385, found 477.11468.

4-Anisyl-(4-nitrophenyl)iodonium tosylate (105)



1-Iodo-4-nitrobenzene (**9**) (2.49 g, 10.0 mmol) and *m*CPBA (2.69 g, 12.0 mmol, 77% active oxidant) was dissolved in $CHCl_3$ (100 mL) and stirred at rt. for 10 min, followed by addition of *p*-TSA monohydrate (2.28 g, 12.0 mmol) and anisole (6.0 mL, 55.0 mmol). The resulting reaction mixture was stirred at 40 °C for 2.5 hr, the volatiles

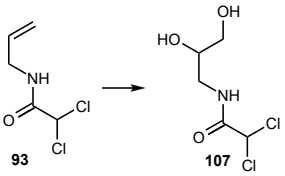
were removed under reduced pressure, and the residue was added diethyl ether (20 mL). The precipitated solid was filtered, washed with ether (3 x 20 mL), and redissolved in MeOH (100 mL at 50 °C). Diethyl ether was added gradually (100 mL) until small amounts of precipitate formed and the beaker was placed in the frigde to crystallize overnight. The solvent was decanted off and the solid was washed with ether (3 x 50 mL) to yield compound **105** as a white crystalline solid (1.18 g, 22%). Spectroscopic data are in accordance with previously reported literature.[111]

IR (neat): ν_{max} 3090, 3066, 3032, 2968, 2917, 1570.

^{1}H NMR (500.13 MHz, CD_3OD): δ 8.33 (d, J = 9.2 Hz, 2H), 8.28 (d, J = 9.2 Hz, 2H), 8.14 (d, J = 9.2 Hz), 7.68 (d, J = 8.2 Hz, 2H), 7.21 (d, J = 8.2 Hz, 2H), 7.08 (d, J = 9.2 Hz, 2H), 3.86 (s, 3H), 2.36 (s, 3H).

^{13}C NMR (125.76 MHz, CD_3OD): δ 164.9, 151.4, 143.6, 141.7, 139.0, 137.1, 129.8, 127.4, 127.0, 122.2, 119.1, 104.5, 56.4, 21.3.

2,2-Dichloro-*N*-(2,3-dihydroxypropyl)acetamide (107)



A 100 mL round-bottom flask was charged K_2OsO_4 x $2H_2O$ (0.22 g, 5 mol%), $K_3Fe(CN)_6$ (11.9 g, 36.0 mmol), K_2CO_3 (4.99 g, 36.0

mmol), *t*-butanol (25 mL), water (25 mL), and the resulting mixture was stirred for 15 min. *N*-Allyl-2,2-dichloroacetamide (**93**) (2.00 g, 12.0 mmol) was added and the reaction mixture was stirred at rt for 2 hr, followed by quenching with Na_2SO_3 (5.00 g). The crude mixture was extracted with EtOAc (4 x 50 mL), and the organic layer was washed with brine (50 mL), dried (Na_2SO_4, filtered, and concentrated *in vacuo* to yield compound **107** as an off-white fused mass (2.20 g, 92%, mp. 55-57 °C), which was essentially pure by ^{1}H NMR.

TLC: R_f 0.23, EtOAc.

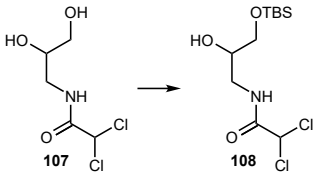
IR (neat): ν_{max} 3315, 3086, 2926, 1672.

^{1}H NMR (400.13 MHz, CD_3OD): δ 6.29 (s, 1H), 3.76-3.71 (m, 1H), 3.52 (dd, J = 11.4 Hz, 5.1 Hz, 1H), 3.49 (dd, J = 11.4 Hz, 5.7 Hz, 1H), 3.43 (dd, J = 13.7 Hz, 4.8 Hz, 1H), 3.25 (dd, J = 13.7 Hz, 7.0 Hz, 1H).

^{13}C NMR (100.61 MHz, CD_3OD): δ 167.0, 71.5, 67.6, 65.1, 44.1.

HRMS (ESI/TOF): Calcd for $C_5H_9NO_3Cl_2Na$ $[M+Na]^+$ 223.98517, found 223.98613.

N-(3-((*tert*-Butyldimethylsilyl)oxy)-2-hydroxypropyl)-2,2-dichloroacetamide (108)



To an oven-dried 250 mL round-bottom flask containing diol **107** (10.1 g, 50.0 mmol) dissolved in anhydr. DCM (50 mL), was added TBSCl (8.29 g, 55.0 mmol), DMAP (0.31 g, 5 mol%), and Et_3N (7.7 mL, 55.0 mmol). The reaction mixture was stirred at rt. under Ar for 17 hr before quenching with sat. aq. NH_4Cl solution. The phases were separated and the aq. phase was extracted with DCM (3 x 20 mL). The combined organic extracts were washed with brine (40 mL), dried ($MgSO_4$), filtered, and concentrated under reduced pressure to yield **108** an off-white powder (15.7 g, 99%, mp. 77-78 °C), which was essentially pure based on ^{1}H NMR.

TLC: R_f 0.23, pet. ether:EtOAc, 7:3.

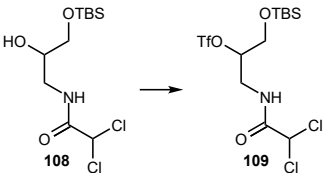
IR (neat): ν_{max} 3325, 3251, 3101, 2928, 2856, 1667.

^{1}H NMR (400.13 MHz, CD_3OD): δ 6.29 (s, 1H), 3.77-3.71 (m, 1H), 3.63 (dd, J = 10.4 Hz, 5.2 Hz, 1H), 3.58 (dd, J = 10.4 Hz, 5.9 Hz, 1H), 3.47 (dd, J = 13.6 Hz, 4.5 Hz, 1H), 3.24 (dd, J = 13.6 Hz, 7.1 Hz, 1H), 0.92 (s, 9H), 0.09 (s, 6H).

^{13}C NMR (100.61 MHz, CD_3OD): δ 166.9, 71.5, 67.6, 66.5, 44.2, 26.4, 19.2, 5.3, 5.3.

HRMS (ESI/TOF): Calcd for $C_{11}H_{23}NO_3Cl_2SiNa$ $[M+Na]^+$ 338.07165, found 338.07266.

1-((*tert*-Butyldimethylsilyl)oxy)-3-(2,2-dichloroacetamido)-propan-2-yl trifluoromethanesulfonate (109)



To an oven-dried 100 mL round-bottom flask containing alcohol **108** (950 mg, 3.00 mmol) dissolved in anhydr. DCM (20 mL), was added Tf_2NPh (1180 mg, 3.30 mmol), DMAP (37 mg, 10 mol%), and Et_3N (1.25 mL, 9.00 mmol). The reaction mixture was stirred at rt. under Ar for 18 hr and concentrated under reduced pressure on celite. The product was isolated by silica-gel column chromatography (pet. ether:EtOAc, 8:2) and concentration of the relevant fractions yielded **109** as a slightly yellow oily liquid (718 mg, 53%).

TLC: R_f 0.40, pet. ether:EtOAc, 7:3.

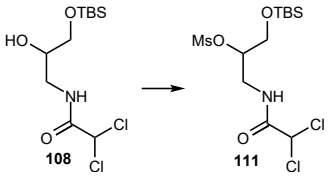
^{1}H NMR (400.13 MHz, $CDCl_3$): δ 6.22 (s, 1H), 4.84-4.77 (m, 1H), 3.97 (dd, J = 14.8 Hz, 9.8 Hz, 1H), 3.88 (dd, J = 14.8 Hz, 7.4 Hz, 1H), 3.83 (dd, J = 11.4 Hz, 3.8 Hz, 1H), 3.69 (dd, J = 11.4 Hz, 4.0 Hz, 1H), 0.88 (s, 9H), 0.08 (s, 3H), 0.07 (s, 3H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 162.3, 82.0, 63.7, 61.6, 56.3, 25.9, 18.4, -5.3, -5.3. CF_3 signal missing.

^{19}F NMR (376.46 MHz, $CDCl_3$): δ -74.1.

LRMS (ESI): Calcd for $C_{11}H_{22}NO_2SiCl_2$ $[M\text{-}OTf]^+$ 298.08, found 297.84.

1-((*tert*-Butyldimethylsilyl)oxy)-3-(2,2-dichloroacetamido)-propan-2-yl methanesulfonate (111)



A solution of alcohol **108** (1898 mg, 6.00 mmol) and Et_3N (1.67 mL, 12.0 mmol) in anhydrous DCM (15 mL) was cooled (ice/water bath), followed by dropwise addition of MsCl (0.56 mL, 7.20 mmol). The reaction mixture was stirred at 0 °C for 30 min, then at rt for 30 min, before quenching with water (10 mL). The phases were separated and the aqueous layer was extracted with DCM (3 x 15 mL). The combined organic phases were washed with 0.1 M HCl solution (2 x 20 mL), water (20 mL), dried (Na_2SO_4), filtered, and concentrated to yield mesylate **111** as a colorless oily liquid (2184 mg, 92%).

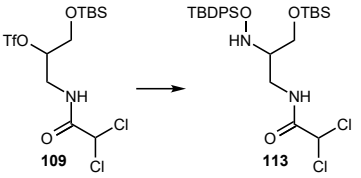
TLC: R_f 0.33, pet. ether:EtOAc, 7:3.

^{1}H NMR (500.13 MHz, $CDCl_3$): δ 7.16 (t, J = 5.3 Hz, NH), 5.96 (s, 1H), 4.79-4.75 (m, 1H), 3.85-3.79 (m, 2H), 3.75 (ddd, J = 14.6 Hz, 6.4 Hz, 3.7 Hz, 1H), 3.53 (ddd, J = 14.6 Hz, 7.0 Hz, 5.6 Hz, 1H), 3.08 (s, 3H), 0.89 (s, 9H), 0.09 (s, 3H), 0.08 (s, 3H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 164.8, 80.4, 66.3, 63.5, 41.4, 38.5, 25.9, 18.4, -5.4.

LRMS (ESI): Calcd for $C_{12}H_{24}Cl_2NO_5SSi$ $[M\text{-}H]^-$ 392.05, found 392.23.

N-(3-((*tert*-Butyldimethylsilyl)oxy)-2-(((tert-butyldiphenylsilyl)oxy)amino)propyl)-2,2-dichloroacetamide (113)



Triflate **109** and TBDPS-hydroxylamine (**101**) was stirred at 40 °C under Ar in anhydrous THF (1.0 mL) for 42 hr. THF was removed under reduced pressure and the residue was subjected to silica-gel autoflash chromatography (pet. ether:EtOAc:DCM, 96:1:3 $\rightarrow$ 89:8:3) to yield compound **113** (80 mg, 70%) as a colorless oil.

TLC: R_f 0.39, pet. ether:EtOAc, 9:1.

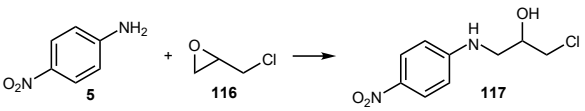
^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.71-7.69 (m, 4H), 7.44-7.35 (m, 6H), 6.13/6.12 (s, 1H, appears as two singlets due to rotamer/tautomer), 5.95 (t, J = 6.0 Hz, NH), 3.97-3.91 (m, 1H), 3.86 (ddd, J = 13.2 Hz, 7.4 Hz, 3.9 Hz, 1H), 3.76 (dd, J = 10.2 Hz, 4.0 Hz, 1H), 3.66 (dd, J = 10.2 Hz, 6.5 Hz, 1H), 3.58 (ddd, J = 13.2 Hz, 7.4 Hz, 5.2 Hz, 1H), 1.11 (s, 9H), 0.94 (s, 9H), 0.11 (s, 6H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 154.2, 135.5, 133.4, 129.8,

127.7, 71.2, 66.3, 64.8, 44.8, 27.2, 26.0, 19.5, 18.4, -5.23, -5.27.

LRMS (ESI): Calcd for $C_{27}H_{43}Cl_2N_2O_3Si_2$ $[M+H]^+$ 569.22, found 569.31.

1-Chloro-3-((4-nitrophenyl)amino)propan-2-ol (117)



(±)-Epichlorohydrin (**116**) (0.75 mL, 9.45 mmol) and *p*-nitroaniline (**5**) (1.24 g, 9.00 mmol) in 5 M lithium perchlorate solution (9 mL) was reacted according to the general procedure for 21 hr. Isolation by silica-gel flash chromatography (DCM) and concentration of the relevant fractions yielded **117** as a yellow waxy oily liquid (1.91 g, 92%). Spectroscopic data are in accordance with previously reported literature.[128]

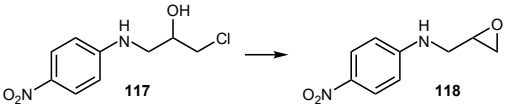
TLC: R_f 0.35, DCM:MeOH 95:5.

IR (neat): ν_{max} 3472 (OH), 3368 (NH), 3083, 2920, 1593.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.09 (d, J = 9.2 Hz, 2H), 6.60 (d, J = 9.2 Hz, 2H), 4.15-4.10 (m, 1H), 3.70 (dd, J = 11.3 Hz, 4.6 Hz, 1H), 3.64 (dd, J = 11.3 Hz, 6.1 Hz, 1H), 3.49 (dd, J = 13.4 Hz, 4.0 Hz, 1H), 3.34 (dd, J = 13.4 Hz, 7.3 Hz, 1H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 153.1, 138.8, 126.6, 111.7, 69.8, 47.4, 46.3.

4-Nitro-*N*-(oxiran-2-ylmethyl)aniline (118)



Sodium hydride (53 mg, 1.32 mmol, 60% dispersion in mineral oil) was added to a stirred solution of 1-chloro-3-((4-nitrophenyl)amino)-propan-2-ol (**117**) (277 mg, 1.20 mmol) in anhydr. THF (5 mL) at 0 °C (ice/water bath). The resulting reaction mixture was stirred for 5 min at 0 °C before adding sat. aq. NH_4Cl (10 mL). The phases were separated and the aq. layer was extracted with DCM (3 x 10 mL). The combined organic phases were dried (Na_2SO_4), filtered, and concentrated under reduced pressure to yield **118** as a yellow crystalline solid (227 mg, 97%, mp. 78-79 °C), which was used without further purification. Spectroscopic data are in accordance with previously reported literature.[129]

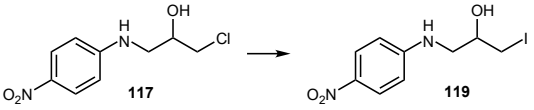
TLC: R_f 0.29, DCM:MeOH, 95:5.

IR (neat): ν_{max} 3360 (NH), 3189, 3074, 3015, 2899, 1595.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.08 (d, J = 9.2 Hz, 2H), 6.59 (d, J = 9.2 Hz, 2H), 4.74 (bs, NH), 3.68 (dd, J = 14.3 Hz, 2.7 Hz, 1H), 3.31 (dd, J = 14.3 Hz, 5.1 Hz, 1H), 3.24-3.21 (m, 1H), 2.86 (dd, J = 4.6 Hz, 4.1 Hz, 1H), 2.68 (dd, J = 4.6 Hz, 2.6 Hz, 1H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 153.2, 138.7, 126.5, 111.5, 50.5, 45.2, 44.4.

1-Iodo-3-((4-nitrophenyl)amino)propan-2-ol (119)



A sealed reactor tube was charged with 1-chloro-3-((4-nitrophenyl)-amino)propan-2-ol (**117**) (98 mg, 0.42 mmol), sodium iodide (315 mg, 2.10 mmol), and acetone (1.5 mL). The reaction mixture was irradiated in the microwave reactor at 120 °C for 10 min. Acetone was removed on a rotary evaporator, the residue was dissolved in DCM (20 mL) and precipitated sodium chloride was removed by filtering through paper. Concentration of the filtrate yielded compound **119** as a yellow crystalline solid (120 mg, 89%, mp. 85-87 °C).

TLC: R_f 0.41, DCM:MeOH, 95:5.

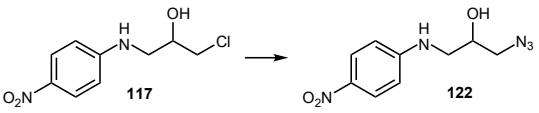
IR (neat): ν_{max} 3470 (OH), 3316 (NH), 3076, 2921, 2851.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.07 (d, J = 9.2 Hz, 2H), 6.59 (d, J = 9.2 Hz, 2H), 3.92-3.87 (m, 1H), 3.52 (dd, J = 13.4 Hz, 3.9 Hz, 1H), 3.39 (dd, J = 10.4 Hz, 4.9 Hz, 1H), 3.32 (dd, J = 10.4 Hz, 6.3 Hz, 1H), 3.31 (dd, J = 13.4 Hz, 7.6 Hz, 1H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 153.1, 138.6, 126.6, 111.7, 69.6, 48.1, 10.9.

HRMS: Sample sent for analysis.

1-Azido-3-((4-nitrophenyl)amino)propan-2-ol (122)



A solution of chlorohydrin **117** (1.75 g, 7.60 mmol), sodium azide (0.99 g, 15.2 mmol), TBAI (140 mg, 5 mol%) in methanol (10 mL) and water (40 mL) was refluxed (oil, 110 °C) for 18 hr. The reaction mixture was cooled to rt, extracted with EtOAc (4 x 25 mL), and the combined organic layers were dried ($MgSO_4$), filtered, and concentrated. The residue was filtered through a short plug of silica (3 cm) with the aid of EtOAc (50 mL) and removal of volatiles under reduced pressure yielded **122** as a yellow waxy liquid (1.64 g, 91%).

TLC: R_f 0.26, pet. ether:EtOAc, 50:50.

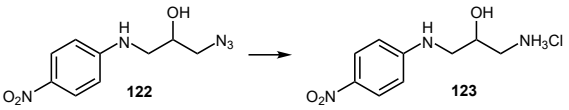
IR (neat): ν_{max} 3367 (OH, NH), 2927, 2869, 2096 (-N_3), 1595.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.08 (d, J = 9.2 Hz, 2H), 6.58 (d, J = 9.2 Hz, 2H), 4.08-4.03 (m, 1H), 3.54 (dd, J = 12.5 Hz, 4.4 Hz, 1H), 3.46 (dd, J = 12.5 Hz, 6.4 Hz, 1H), 3.40 (dd, J = 13.2 Hz, 4.2 Hz, 1H), 3.28 (dd, J = 13.2 Hz, 7.4 Hz, 1H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 153.2, 138.7, 126.6, 111.6, 69.0, 54.8, 46.4.

HRMS (ESI/TOF): Calcd for $C_9H_{11}N_5NaO_3$ $[M+Na]^+$ 260.07541, found 260.07611.

1-Amino-3-((4-nitrophenyl)amino)propan-2-ol hydrochloride (123)



A solution of azide **122** (607 mg, 2.56 mmol) and PPh_3 (740 mg, 2.82 mmol) in THF (9 mL) and water (1 mL) was stirred at 50 °C under Ar for 5 hr. THF was removed under reduced pressure, and EtOAc (20 mL) and 6 M aq. hydrochloric acid (20 mL) was added. The phases were separated and the organic layer was extracted with water (2 x 10 mL). The combined aq. phases were washed with Et_2O (40 mL) and concentrated under reduced pressure. Traces of water was azeotropically removed with toluene (3 x 5 mL) to yield **123** as sharp yellow crystals (472 mg, 74%, mp. 203-205 °C).

TLC: R_f 0.17 as freebase, DCM:MeOH:20% NH_3, 89:10:1.

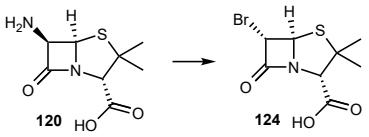
IR (neat): ν_{max} 3293, 3247, 3044, 2921, 1601.

^{1}H NMR (400.13 MHz, D_2O): δ 8.08 (d, J = 9.3 Hz, 2H), 6.73 (d, J = 9.3 Hz, 2H), 4.16-4.10 (m, 1H), 3.46 (dd, J = 14.4 Hz, 4.8 Hz, 1H), 3.37 (dd, J = 14.4 Hz, 7.0 Hz, 1H), 3.36 (dd, J = 13.1 Hz, 2.3 Hz, 1H), 3.02 (dd, J = 13.1 Hz, 10.1 Hz, 1H).

^{13}C NMR (100.61 MHz, CD_3OD): δ 155.3, 138.8, 127.2, 112.7, 67.3, 48.0, 44.0.

HRMS (ESI/TOF): Calcd for $C_9H_{13}N_3O_3$ $[M+H]^+$ 212.10297, found 212.10366.

(2*S*,5*R*,6*S*)-6-Bromo-3,3-dimethyl-7-oxo-4-thia-1-azabicyclo-[3.2.0]heptane-2-carboxylic acid (124)



6-Aminopenicillanic acid (**120**) (2.60 g, 12.0 mmol) and KBr (7.14 g, 60.0 mmol) was dissolved in EtOH (25 mL) and 2.5 N sulfuric acid (30 mL, 38.0 mmol), and cooled to 0 °C (ice/water). A solution of $NaNO_2$ (1.24 g, 18.0 mmol) in water (6 mL) was added dropwise over a period of 20 min. The resulting reaction mixture was stirred at 0 °C for 3 hr followed by addition of chloroform (50 mL) and brine (20 mL). The phases were separated and the aq. phase was extracted with chloroform (3 x 20 mL). The combined organic layers were washed with water (2 x 30 mL), dried ($MgSO_4$), filtered, and concentrated to yield **124** as a waxy oily liquid (2.81 g, 84%). Spectroscopic data are in accordance with previously reported literature.[130]

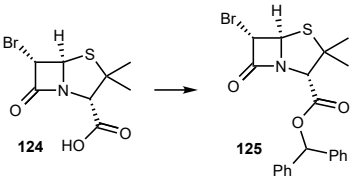
TLC: R_f 0.36, ACN:H_2O, 90:10.

IR (neat): ν_{max} 3132 (br, COOH), 2974, 2930, 1773, 1721.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 10.3 (bs, COOH), 5.38 (d, J = 1.3 Hz, 1H), 4.82 (d, J = 1.3 Hz, 1H), 4.56 (s, 1H), 1.62 (s, 3H), 1.54 (s, 3H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 171.9, 167.9, 70.4, 69.8, 65.0, 49.1, 33.6, 25.9.

Benzhydryl (2*S*,5*R*,6*S*)-6-bromo-3,3-dimethyl-7-oxo-4-thia-1-azabicyclo[3.2.0]heptane-2-carboxylate (125)



To a solution of penicillanic acid **124** (2.80 g, 10.0 mmol), diphenylmethanol (1.84 g, 10.0 mmol), and DMAP (61 mg, 5 mol%) in anhydr. DCM (50 mL) was added DIC (1.57 mL, 10.0 mmol). The resulting reaction mixture was stirred at rt. under Ar for 24 hr before adding water (20 mL) and brine (20 mL). The phases were separated and the aq. layer was extracted with DCM (3 x 40 mL). The combined organic layers were washed with water (3 x 40 mL), dried (Na_2SO_4), filtered, and concentrated under reduced pressure. The product was isolated by silica-gel flash chromatography (pet. ether:EtOAc, 95:5) and concentration of the relevant fractions yielded **125** as a sticky oil (2.80 g, 63%). Spectroscopic data are in accordance with previously reported literature.[130]

TLC: R_f 0.26, pet. ether:EtOAc, 90:10.

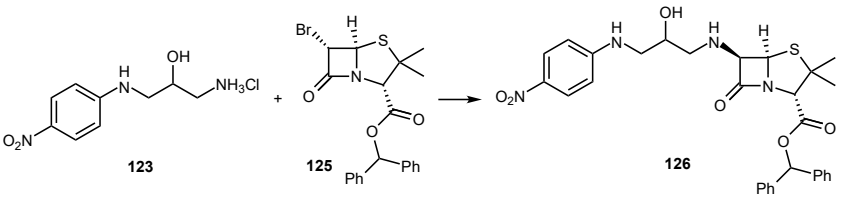
IR (neat): ν_{max} 3060, 3030, 3014, 2977, 2956, 1776, 1743.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.38-7.29 (m, 10H), 6.95 (s, 1H), 5.45 (d, J = 1.4 Hz, 1H), 4.82 (d, J = 1.4 Hz, 1H), 4.66 (s, 1H), 1.60 (s, 3H), 1.27 (s, 3H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 167.3, 166.1, 139.1, 139.0,

128.70, 128.66, 128.5, 128.3, 127.5, 127.1, 78.6, 70.7, 70.0, 65.3, 49.6, 34.2, 25.5.

Benzhydryl (2*S*,5*R*,6*R*)-6-((2-hydroxy-3-((4-nitrophenyl)-amino)propyl)amino)-3,3-dimethyl-7-oxo-4-thia-1-azabi-cyclo[3.2.0]heptane-2-carboxylate (126)



Amine **123** (84 mg, 0.34 mmol), bromide **125** (153 mg, 0.34 mmol), and K_2CO_3 (117 mg, 0.85 mmol) was stirred in anhydrous DMF (2 mL) for 15 hr at rt under Ar. DMF was removed under reduced pressure, the reaction mixture was evaporated on celite, and subjected to autoflash column chromatography (pet. ether:EtOAc, 80:20 $\rightarrow$ 0:100) to yield the target compound **126** as a yellow solid (68 mg, 35%).

TLC: R_f 0.58, EtOAc.

IR (neat): ν_{max} 3379 (OH, NH), 3060, 2923, 1733, 1630, 1597.

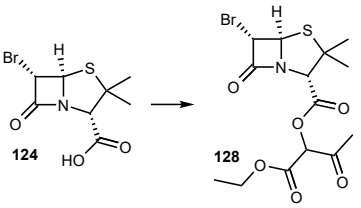
^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.99 (d, J = 8.8 Hz, 2H), 7.59 (d, J = 6.0 Hz, 1H), 7.34-7.28 (m, 11H), 6.95 (d, J = 1.8 Hz, 1H), 6.50 (d, J = 8.8 Hz, 2H), 6.29 (bs, 1H), 5.50 (bs, 1H), 4.10 (d, J = 1.8 Hz, 1H), 3.96-3.91 (m, 1H), 3.51-3.38 (m, 2H), 3.22 (dd, J = 13.2 Hz, 5.0 Hz, 1H), 3.17 (dd, J = 13.2 Hz, 6.9 Hz, 1H), 1.47

(app. s, 3H), 1.06 (app. s, 3H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 168.8, 168.1, 153.8, 139.1, 137.8, 134.91, 134.87, 128.8, 128.75, 128.69, 128.4, 127.8, 126.9, 126.5, 111.3, 91.1, 79.3, 69.9/69.8, 63.54/63.50, 46.5, 44.5/44.4, 37.8, 27.3/27.2, 23.4/23.3.

HRMS (ESI/TOF): Calcd for $C_{30}H_{33}N_4O_6S$ $[M+H]^+$ 577.21153, found 577.21123.

1-Ethoxy-1,3-dioxobutan-2-yl (2*S*,5*R*,6*S*)-6-bromo-3,3-dimethyl-7-oxo-4-thia-1-azabicyclo[3.2.0]heptane-2-carboxylate (128), diasteromeric mixture



Penicillanic acid **124** (388 mg, 1.39 mmol), ethyl 2-chloroacetoacetate (**127**) (0.22 mL, 1.60 mmol), and K_2CO_3 (105 mg, 0.76 mmol) was stirred in anhydrous DMF (3 mL) at rt under Ar for 24 hr. Water (10 mL) was added followed by extraction with DCM (3 x 15 mL). The combined organic layers were washed with water (2 x 10 mL), dried ($MgSO_4$), filtered, and concentrated under reduced pressure to yield compound **128** as a colorless oily liquid (563 mg, 88%).

TLC: R_f 0.57, pet. ether:EtOAc, 5:5.

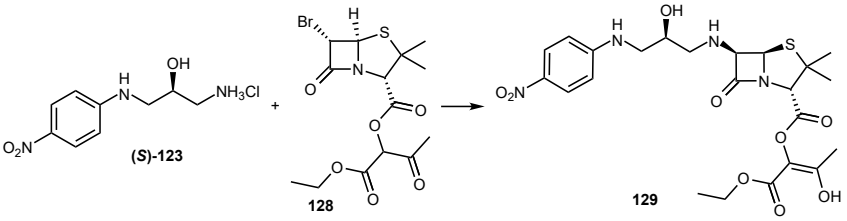
^{1}H NMR (400.13 MHz, $CDCl_3$): δ 5.55 (s, 1H), 5.44/5.43 (d, J

=1.4 Hz, 1H), 4.82 (d, J = 1.4 Hz, 1H, two overlapping doublets), 4.69 (s, 1H, two overlapping singlets), 4.34-4.27 (m, 2H), 2.39 (s, 3H), 1.68 (s, 3H), 1.60 (s, 3H, two overlapping singlets), 1.33 (t, J = 7.1 Hz, 3H, two overlapping triplets).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 196.1/196.0, 167.6/167.5, 165.8/165.7, 163.8/163.7, 78.4/78.3, 70.6/70.5, 69.9/69.9, 65.5/65.5, 63.1/63.0, 49.3/49.1, 33.7/33.5, 27.6/27.6, 25.8/25.8, 14.1.

LRMS (ESI): Calcd for $C_{14}H_{18}BrNO_6SNa$ $[M+Na]^+$ 429.99, found 429.86.

1-Ethoxy-1,3-dioxobutan-2-yl (2*S*,5*R*,6*R*)-6-(((*S*)-2-hydroxy-3-((4-nitrophenyl)amino)propyl)amino)-3,3-dimethyl-7-oxo-4-thia-1-azabicyclo[3.2.0]heptane-2-carboxylate (129)



Amine hydrochloride **(*S*)-123** (211 mg, 0.85 mmol), penicillinate **128** (361 mg, 0.88 mmol), and K_2CO_3 (122 mg, 0.88 mmol) was stirred in anhydrous DMF (2 mL) at rt under Ar for 24 hr. DMF was removed under reduced pressure, the reaction mixture was evaporated on celite, and subjected to flash column chromatography (pet. ether:EtOAc, 4:6). Concentration of the relevant fractions yielded

compound **129** as a yellow solid (262 mg, 57%).

TLC: R_f 0.42, pet. ether:EtOAc, 2:8.

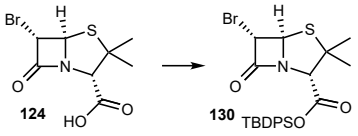
IR (neat): ν_{max} 3464 (OH), 3372 (NH), 3199, 2979, 1782, 1732.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.55 (bs, NH), 7.98 (d, J = 9.2 Hz, 2H), 6.52 (d, J = 9.2 Hz, 2H), 5.38 (d, J = 1.4 Hz, 1H), 5.27 (t, J = 5.3 Hz, NH), 4.82 (d, J = 1.4 Hz, 1H), 4.63 (s, 1H), 4.11-4.04 (m, 2H), 4.02-3.97 (m, 1H), 3.40-3.18 (m, 4H), 1.90 (s, 3H), 1.64 (s, 3H), 1.60 (s, 3H), 1.18 (t, J = 7.1 Hz, 3H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 167.8, 167.2, 165.2, 154.9, 153.5, 138.0, 126.5, 111.9, 111.4, 70.3, 70.1, 69.3, 65.3, 60.0, 48.7, 46.8, 46.6, 33.0, 25.9, 14.5, 13.3.

HRMS (ESI/TOF): Sample gave signals that could not be attributed to compound **129**. Detected 598.78582 and 644.76865.

***tert*-Butyldiphenylsilyl (2*S*,5*R*,6*S*)-6-bromo-3,3-dimethyl-7-oxo-4-thia-1-aza bicyclo[3.2.0]heptane-2-carboxylate (130)**



TBDPSCl (0.59 mL, 2.25 mmol) dissolved in anhydr. THF (5 mL) was added dropwise to a stirred solution of 6-bromopenicillanic acid (**124**) (630 mg, 2.25 mmol) and *N*-methylmorpholine (0.25 mL, 2.25 mmol) in anhydr. THF (10 mL) over a period of 5 min. The resulting reaction mixture was stirred at rt. for 48 hr followed by concen-

tration under reduced pressure. The residue was filtered through a short plug of silica (5 cm) with 1:1 pet. ether:DCM (150 mL). Concentration of the filtrate under reduced pressure yielded compound **130** as an off-white oily liquid (1125 mg, 97%).

TLC: R_f 0.59, PE:EtOAc, 5:5.

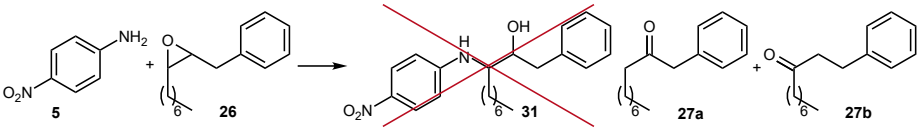
IR (neat): ν_{max} 3071, 3049, 2956, 2932, 2891, 2858, 1725 (ester and amide C=O overlapping), 1589.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.70-7.66 (m, 4H, ArH), 7.43-7.38 (m, 6H, ArH), 5.39 (d, J = 1.4 Hz, 1H), 4.81 (d, J = 1.4 Hz, 1H), 4.62 (s , 1H), 1.62 (s, 3H), 1.43 (s, 3H), 1.16 (s, 9H).

^{13}C NMR (100.61 MHz, $CDCl_3$): δ 167.6, 166.0, 135.6, 130.7, 128.1, 128.0, 127.9, 71.3, 70.6, 65.3, 49.5, 34.3, 27.1, 26.0, 19.3.

VII.2.1 Attempted syntheses

3-((4-Nitrophenyl)amino)-1-phenyldecan-2-ol (31)

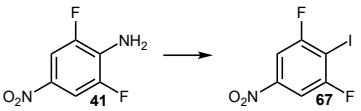


p-Nitroaniline (**5**) (152 mg, 1.10 mmol) and *cis*-Benzylheptyloxirane (**26**) (232 mg, 1.00 mmol) was refluxed in 5 M LPDE (5 mL) under Ar for 24 hr. Following extraction with DCM, ^{1}H-NMR analysis showed only starting materials. 5 M LPEtOAc was used instead and the reaction mixture was stirred at 80 °C under Ar for 48 hr. ^{1}H NMR shows a mixture of *p*-nitroaniline (**5**) and two isomeric ketones

27a and **27b** in 77:33 ratio.

^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.06 (d, J = 9.1 Hz, 2H, *p*-nitroaniline), 7.35-7.27 (m, 3H, a/b), 7.21-7.17 (m, 2H, a/b), 6.60 (d, J = 9.1 Hz, 2H, *p*-nitroaniline), 3.68 (s, 2H, a), 2.90 (t, J = 7.6 Hz, 2H, b), 2.73 (t, J = 7.6 Hz, 2H, b), 2.44 (t, J = 7.4 Hz, 2H, a), 2.38 (t, J = 7.4 Hz, 2H, b), 1.59-1.51 (m, 2H, a/b), 1.32-1.20 (m, 10H a, 8H b), 0.87 (t, J = 6.9 Hz, 3H, a/b).
^{13}C NMR (100.61 MHz, $CDCl_3$): δ 210.6 (C=O, b), 208.9 (C=O, a), 50.2 (a), 44.3 (b), 43.2 (b), 42.1 (a), 31.9 (a), 31.8 (b), 29.9 (b), 29.4 (a), 29.3 (b), 29.2 (a/b overlapping), 23.91 (b), 23.85 (a), 22.73 (a), 22.69 (b), 14.18 (a), 14.16 (b).

2,6-Difluoro-4-nitro-1-iodobenzene (67)

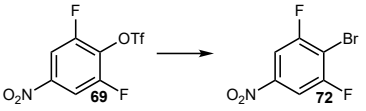


$NaNO_2$ (23 mg, 0.33 mmol) was added to a stirred solution of 2,6-difluoro-4-nitroaniline (**41**) (52 mg, 0.30 mmol) and CSA (77 mg, 0.33 mmol) in AcOH (3 mL) at rt. The resulting reaction mixture was stirred at rt for 50 minutes before adding more CSA (77 mg, 0.33 mmol) and $NaNO_2$ (46 mg, 0.66 mmol). The reaction was stirred for another 30 min, and the mixture was poured into a beaker containing water (50 mL), K_2CO_3 (1.00 g) and Na_2SO_3 (1.00 g). DCM (15 mL) was added, the phases were separated, and the aqueous

layer was extracted with DCM (3 x 10 mL). The combined organic phases were washed with sat. $NaHCO_3$ solution (20 mL) and brine (20 mL), and was concentrated under reduced pressure to yield a crude residue containing compound **67** with some impurities.

TLC: R_f 0.55, pet. ether:EtOAc, 9:1.
^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.78 (d, J = 5.3 Hz, 2H).
^{13}C NMR (100.61 MHz, $CDCl_3$): δ 162.8 (dd, J = 251.2 Hz, 6.1 Hz), 149.7 (t, J = 9.6 Hz), 107.1 (dd, J = 30.1 Hz, 3.1 Hz), 80.7 (t, J = 29.9 Hz).
^{19}F NMR (376.46 MHz, $CDCl_3$): δ -86.7.

1-Bromo-2,6-Difluoro-4-nitrobenzene (72)

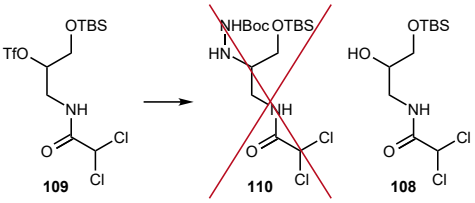


Triflate **69** (161 mg, 0.52 mmol) and TBAB (503 mg, 1.56 mmol) was refluxed in toluene (2.0 mL) under Ar for 20 hr. The reaction solution was filtered through a short plug of silica (5 cm) with the aid of pet. ether:EtOAc (80:20, 50 mL), the filtrate was concentrated under reduced pressure, and analysed with ^{1}H and ^{19}F NMR. Trace amounts of compound **72** was detected in a mixture concisting of mainly TBAB. The reaction was scaled up 4 times and a purification with silica-gel flash column chromatography (pet.

ether:EtOAC, 99:1), yielded compound **72** (73 mg, 14%). The reaction was repeated two more times, with isolated yields of 0.3% and 6%. Sufficient amounts were not obtained.

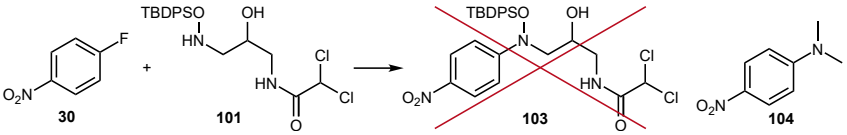
TLC: R_f 0.29, pet. ether:EtOAc, 95:5.
^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.87 (d, J = 5.6 Hz, 2H).
^{13}C NMR (100.61 MHz, $CDCl_3$): δ 159.9 (dd, J = 253.7 Hz, 4.3 Hz), 147.8 (t, J = 10.0 Hz), 108.0 (dd, J = 28.2 Hz, 3.1 Hz), 106.7 (t, J = 24.6 Hz).
^{19}F NMR (376.46 MHz, $CDCl_3$): δ -99.2.

tert-Butyl 2-(1-((*tert*-butyldimethylsilyl)oxy)-3-(2,2-dichloroacetamido)propan-2-yl)hydrazine-1-carboxylate (110)

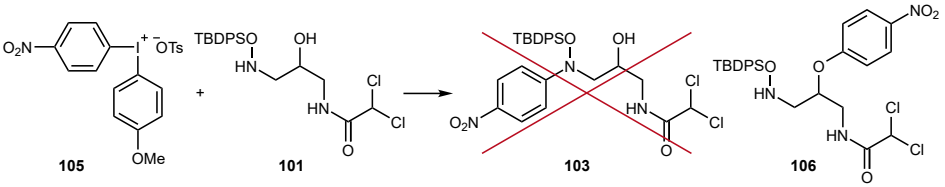


A solution of triflate **109** (112 mg, 0.25 mmol) and *tert*-butyl carbazate (330 mg, 2.50 mmol) in anhydrous THF (0.5 mL) was stirred at rt under Ar for 20 hr. The reaction mixture was evaporated on celite and subjected to silica-gel flash column chromatography (pet. ether:EtOAc, 8:2) and concentration of the relevant fractions yielded *N*-(3-((*tert*-Butyldimethylsilyl)oxy)-2-hydroxypropyl)-2,2-dichloroacetamide (**108**) (12 mg, 15%).

N-(3-(((*tert*-Butyldiphenylsilyl)oxy)(4-nitrophenyl)amino)-2-hydroxypropyl)-2,2-dichloroacetamide (103)



Method 1: TBDPS-protected hydroxylamine (**101**) (315 mg, 0.69 mmol), 1-fluoro-4-nitrobenzene (**30**) (87 mg, 0.62 mmol), and Et_3N (0.38 mL, 2.76 mmol) was dissolved in ACN (1.5 mL) and stirred at 40 °C under Ar for 18 hr, but no reaction was observed. The temperature was increased to 60 °C and after 2 hr there was still no formation of a new product. The same result was observed at 80 °C for 2 hr. ACN and Et_3N was removed under reduced pressure and replaced with DMF (1.0 mL) and DIPEA (0.48 mL, 2.76 mmol). The reaction mixture was stirred at 100 °C under Ar for 24 hr, and then evaporated directly on celite and subjected to silica-gel flash column chromatography (pet. ether:EtOAc, 9:1 → 8:2). Concentration of the relevant fractions yielded *N,N*-dimethyl-4-nitroaniline (**104**) (12 mg, 11%) as a bright yellow solid. Spectroscopic data are in accordance with previously reported literature.[109,110]



Method 2: TBDPS-protected hydroxylamine (**101**) (218 mg, 0.48 mmol) and Cs_2CO_3 (235 mg, 0.72 mmol) was stirred in toluene (2.0

mL) for 5 min before adding diaryliodonium salt **105** (306 mg, 0.58 mmol) in one portion. The solution immediately turned bright yellow. The reaction mixture was stirred at rt for 2.5 hr, and then evaporated directly on celite and subjected to silica-gel flash column chromatography (pet. ether:EtOAc, 9:1 → 7:3). Concentration of the relevant fractions yielded oxygen-arylated compound **106** (150 mg, 54%) as a yellow solid.

***N*,*N*-Dimethyl-4-nitroaniline (104)**:

TLC: R_f 0.20, pet. ether:EtOAc, 8:2.

^{1}H NMR (500.13 MHz, $CDCl_3$): δ 8.12 (d, J = 9.4 Hz, 2H), 6.60 (d, J = 9.4 Hz, 2H), 3.11 (s, 3H).

^{13}C NMR (125.76 MHz, $CDCl_3$): δ 154.3, 137.1, 126.2, 110.4, 40.4.

***N*-(3-(((*tert*-Butyldiphenylsilyl)oxy)amino)-2-(4-nitrophenoxy)propyl)-2,2-dichloroacetamide (106)**:

TLC: R_f 0.43, pet. ether:EtOAc, 7:3.

^{1}H NMR (500.13 MHz, CD_3CN): δ 8.16 (d, J = 9.3 Hz, 2H), 7.68 (dd, J = 8.1 Hz, 1.4 Hz, 2H), 7.63 (dd, J = 8.1 Hz, 1.4 Hz, 2H), 7.46-7.33 (m, 6H), 7.29 (t, J = 5.6 Hz, NH), 7.15 (d, J = 9.3 Hz, 2H), 6.05 (s, 1H), 6.00 (dd, J = 9.1 Hz, 5.3 Hz, NH), 5.03-4.96 (m, 1H), 3.54-3.49 (m, 1H), 3.44-3.39 (m, 1H), 3.12-3.07 (m, 1H), 2.99 (ddd, J = 13.9 Hz, 7.1 Hz, 5.5 Hz, 1H), 1.03 (s, 9H).

^{13}C NMR (125.76 MHz, CD_3CN): δ 165.5, 164.6, 142.6, 136.5, 136.4, 134.52, 134.47, 130.8, 130.7, 128.60, 128.59, 126.7, 116.8, 74.2, 67.5, 55.6, 42.2, 27.6, 19.6.

LRMS (ESI): Calcd for $C_{27}H_{30}Cl_2N_3O_5Si$ $[M\text{-}H]^-$ 574.13, found 574.19.

VII.2.2 Photochemical reactions

Photolysis of 1-phenyl-3-(4-nitrophenylamino)propan-2-ol (1)

A solution of compound **1** (≈28 mg, ≈0.10 mmol) was photolysed in a 150 mL (0.70 mM) photochemical reactor according to the general procedure with a 125 W medium-pressure mercury-vapour lamp for 2 hr at pH 1, 3, 5, 7, 9, 11, and 13. The aqueous phase was extracted with DCM, and for experiments performed at pH 7, 9, 11, and 13, the pH was adjusted to ≈ 2 after the initial extraction, and extracted again.

Photolysis of 1-phenyl-3-(3-nitrophenylamino)propan-2-ol (2)

A solution of compound **2** (≈28 mg, ≈0.10 mmol) was photolysed in a 150 mL (0.70 mM) photochemical reactor according to the general procedure with a 125 W medium-pressure mercury-vapour lamp for 2 hr at pH 1, 3, 5, 7, 9, 11, and 13. The aqueous phase was extracted with DCM, and for experiments performed at pH 7, 9, 11, and 13, the pH was adjusted to ≈ 2 after the initial extraction, and extracted again.

1-(4-Nitrophenyl)-3-(phenylamino)propan-2-ol (3)

A solution of compound **3** (≈28 mg, ≈0.10 mmol) was photolysed in a 150 mL (0.70 mM) photochemical reactor according to the general procedure with a 125 W medium-pressure mercury-vapour lamp for 2 hr at pH 1, 3, 5, 7, 9, 11, and 13. The aqueous phase was extracted with DCM, and for experiments performed at pH 7, 9, 11, and 13, the pH was adjusted to ≈ 2 after the initial extraction, and extracted again.

1-(3-Nitrophenyl)-3-(phenylamino)propan-2-ol (4)

A solution of compound **4** (≈28 mg, ≈0.10 mmol) was photolysed in a 150 mL (0.70 mM) photochemical reactor according to the general procedure with a 125 W medium-pressure mercury-vapour lamp for 2 hr at pH 1, 3, 5, 7, 9, 11, and 13. The aqueous phase was extracted with DCM, and for experiments performed at pH 7, 9, 11, and 13, the pH was adjusted to ≈ 2 after the initial extraction, and extracted again.

3-((4-Nitrophenyl)amino)-1-phenyldecan-2-ol (31)

A solution of compound **31** (23.6 mg, 0.064 mmol) was photolysed in a 150 mL (0.42 mM) photochemical reactor according to the general procedure with a 125 W medium-pressure mercury-vapour lamp for 4 hr at pH 11. The aqueous phase was extracted with DCM, the pH was adjusted to ≈ 2 after the initial extraction, and extracted again.

1-((2,5-Dichloro-4-(1,1,2,3,3,3-hexafluoropropoxy)phenyl)-amino)-3-(4-nitrophenyl)propan-2-ol (63)

A solution of compound **63** (26.6 mg, 0.052 mmol) was photolysed in a 75 mL (0.70 mM) photochemical reactor according to the general procedure with a 6 W medium-pressure mercury-vapour lamp for 24 hr at pH 13 and 8. The aqueous layer was extracted with EtOAc, the pH was adjusted to $\approx$ 2 after the initial extraction, and extracted again.

1-((3,5-Dichloro-2-fluorophenyl)amino)-3-(2,6-difluoro-3-nitrophenyl)propan-2-ol (78)

A solution of compound **78** (20.4 mg, 0.052 mmol) was photolysed in a 75 mL (0.69 mM) photochemical reactor according to the general procedure with a 6 W medium-pressure mercury-vapour lamp for 24 hr at pH 13 and 8. The aqueous layer was extracted with EtOAc, the pH was adjusted to $\approx$ 2 after the initial extraction, and extracted again.

1-((3,5-Dichloro-2,4-difluorophenyl)amino)-3-(2,6-difluoro-3-nitrophenyl)propan-2-ol (79)

A solution of compound **79** (21.0 mg, 0.051 mmol) was photolysed in a 75 mL (0.68 mM) photochemical reactor according to the general procedure with a 6 W medium-pressure mercury-vapour lamp for 24 hr at pH 13 and 8. The aqueous layer was extracted with EtOAc, the

pH was adjusted to ≈ 2 after the initial extraction, and extracted again.

1-((2,5-Dichloro-4-(1,1,2,3,3,3-hexafluoropropoxy)phenyl)-amino)-3-(2,6-difluoro-3-nitrophenyl)propan-2-ol (80)

A solution of compound **80** (26.8 mg, 0.049 mmol) was photolysed in a 75 mL (0.66 mM) photochemical reactor according to the general procedure with a 6 W medium-pressure mercury-vapour lamp for 24 hr at pH 13 and 8. The aqueous layer was extracted with EtOAc, the pH was adjusted to ≈ 2 after the initial extraction, and extracted again.

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Paper I

Photodegradable antimicrobial agents - synthesis, photodegradation, and biological evaluation

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Photodegradable antimicrobial agents – synthesis, photodegradation, and biological evaluation†

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Multi-drug resistant (MDR) bacteria are already a significant health-care problem and are making the combat of infections quite challenging. Here we report the synthesis of several new compounds containing an ethanolamine moiety, of which two exhibit promising antimicrobial activity (at the 6 μM level). All the compounds are degraded when exposed to light and form inactive products.

Introduction

Alexander Fleming's discovery of penicillin in 1928 [1] started a revolution in the treatment of infections that over the years has saved millions of lives.[2,3] However, the extensive use of penicillin and other antibiotics is about to make them less effective due to the development of multi-drug resistant (MDR) bacteria.[4–6] Such bacteria are already a widespread problem, and several public-health organisations describe the situation as critical and predict catastrophic consequences with an annual death toll in the millions by 2050.[2,7–10] Thus, there is a need for new antibiotics that both combat existing MDR bacteria and prevent formation of new ones.[11]

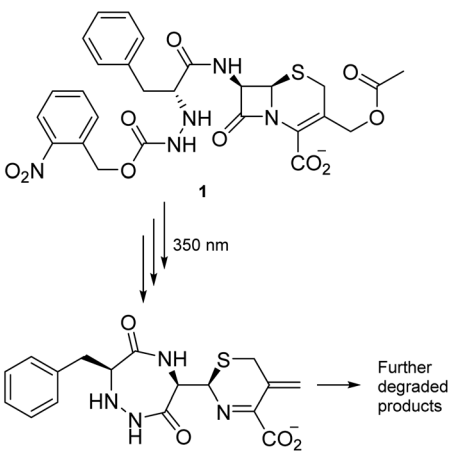
There are several causes for the MDR-bacteria problems,[2,7,8] but a most important factor is the excessive use, both in human health care and agriculture,[4] of antibiotics, which enter the environment more or less metabolized through sewage and run-off from farmland.[12–15] A proof of the alarming state of affairs is the increasing levels of antibiotic residues in drinking water, wastewater, ground water, coastal waters, and marine organisms.[12,16–20] In order to stop this development, it is necessary to develop antibiotics based on scaffolds with a structural motif that will enable their degradation under ambient conditions so that accumulation in the biosphere does not take place.

Most antibiotics act inside the body, so the stability of conceivable new agents must necessarily be significant under aqueous and physiological conditions. Our search for degradable moieties was therefore turned to motifs that are known to react with light, another reagent present in abundance in the environment. Light has already been applied repeatedly to turn compounds into derivatives for medical application,[21–23] and irradiation of a carbamate group has successfully been utilized by Lee *et al.* to facilitate the decomposition of the β-lactam moiety embedded in cephalosporanic-acid antibiotic (**1**) (Scheme 1) in a cascade reaction resulting in fragments with no antibiotic activity.[24]

The decomposition illustrated in Scheme 1 appeared to be made possible by the nitrobenzyl group, which has a well-documented reactivity in the excited state.[25] This reactivity has been utilized by Wan and Muralidharan to carry out photo-*retro*-aldol reactions with a variety of 1-(4-nitrobenzyl)-1-alkanols, the result of which was fragmentation and formation of 4-nitrotoluene, 4,4′-dinitrobibenzyl, and the corresponding alkanal in yields that were pH dependent (Scheme 2).[26] We therefore speculated if such alkanols have the potential to function as scaffolds for new photodegradable antibiotics provided substituents that induce biological activity are attached in appropriate positions (Fig. S1†).

Synthesis

The substituents we had in mind were halogenated anilines that were present in some biologically active compounds



Scheme 1 Light-induced degradation of a cephalosporanic acid analogue.

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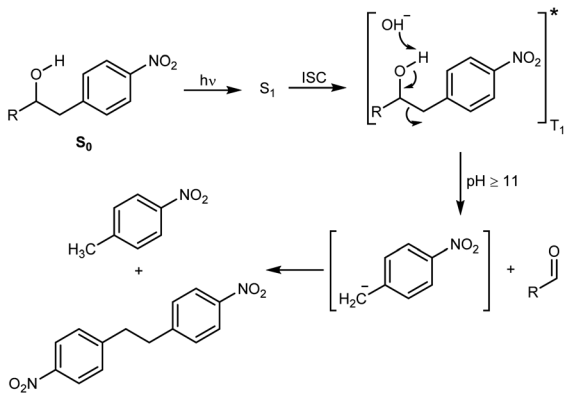


Scheme 2 The photo-*retro*-aldol type reaction of nitrophenethyl alcohols. R = Ph, CH_3. Irradiation with a 254, 300, or 350 nm lamp yields the first excited singlet state (S_1), which undergoes intersystem crossing (ISC) and formation of triplet-excited (T_1) alkanols that suffer fragmentation.

prepared and tested for other purposes (Fig. S2†). In order to make the alkanols as biocompatible as possible, we opted for 2-(arylamino)-1-(nitrobenzyl)ethanol derivatives (**2–5**), which were prepared in quite satisfactory yields by application of standard coupling and oxidation reactions (Scheme 3).

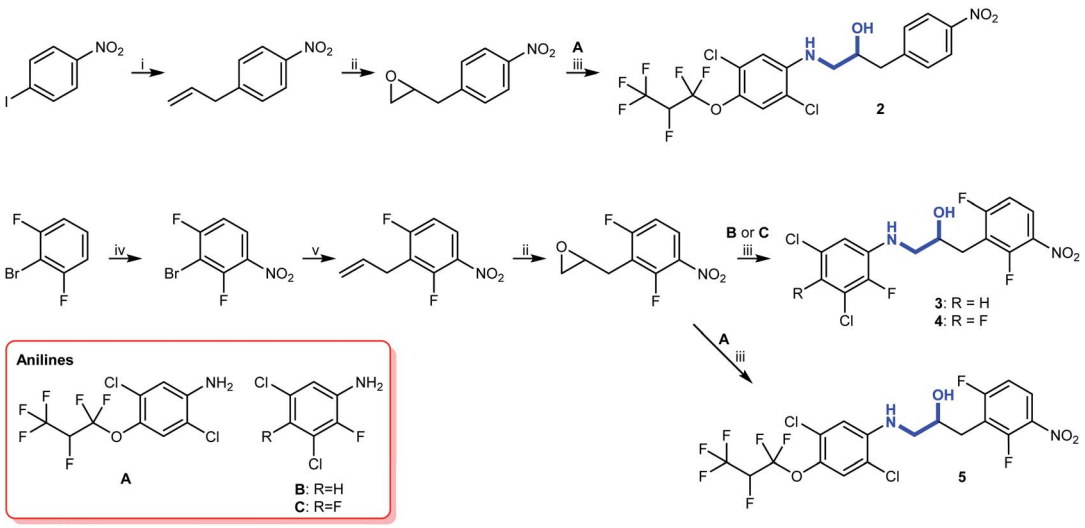
To this end, a Suzuki–Miyaura cross-coupling reaction with allylboronic acid pinacol ester, using a method described by Kotha and co-workers,[27] yielded the allylated product (Scheme 3). Subsequent treatment with *m*CPBA gave the corresponding epoxide and a Lewis acid-promoted epoxide ring-opening reaction using 5 M lithium perchlorate-diethyl ether (LPDE) solution gave aminol **2**. The remaining three compounds were prepared by a selective meta nitration followed by a Stille cross-coupling reaction with allyltributylstannane yielding allylbenzene. Subsequent epoxidation with *m*CPBA gave the required epoxide and a Lewis acid-promoted epoxide ring opening with anilines **A**, **B**, and **C** gave aminols **3–5**.

Photodecomposition

The photodegradation of compounds **2–5** was studied at pH 8 and 13 following the procedure reported by Wan and Muralidharan[26] using a low-pressure mercury-vapor lamp emitting light mainly at 254 nm (Fig. S3†). The reaction was monitored by proton NMR spectroscopy, which proved that almost complete conversion was achieved after 24 hours, for compounds **2–4** at pH 13 and compounds **3** and **5** at pH 8 (Table S1†). Control experiments show that compounds **2–5** are stable for more than 24 hours in the dark at pH 8 and compound **2** is even stable at pH up to 13 for 24 hours. The changes that took place are illustrated by three spectra associated with photolysis of aminol **2**. Sharp, intense peaks in the spectrum of **2** (A in Fig. 1) are essentially absent in the spectrum of the reaction mixture after irradiation for 24 hours (B in Fig. 1), which on the other hand, contains strong peaks due to aniline **A** (Scheme 3; C in Fig. 1) formed during the degradation of **2** (see also Fig. S4†). MS analysis of the decomposition mixture also disclosed a mass equivalent to *para*-nitrobenzoic acid, which must have derived from the corresponding aldehyde formed under the photodecomposition.

Antibacterial activity

With compounds **2–5** and the corresponding crude degradation mixtures (**2d–5d**) at hand, the minimum inhibitory concentration (MIC) was determined by screening the compounds against several laboratory strains of Gram-positive and Gram-negative bacteria, including *E. faecalis*, *E. coli*, *P. aeruginosa, S. aureus*, *S. agalactiae*, and *S. epidermidis* (Tables 1 and S2†). As the results in Table 1 show, compounds **2** and **5** inhibited the growth of *S. agalactiae* at 6.3 μM while compounds **3** and **4**



Scheme 3 Synthesis of aminols **2–5**. Reagents and conditions: (i) $Pd(PPh_3)_4$, CsF, AllylBpin, THF, reflux;[27] (ii) *m*CPBA, DCM, rt; (iii) 5 M LPDE, 40 °C; (iv) H_2SO_4, HNO_3, 0 °C; (v) $Pd(PPh_3)_4$, $Bu_3SnAllyl$, DMF, 110 °C.

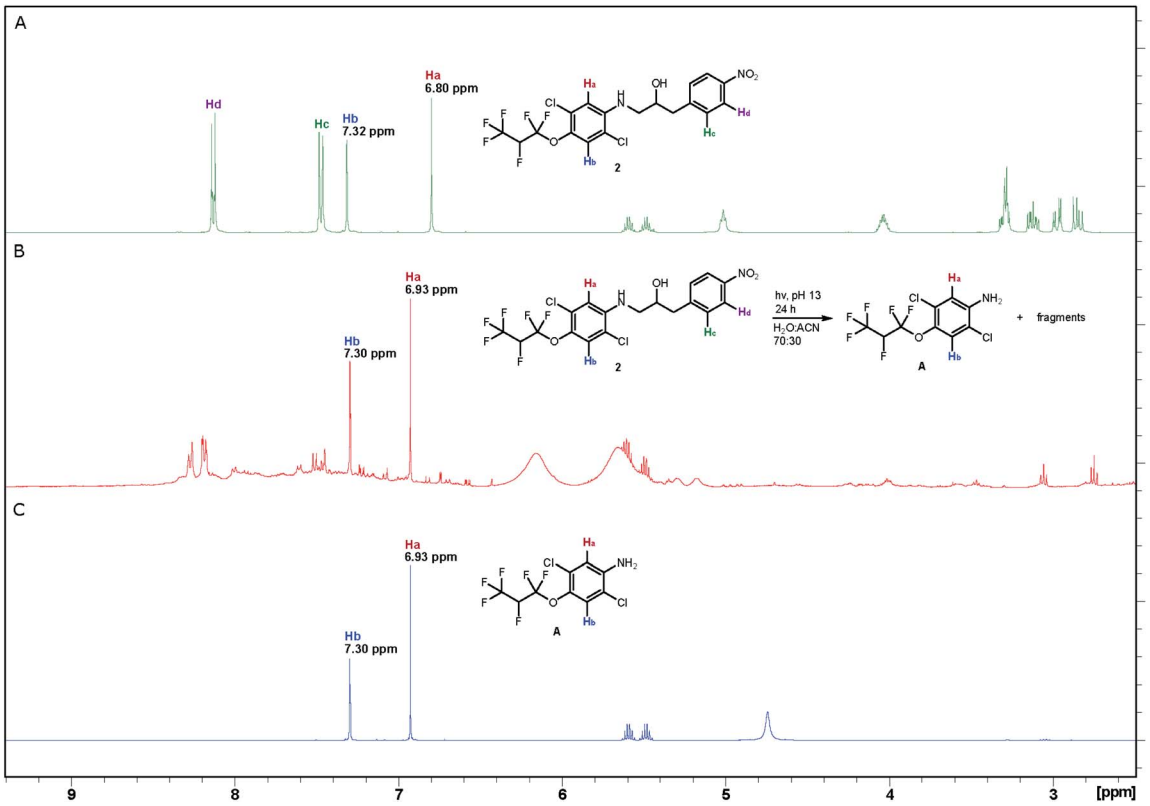


Fig. 1 ^{1}H NMR spectra (A) compound **2** (0.7 mM) before irradiation; (B) reaction mixture after photolysis of compound **2** for 24 hours; (C) decomposition product (A). The NMR analyses were recorded in CD_3CN at 400 MHz.

inhibited bacteria growth at 50 μM. The corresponding degradation product mixtures exhibited no antimicrobial activity whatsoever and showed no toxicity towards MRC5 and HepG2 cell lines either (Table 1). However, the two most active antimicrobial agents, *viz.* compounds **2** and **5**, were somewhat toxic (Table 1), but at a much higher concentration then that required to reveal antimicrobial activity.

Table 1 Minimum inhibitory concentration (MIC) in μM for compounds **2–5** against Gram-positive bacteria and toxicity tests against MRC5 and HepG2 cell lines. The activity was screened at the following concentrations: 100, 75, 50, 25, 12.5, 6.3, 3.1, and 1.6 μM[a]

	MIC			Tox	
Compound	*S. epidermidis*	*S. aureus*	*S. agalactiae*	MRC5	HepG2
2	I	I	6.3	50	75
3	50	50	50	50	75
4	I	50	I	50	50
5	I	I	6.3	25	50
2d	I	I	I	I	I
3d	I	I	I	I	I
4d	I	I	I	I	I
5d	I	I	I	I	I

[a] I = inactive at the tested concentrations.

Experimental

General information

IR spectra were recorded on an Agilent Cary 630 FT-IR spectrophotometer equipped with an attenuated total reflectance (ATR) attachment. Samples were analysed neat on a ZnSe crystal and the absorption frequencies are given in wave numbers (cm^{-1}).

UV-vis spectra were obtained on an Agilent 8453 single-beam UV-vis spectrophotometer with a deuterium-discharge lamp for the UV range and a tungsten lamp for the visible wavelength range. Samples were analysed in an Agilent open-top UV quartz cell (10 mm, 3.0 mL) with ethanol as solvent. The wavelengths are reported in nm and molar attenuation coefficients in M^{-1} cm^{-1}.

NMR spectra were recorded on a Bruker Ascend™ 400 spectrometer (400.13 MHz for ^{1}H, 100.61 MHz for ^{13}C, 376.46 MHz for ^{19}F) or a Bruker Ascend™ 850 spectrometer (850.13 MHz for ^{1}H and 213.77 MHz for ^{13}C). Coupling constants (J) are given in Hz and the multiplicity is reported as singlet (s), doublet (d), triplet (t), sextet (sxt), multiplet (m), and broad singlet (bs). The chemical shift are reported in ppm in the order downfield to upfield and calibration is done using the residual solvent signals for chloroform-d (^{1}H 7.26 ppm; ^{13}C 77.16 ppm) or acetonitrile-d_3 (^{1}H 1.94 ppm; ^{13}C 1.32 ppm).[28] Calibration for ^{19}F NMR is done using α,α,α-trifluorotoluene as internal

standard in chloroform-*d* (−62.61 ppm) and acetonitrile-d_3 (−63.10 ppm).[29]

High-resolution mass spectra were obtained on a JEOL AccuTOF™ T100GC mass spectrometer. The instrument was operated with an orthogonal electrospray ionization source (ESI), an orthogonal accelerated time of flight (TOF) single stage reflectron mass analyzer and a dual micro channel plate (MCP) detector at the following instrumental settings/conditions; ionization mode: positive, desolvating temperature/ion source temperature = 250 °C, needle voltage = 3000 V, desolvation gas flow = 2.0 L min^{-1}, nebulising gas flow = 1.0 L min^{-1}, orifice1 temperature = 120 °C, orifice1 voltage = 24 V, ring lens voltage = 12 V, orifice2 voltage = 6 V, ion guide peak voltage = 800 V, detector voltage = 2300 V, acquisition range = 4–1000 *m*/*z*, spectral recording interval = 0.5 s, wait time = 0.03 ns and data sampling interval = 0.5 ns. Mass calibration were performed using the internal standard method and mass drift compensation was performed in each acquisition.

Thin-layer chromatography (TLC) was carried out with silica gel (60 F_{254}) on aluminium sheets with solvent systems consisting of various mixtures of petroleum ether, ethyl acetate, and dichloromethane. Visualization was achieved with either UV light (254 and/or 365 nm) or a potassium permanganate stain. Flash chromatography was performed with a hand pump and 230–400 mesh silica gel or an Interchim Puriflash 215 autoflash chromatography system with Biotage Snap Ultra HP-Sphere™ 25 μm silica-gel columns.

General procedure

Lewis-acid promoted epoxide ring opening. Lithium perchlorate was dried under vacuum for 1 h and dissolved in anhydrous diethyl ether to a 5 M solution. The appropriate aniline (1.0–1.3 equiv.), (∼0.2 M) and epoxide (1.0 equiv.), (∼0.2 M) was added and the reaction mixture was stirred at 40 °C under Ar overnight. DCM and water were added, the phases were separated, and the aqueous layer was extracted with DCM (3 × 10 mL). The combined organic phases were evaporated on Celite and subjected to silica-gel flash chromatography (pet. ether : DCM, 3 : 7), and concentration of the relevant fractions yielded the nitrophenethyl alcohols.

Synthesis of 1-allyl-4-nitrobenzene. An oven-dried 25 mL round-bottom flask fitted with a condenser was charged with 1-iodo-4-nitrobenzene (1.00 g, 4.00 mmol), CsF (1.52 g, 10.0 mmol), $Pd(PPh_3)_4$ (0.70 g, 15 mol%), THF (20 mL), and water (2 mL). The mixture was stirred at rt under Ar for 30 min, followed by addition of allylboronic acid pinacol ester (1.36 mL, 7.20 mmol) in THF (8 mL). The reaction mixture was refluxed (oil bath, 95 °C) for 22 h. After cooling to rt the product mixture was evaporated onto Celite and purified by silica-gel flash chromatography (pet. ether). Concentration of the relevant fractions (R_f = 0.47 (pet. ether : EtOAc, 80 : 20 v/v)) yielded 1-allyl-4-nitrobenzene (0.35 g, 52%) as a slightly yellow liquid. The spectroscopic data were in accordance with previously reported data.[30]

IR (neat): ν_{max} 3091, 3017, 2918, 1593, 1502; ^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.15 (d, J = 8.8 Hz, 2H), 7.34 (d, J = 8.8 Hz, 2H), 5.99–5.89 (m, 1H), 5.18–5.09 (m, 2H), 3.49 (d, J = 6.7 Hz, 2H); ^{13}C NMR (100.61 MHz, $CDCl_3$): δ 147.9, 146.7, 135.6, 129.5, 123.8, 117.5, 40.0.

Synthesis of 2-(4-nitrobenzyl)oxirane. An oven-dried 25 mL round-bottom flask charged with 1-allyl-4-nitrobenzene (0.35 g, 2.16 mmol) in anhydrous DCM (12 mL) was cooled (ice/water bath) and stirred for 5 min under Ar followed by addition of *m*CPBA (0.59 g, 2.63 mmol). The reaction mixture was stirred at 0 °C for 2 h, then at rt for 15 h. More *m*CPBA (0.12 g, 0.54 mmol) was added and stirring continued for another 31 h before quenching the reaction with 1 M aq. NaOH solution (10 mL). The phases were separated and the aq. phase was extracted with DCM (3 × 15 mL). The combined organic phases were washed with water (20 mL), brine (20 mL), dried (MgSO4), filtered, and concentrated *in vacuo*. The product was isolated by silica-gel autoflash chromatography (pet. ether : EtOAc : DCM, 93 : 2 : 5 → 40 : 55 : 5) and concentration of the relevant fractions (R_f = 0.26 (pet. ether : DCM, 50 : 50 v/v)) yielded 2-(4-nitrobenzyl) oxirane as a yellow oily liquid (0.20 g, 52%). The spectroscopical data were in full accord with the previously reported data.[31]

IR (neat): ν_{max} 2957, 2923, 2853, 1599, 1516, 1345; ^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.18 (d, J = 8.5 Hz, 2H), 7.43 (d, J = 8.5 Hz, 2H), 3.20–3.16 (m, 1H), 3.06 (dd, J = 14.8 Hz, 4.2 Hz, 1H), 2.90 (dd, J = 14.8 Hz, 6.4 Hz, 1H), 2.84 (dd, J = 4.7 Hz, 4.0 Hz, 1H), 2.55 (dd, J = 4.7 Hz, 2.6 Hz, 1H); ^{13}C NMR (100.61 MHz, $CDCl_3$): δ 147.1, 145.0, 130.0, 123.9, 51.7, 46.8, 38.6.

Synthesis of 1-((2,5-dichloro-4-(1,1,2,3,3,3-hexafluoropropoxy)phenyl)amino)-3-(4-nitrophenyl)propan-2-ol (2). 2,5-Dichloro-4-(1,1,2,3,3,3-hexafluoropropoxy)aniline (111 mg, 0.34 mmol) and 2-(4-nitrobenzyl)oxirane (62 mg, 0.34 mmol) was reacted according to the general procedure for 19 h to yield aminol **2** (R_f = 0.36 (pet. ether : DCM, 3 : 7)) as a white solid (87 mg, 50%, mp. 121–123 °C) along with 40% recovery of epoxide.

IR (neat): ν_{max} 3507, 3420, 3376, 2980, 2918, 2857, 1599; UV/vis (EtOH): λ_{max} 255 nm (ε 18 339 M^{-1} cm^{-1}); ^{1}H NMR (850.13 MHz, CD_3CN): δ 8.14 (d, J = 8.7 Hz, 2H), 7.49 (d, J = 8.7 Hz, 2H), 7.33 (s, 1H), 6.81 (s, 1H), 5.55 (dsxt, J = 42.8 Hz, 5.8 Hz, 1H), 5.03 (t, J = 5.6 Hz, NH), 4.06–4.02 (m, 1H), 3.30 (ddd, J = 13.3 Hz, 6.4 Hz, 3.9 Hz, 1H), 3.27 (d, J = 4.4 Hz, OH), 3.15–3.11 (m, 1H), 2.98 (dd, J = 13.8 Hz, 4.5 Hz, 1H), 2.85 (dd, J = 13.8 Hz, 8.4 Hz, 1H); ^{13}C NMR (213.77 MHz, CD_3CN): δ 148.2, 147.7, 145.2, 134.5, 131.5, 127.9, 125.6, 124.3, 121.3 (qd, J = 281 Hz, 25 Hz), 119.1 (td, J = 271 Hz, 23 Hz), 117.7, 112.6, 85.4 (dsxt, J = 198 Hz, 35 Hz), 70.8, 49.7, 41.7; ^{19}F NMR (376.46 MHz, CD_3CN): δ −75.6–75.7 (m, 3F), −78.4–80.4 (m, 2F), −213.3 (sxt, J = 12 Hz, 1F); HRMS: (ESI/TOF) *m*/*z*: $[M + Na]^+$ calcd for $C_{18}H_{14}Cl_2F_6N_2O_4Na^+$ 529.01270; found 529.01286.

Synthesis of 2-bromo-1,3-difluoro-4-nitrobenzene. 2-Bromo-1,3-difluorobenzene (579 mg, 3.00 mmol) was dissolved in 95–97% sulfuric acid (4 mL) and cooled to 0 °C. To this was added an ice-cold mixture of 95–97% sulfuric acid (4 mL) and 65% nitric acid (5.2 mL) dropwise over 15 min. The reaction mixture was stirred at 0 °C for 1 h, poured over ice, and extracted with DCM (3 × 15 mL). The combined organic layers were washed with sat. aq. $NaHCO_3$ solution (20 mL), dried ($MgSO_4$), filtered, and concentrated under reduced pressure to yield 2-bromo-1,3-

difluoro-4-nitrobenzene as a white crystalline solid (691 mg, 97%, mp 52–53 °C).

IR (neat): ν_{max} 3099, 1916, 1591, 1530; ^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.12 (ddd, J = 9.4 Hz, 8.0 Hz, 5.5 Hz, 1H), 7.14 (ddd, J = 9.4 Hz, 7.0 Hz, 2.0 Hz, 1H); ^{13}C NMR (100.61 MHz, $CDCl_3$): δ 163.1 (dd, J = 260 Hz, 3 Hz), 154.3 (dd, J = 267 Hz, 5 Hz), 134.8, 126.2 (dd, J = 10 Hz, 2 Hz), 112.1 (dd, J = 24 Hz, 4 Hz), 101.3 (dd, J = 25 Hz, 24 Hz); ^{19}F NMR (376.46 MHz, $CDCl_3$): δ −92.0 (d, J = 9.5 Hz), −104.4 (d, J = 9.5 Hz); HRMS: (EI/TOF) m/z: $[M]^+$ calcd for $C_6H_2BrF_2NO_2^+$ 236.92315; found 236.92306.

Synthesis of 2-allyl-1,3-difluoro-4-nitrobenzene. 2-Bromo-1,3-difluoro-4-nitrobenzene (870 mg, 3.66 mmol), $Pd(PPh_3)_4$ (634 mg, 15 mol%), and $Bu_3SnAllyl$ (1.53 mL, 4.39 mmol) was dissolved in anhydrous DMF (8 mL). 4 Å molecular sieves were added and the reaction mixture was stirred at 100 °C under Ar for 24 h. The solids were filtered off through cotton with the aid of DCM (50 mL), and the volatiles were removed under reduced pressure. The title compound was isolated by silica-gel flash chromatography (pet. ether : EtOAc, 99 : 1) and concentration of the relevant fractions (R_f = 0.32 (pet. ether : EtOAc, 95 : 5, v/v)) yielded 2-allyl-1,3-difluoro-4-nitrobenzene as a colourless liquid (194 mg, 27%).

IR (neat): ν_{max} 3087, 2957, 2925, 2855, 1623, 1596; ^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.01 (ddd, J = 9.0 Hz, 8.5 Hz, 5.7 Hz, 1H), 7.04–6.99 (m, 1H), 5.95–5.85 (m, 1H), 5.13–5.08 (m, 2H), 3.51–3.48 (m, 2H); ^{13}C NMR (100.61 MHz, $CDCl_3$): δ 164.2 (dd, J = 258 Hz, 8 Hz), 155.2 (dd, J = 266 Hz, 10 Hz), 134.5–134.4 (m), 132.9, 125.4 (dd, J = 11 Hz, 2 Hz), 118.8 (dd, J = 22 Hz, 19 Hz), 117.4, 111.7 (dd, J = 25 Hz, 4 Hz), 26.8 (t, J = 3 Hz); ^{19}F NMR (376.46 MHz, $CDCl_3$): δ −102.5 (d, J = 14.1 Hz), −116.6 (d, J = 14.1 Hz); HRMS: (EI/TOF) m/z: $[M]^+$ calcd for $C_9H_7F_2NO_2^+$ 199.04394; found 199.04399.

Synthesis of 2-(2,6-difluoro-3-nitrobenzyl)oxirane. To a stirred solution of 2-allyl-1,3-difluoro-4-nitrobenzene (194 mg, 0.97 mmol) at 0 °C was added *m*CPBA (425 mg, 1.94 mmol). The reaction mixture was stirred for 2 h at 0 °C and 5 d at rt, followed by addition of a 1 : 1 sat. aq. $NaHCO_3$: 10% $Na_2S_2O_3$ solution (30 mL). The phases were separated and the aqueous layer was extracted with DCM (3 × 15 mL). The combined organic phases were washed with a 1 : 1 sat. aq. $NaHCO_3$:10% $Na_2S_2O_3$ solution (30 mL), sat. aq. $NaHCO_3$ (30 mL), water (30 mL), dried ($MgSO_4$), filtered and concentrated under reduced pressure to yield 2-(2,6-difluoro-3-nitrobenzyl)oxirane as a slightly yellow oily liquid (183 mg, 88%). The product was used without further purification.

IR (neat): ν_{max} 3104, 3000, 2926, 1728, 1624; ^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.05 (ddd, J = 9.2 Hz, 8.5 Hz, 5.7 Hz, 1H), 7.07–7.02 (m, 1H), 3.22–3.17 (m, 1H), 3.16–3.11 (m, 1H), 3.03–2.97 (m, 1H), 2.81–2.79 (m, 1H), 2.56 (dd, J = 4.8 Hz, 2.5 Hz, 1H); ^{13}C NMR (100.61 MHz, $CDCl_3$): δ 164.5 (dd, J = 259 Hz, 8 Hz), 155.5 (dd, J = 266 Hz, 9 Hz), 134.5, 126.1 (dd, J = 11 Hz, 1 Hz), 115.8 (dd, J = 22 Hz, 20 Hz), 111.8 (dd, J = 25 Hz, 4 Hz), 50.1, 46.9, 25.8 (t, J = 2 Hz); ^{19}F NMR (376.46 MHz, $CDCl_3$): δ −101.5 (d, J = 13.6 Hz), −115.7 (d, J = 13.6 Hz); HRMS; (EI/TOF) m/z: $[M–C_2H_2O]^+$ calcd for $C_7H_4F_2NO_2^+$ 172.02046; found 172.02080.

Synthesis of 1-((3,5-dichloro-2-fluorophenyl)amino)-3-(2,6-difluoro-3-nitrophenyl)propan-2-ol (3). 3,5-Dichloro-2-fluoroaniline (82 mg, 0.46 mmol) and 2-(2,6-difluoro-3-nitrobenzyl)oxirane (77 mg, 0.36 mmol) was reacted according to the general procedure for 20 h to yield aminol **3** (pet. ether:DCM, 3 : 7 (R_f = 0.36)) as a sticky colourless oil (60 mg, 42%) along with 7% recovery of epoxide.

IR (neat): ν_{max} 3565, 3425, 3101, 2926, 2857, 1597; UV/vis (EtOH): λ_{max} 254 nm (ε 16 575 M^{-1} cm^{-1}); ^{1}H NMR (400.13 MHz, CD_3CN): δ 8.05 (ddd, J = 9.3 Hz, 8.5 Hz, 5.7 Hz, 1H), 7.14 (ddd, J = 9.3 Hz, 8.5 Hz, 1.9 Hz, 1H), 6.73–6.69 (m, 2H), 4.93 (bs, NH), 4.06–3.98 (m, 1H), 3.34 (d, J = 5.3 Hz, OH), 3.32 (ddd, J = 13.5 Hz, 6.7 Hz, 4.0 Hz, 1H), 3.20–3.13 (m, 1H), 2.99–2.87 (m, 2H); ^{13}C NMR (100.61 MHz, CD_3CN): δ 165.5 (dd, J = 256 Hz, 8 Hz), 156.2 (dd, J = 263 Hz, 10 Hz), 146.8 (d, J = 239 Hz), 139.9 (d, J = 12 Hz), 135.5, 130.5 (d, J = 4 Hz), 126.7 (dd, J = 12 Hz, 2 Hz), 121.4 (d, J = 16 Hz), 118.6 (dd, J = 22 Hz, 19 Hz), 116.5 (d, J = 1 Hz), 112.6 (dd, J = 25 Hz, 4 Hz), 111.6 (d, J = 4 Hz), 69.3, 49.5, 29.1; ^{19}F NMR (376.46 MHz, CD_3CN): δ −103.0 (d, J = 13.6 Hz, 1F), −117.5 (d, J = 13.6 Hz, 1F), −141.7 (s, 1F); HRMS: (ESI/TOF) m/z: $[M + H]^+$ calcd for $C_{15}H_{12}Cl_2F_3N_2O_3^+$ 395.01716; found 395.01724.

Synthesis of 1-((3,5-dichloro-2,4-difluorophenyl)amino)-3-(2,6-difluoro-3-nitrophenyl)propan-2-ol (4). 3,5-Dichloro-2,4-difluoroaniline (65 mg, 0.33 mmol) and 2-(2,6-difluoro-3-nitrobenzyl)-oxirane (71 mg, 0.33 mmol) was reacted according to the general procedure for 22 h to yield aminol **4** (R_f = 0.30 (pet. ether:DCM, 3 : 7)) as a white solid (45 mg, 33%, mp. 110–112 °C) along with 46% recovery of epoxide.

IR (neat): ν_{max} 3433, 3382, 3103, 2935, 2857, 1625; UV/vis (EtOH): λ_{max} 242 nm (ε 16 289 M^{-1} cm^{-1}); ^{1}H NMR (400.13 MHz, CD_3CN): δ 8.04 (ddd, J = 9.2 Hz, 8.6 Hz, 5.7 Hz, 1H), 7.13 (ddd, J = 9.2 Hz, 8.5 Hz, 1.8 Hz, 1H), 6.79 (dd, J = 8.6 Hz, 7.1 Hz, 1H), 4.72 (bs, NH), 4.05–3.97 (m, 1H), 3.34 (d, J = 5.3 Hz, OH), 3.92 (ddd, J = 13.4 Hz, 6.6 Hz, 4.1 Hz, 1H), 3.16–3.10 (m, 1H), 2.98–2.87 (m, 2H); ^{13}C NMR (100.61 MHz, CD_3CN): δ 165.5 (dd, J = 256 Hz, 8 Hz), 156.2 (dd, J = 263 Hz, 10 Hz), 146.9 (dd, J = 242 Hz, 1 Hz), 146.3 (dd, J = 237 Hz, 2 Hz), 135.7 (dd, J = 12 Hz, 3 Hz), 135.4 (dd, J = 8 Hz, 3 Hz), 126.7 (d, J = 12 Hz), 118.5 (dd, J = 22 Hz, 20 Hz), 117.3 (dd, J = 18 Hz, 4 Hz), 112.6 (dd, J = 25 Hz, 4 Hz), 111.4–111.0 (m, 1C), 111.2 (d, J = 4 Hz), 69.4, 49.7, 29.1; ^{19}F NMR (376.46 MHz, CD_3CN): δ −103.0 (d, J = 14.0 Hz, 1F), −117.5 (d, J = 14.0 Hz, 1F), −133.9 (d, J = 4.0 Hz, 1F), −137.0 (d, J = 4.0 Hz, 1F); HRMS: (ESI/TOF) m/z: $[M + H]^+$ calcd for $C_{15}H_{11}Cl_2F_4N_2O_3^+$ 413.00774; found 413.00790.

Synthesis of 1-((2,5-dichloro-4-(1,1,2,3,3,3-hexafluoropropoxy)phenyl)amino)-3-(2,6-difluoro-3-nitrophenyl)propan-2-ol (5). 2,5-Dichloro-4-(1,1,2,3,3,3-hexafluoropropoxy)aniline (151 mg, 0.46 mmol) and 2-(2,6-difluoro-3-nitrobenzyl)-oxirane (100 mg, 0.46 mmol) was reacted according to the general procedure for 18 h to yield aminol **5** (R_f = 0.36 (pet. ether:DCM, 3 : 7)) as a sticky colourless oil (104 mg, 42%) along with 16% recovery of epoxide.

IR (neat): ν_{max} 3550, 3411, 3103, 2937, 1624; UV/vis (EtOH): λ_{max} 254 nm (ε 25 248 M^{-1} cm^{-1}); ^{1}H NMR (850.13 MHz, CD_3CN): δ 8.06–8.04 (m, 1H), 7.33 (s, 1H), 7.15–7.13 (m, 1H), 6.86 (s, 1H), 5.55 (dsxt, J = 42.7 Hz, 5.8 Hz, 1H), 5.04 (t, J = 5.7 Hz, NH), 4.07–4.04 (m, 1H), 3.39 (d, J = 5.2 Hz, OH), 3.36 (ddd, J = 13.3 Hz, 6.4 Hz, 4.1 Hz, 1H), 3.22–3.19 (m, 1H), 2.97

(dd, J = 13.9 Hz, 4.8 Hz, 1H), 2.93 (dd, J = 13.9 Hz, 8.3 Hz, 1H); ^{13}C NMR (213.77 MHz, CD_3CN): δ 165.5 (dd, J = 256 Hz, 8 Hz), 156.2 (dd, J = 263 Hz, 10 Hz), 145.2, 135.5 (d, J = 5 Hz), 134.5, 127.9, 126.7 (d, J = 12 Hz), 125.6, 121.3 (qd, J = 281 Hz, 25 Hz), 119.1 (td, J = 271 Hz, 23 Hz), 118.5 (dd, J = 22 Hz, 19 Hz), 117.7, 112.73–112.60 (m, 1C), 112.71, 85.4 (dsxt, J = 198 Hz, 35 Hz), 69.3, 49.6, 29.2; ^{19}F NMR (376.46 MHz, CD_3CN): δ −75.6–75.7 (m, 3F), −78.5–80.4 (m, 2F), −102.9 (d, J = 14.0 Hz, 1F), −117.4 (d, J = 14.0 Hz, 1F), −213.3 (sxt, J = 11.2 Hz, 1F); HRMS: (ESI/TOF) m/z: $[M + H]^+$ calcd for $C_{18}H_{13}Cl_2F_8N_2O_4^+$ 543.01191; found 543.01196.

Photodegradation reactions at ∼0.7 mM for NMR analysis

All reactions were carried out in a 75 mL gas inlet flask (∼0.25–0.35 mM) under a constant stream of nitrogen with a quartz well water-cooled cold finger for 24 hours. The appropriate aminol (∼0.02 mmol) was dissolved in acetonitrile (23 mL), diluted with distilled water adjusted to pH 8 with sodium hydroxide (52 mL), and photolyzed using a 254 nm low-pressure mercury lamp. The aqueous solution was extracted with DCM (3 × 20 mL) and the combined organic layers were dried ($MgSO_4$), filtered, and concentrated to yield a black residue, which was analysed by ^{1}H NMR spectroscopy.

Conclusions

In conclusion, two compounds based on a new structural scaffold, **2** and **5**, possess both antimicrobial activity at the 6 μM level and the ability to decompose upon exposure to light. The decomposition products do not exhibit antimicrobial activity or toxicity. This opens for the development of antibiotics that when released to the environment after treatment will be removed and not contribute to the evolution of MDR bacteria. Both **2** and **5** were not active towards Gram-negative bacteria, but work by Richter *et al.* outlines techniques that can be applied to expand their capability to encompass Gram-negative bacteria as well.[32] Work directed towards this is ongoing in these laboratories.

Author contributions

LKS and MOS had the initial idea; VE, LKS, and MOS developed the idea; VE performed the experimental work; VE drafted the manuscript; VE, LKS, and MOS revised the manuscript.

Conflicts of interest

There are no conflicts to declare.

Acknowledgements

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Paper II

Photodegradable Antimicrobial Agents - Synthesis and Mechanism of Degradation

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Prepared manuscript

Photodegradable Antimicrobial Agents – Synthesis and Mechanism of Degradation

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ABSTRACT: As a strategy to inactivate antimicrobial agents after use we designed a range of ethanolamine derivatives where four of them possessed interesting activity. The ethanolamine moiety facilitates the photodecomposition, which in a potential drug will take place after use. Herein the synthetic preparation of these compounds and mechanism of photoinactivation is described.

INTRODUCTION

Since Alexander Fleming discovered penicillin in 1928,[1] application of this antibiotic has saved millions of lives,[2,3] but excessive use of this and other similar drugs in agriculture and human medicine has resulted in the development of antibiotic-resistant bacteria strains.[4] As a result, many infections that used to be easy to cure, such as pneumonia and post-operative infections, have gradually become a threat.[5,6] Multi-drug resistant (MDR) bacteria are already a global problem, and several health organisations describe the situation as critical,[2,7,8] whereas Mah describes antimicrobial resistance (AMR) as a silent pandemic in a recent commentary.[9] Currently an estimated 700.000 people die annually due to MDR bacteria,[10] and the future situation looks a lot worse with resistance levels projected to rise.[11-13] Despite this prediction, pharmaceutical companies seem uninterested in developing new antibiotics because these drugs will probably not be used in financially sustainable quantities unless existing treatments fail totally.[10]

A consequence of the enormous consumption of antibiotics is high levels of pharmaceuticals and their metabolites in drinking water, waste water, ground water, and coastal waters as well as marine organisms.[14-18] This has led to environmental exposure to antibacterial agents such as ciprofloxacin, sulfamethoxazole, chloramphenicol, and amoxicillin,[19-25] which contribute to the development of antimicrobial resistance.

In recent years technologies have been developed to address this issue. A promising tool is photopharmacology, which apply light to activate and deactivate drugs. [26-28] Another approach is light-induced complete degradation of drugs, of which the photodecomposition of a cephalosporanic acid (**1**) (Figure 1A) is an example;[29] excitation of the nitrobenzyl carbamate moiety leads to ring opening of the β-lactam and formation of hydrazide **2**. Another example is the phosphine-tungsten complex phosphopyricin (**3**), which is degraded and deactivated when exposed to white light (Figure 1B).[30]

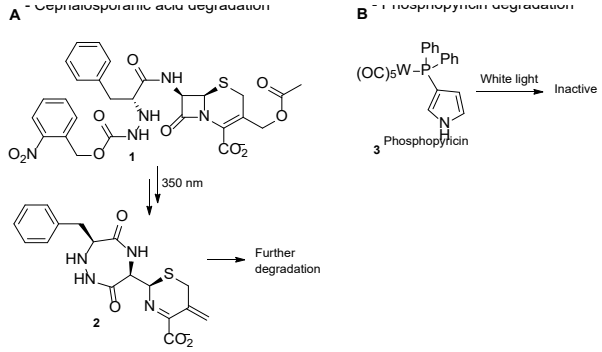


Figure 1. Photodegradable antibiotics (A) Cephalosporanic acid; (B) Phosphopyricin.

We recently communicated synthesis and biological evaluation of four antimicrobial agents based on a new scaffold. which facilitated light-induced fragmentation leading to formation of inactive and non-toxic compounds.[31] This chemistry opens up the possibility to prepare new antimicrobial agents that decompose in the environment after release.

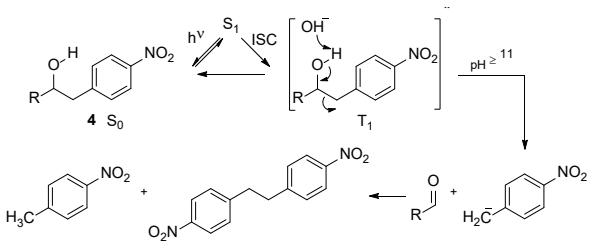
Here a full account of the synthetic work and studies of the photodecomposition mechanisms is presented.

RESULTS AND DISCUSSION

Synthesis of model compounds

Our investigation was inspired by the work of Wan and Muralidharan who studied the so-called photo-retro-aldol reaction, summarized in Scheme 1.[32,33] The idea was to incorporate similar benzylic-alcohol motifs in biologically active molecules that would undergo the same photofragmentation and furnish biologically inactive fragments after use.

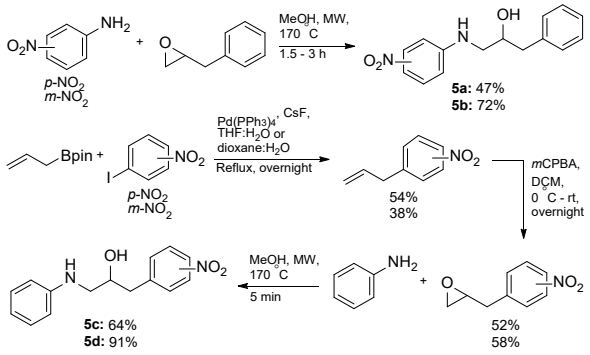
Scheme 1. Photo-retro-aldol type reaction of nitrophenethyl alcohols. R = Ph, CH_3. Irradiation with 254, 300, or 350 nm lamp yields the first excited singlet state (S_1) of the molecules, which undergo intersystem crossing (ISC) to the reactive triplet state (T_1).



As a starting point, we decided to study model compounds closely related to those investigated by Wan and Muralidharan. The candidates were arrived at by attaching a *N*-arylaminomethyl group to C1 in ethanol derivative **4** (Scheme 2), which corresponds to the incorporation of an ethanolamine scaffold. Four such compounds were prepared by treating 2-arylmethylepoxides with three anilines under microwave irradiation (MW). The aryl groups and the anilines contained either a phenyl or a nitrophenyl moiety, which were those utilized by Wan and Muralidharan.[32] When benzyloxirane reacted with *p*- and *m*-nitroaniline, the corresponding aminols 1-(*p*-nitrophenylamino)-3-phenylpropan-2-ol (**5a**) and 1-(*m*-nitrophenylamino)-3-phenyl-propan-2-ol (**5b**) were obtained in moderate yields (Scheme 2). The outcome was better when *p*- and *m*-nitrobenzylepoxide, prepared from allyl boronate by a Suzuki-Miyaura cross-coupling according to Kotha and co-workers[34] followed by treatment with *m*CPBA, were treated with aniline and gave 1-phenylamino-3-(*p*-nitrophenylpropan-2-ol (**5c**) and 1-phenylamino-3-(*m*-aminophenyl)propan-2-ol (**5d**) in moderate overall yield (Scheme 2). The reason for this is probably the better nucleophilicity of aniline compared to the nitroanilines. Attempts were made to improve the yield of the Suzuki-Miayura allylation by running the reaction under anhydrous conditions, but they all failed.

A requirement for photodecomposition of the aminols in the environment is absorption of light with wavelengths above approximately 300 nm. The UV spectra of **5a – 5d** were therefore recorded, and this revealed that whereas **5a** and **5b**, both with a nitrophenylamino moiety attached to C1, had a strong absorption with λ_{max} around 400 nm, **5c** and **5d** barely absorbed light above 330 nm (Figure 2). Under natural conditions, therefore, aminols **5a** and **5b** should absorb light the best and be more prone than **5c** and **5d** to suffer light-induced decomposition.

Scheme 2. Preparation of model compounds 5a – 5d



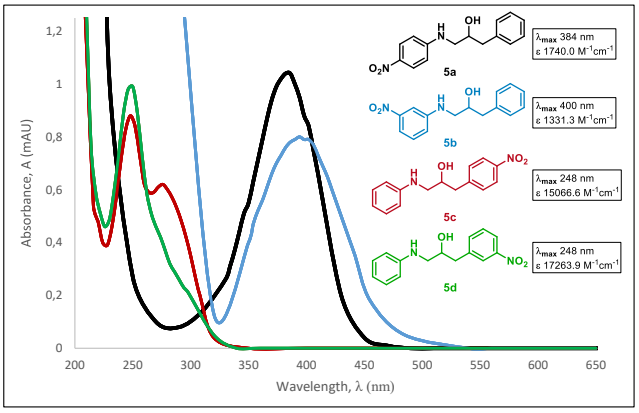


Figure 2. UV/vis spectroscopic data for compounds **5a** - **5d**.

Photodegradation studies on the model compounds

Following the procedure by Wan and Muralidharan,[32] aminoalcohols **5a – 5d** in a mixture of acetonitrile (ACN) and water (7/3, v/v) were irradiated through a Pyrex filter over a wide pH range. Following liquid-liquid extraction with DCM, all reaction mixtures were analysed by ^{1}H NMR spectroscopy. Dramatic differences in decomposition were observed for the different compounds (Table 1). When the nitro group is *para* to the amino moiety (**5a**), a 100% conversion is achieved at pH ≥ 11, whereas the *meta*-analogue (**5b**) undergoes 11% degradation at pH 11 and 17% at pH 13. At pH lower than 7, only trace amounts of decomposition products were detected by ^{1}H-NMR analysis for compound **5b**. For compounds **5c** and **5d** a pH ≥ 11 is required for degradation to occur.

Table 1. Consumption (%) of compounds 5a – 5d at various pH as determined by ^{1}H-NMR analysis

Compound	**5a** (%)	**5b** (%)	**5c** (%)	**5d** (%)
pH 1	9	Trace	ND	ND
pH 3	9	Trace	ND	ND
pH 5	9	Trace	ND	ND
pH 7	32	4	ND	ND
pH 9	75	5	ND	ND
pH 11	100	11	38	20
pH 13	100	17	40	56

ND = no consumption detected.

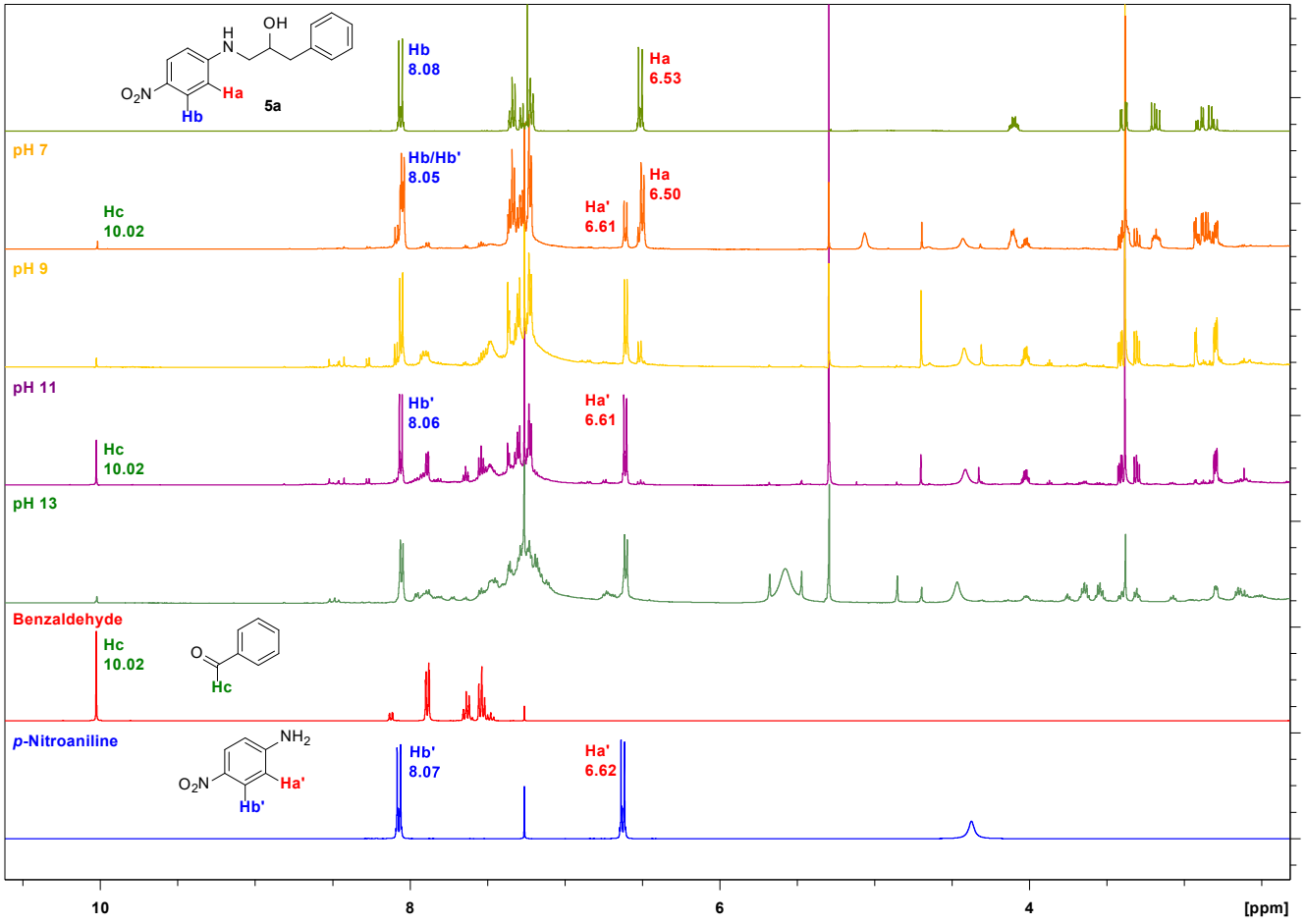
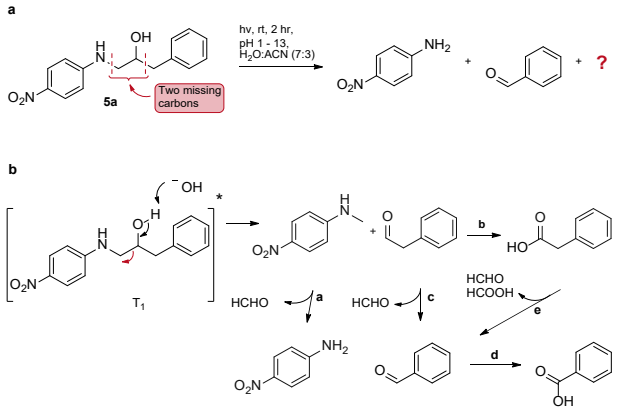


Figure 3. ^{1}H-NMR spectra of the photolysate from compound **5a** irradiated at pH from 7 to 13 and reference spectra of benzaldehyde and *p*-nitroaniline.

Detailed photodecomposition studies were then performed with ethanolamine **5a** at pH > 5 using ^{1}H NMR to monitor the reaction. The development of an aldehyde singlet at δ 10.02 ppm and a doublet at 7.89 ppm (Figure 3d) is consistent with benzaldehyde formation (Figure 3f) and doublets at δ 6.61 and 8.06 ppm are in accordance with formation of *p*-nitroaniline (Figure 3g). At pH 13 (Figure 3e), the ratio between benzaldehyde and *p*-nitroaniline is much lower than at pH 11 (Figure 3d), suggesting further degradation of the aldehyde at higher pH. A conceivable product is benzoic acid this conclusion is supported by a signal at 171.1 ppm in the ^{13}C-NMR spectrum, supposedly due to a carboxylate group. The rate of decomposition is clearly pH dependent; it is peaking around pH 11, and gradually decreasing as the pH drops.

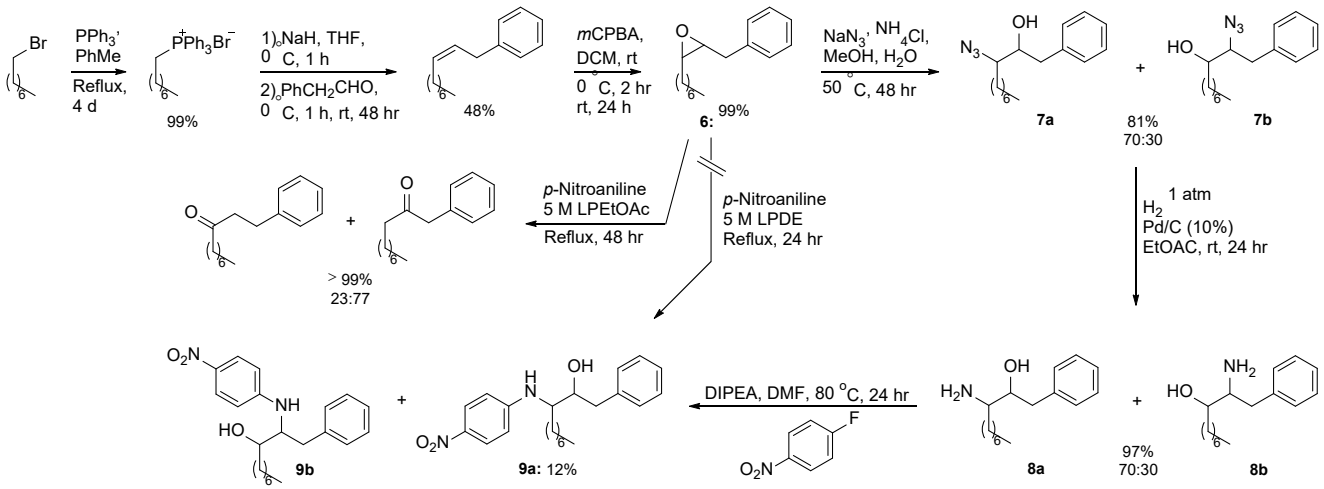
As two of the products are undoubtedly *p*-nitroaniline and benzaldehyde, two carbons are unaccounted for in the analysis of the reaction mixture (Scheme 3a). A reasonable assumption is that the two missing carbons form products that either diffuse into the aqueous phase or disappear as volatiles during work-up. It seems likely that the suggested products originate from a photo-retro-aldol type reaction, initially forming *N*-methyl-4-nitroaniline and phenylacetaldehyde (Scheme 3). The former product, known to undergo photochemical *N*-demethylation,[35,36] subsequently reacts and furnish formaldehyde and *p*-nitroaniline (Scheme 3b, reaction a). Phenylacetaldehyde then undergoes oxidation to phenylacetic acid (reaction b) (which can suffer cleavage and form benzaldehyde and formaldehyde/formic acid (reaction e)[37]) or reacts in a Norrish type 1 process and forms formaldehyde and benzaldehyde (reaction c). And indeed, phenylacetaldehyde was converted to benzaldehyde when irradiated under the same conditions as compounds **5a**-**5d**.

Scheme 3. Suggested reaction pathways for photodegradation of compound 5a



To shed more light on the decomposition process, we designed an analogue to **5a** with a long alkyl chain at C1 to

prevent that the two carbon atoms not accounted for in the ethanolamine scaffold do not disappear during work-up of the photolysate. The synthesis (Scheme 4) commenced with a Wittig reaction involving octyltriphenylphosphonium bromide and phenylacetaldehyde resulting in formation of the desired compound as a 94:6 mixture of the *Z*/*E* isomers as evident from the integration of the doublets at 3.41 ppm and 3.34 ppm in the ^{1}H NMR spectra, respectively, a method used by Krasovskaya et al. in order to establish *Z*/*E* ratio for similar products.[38] The mixture was subjected to epoxidation upon treatment with *m*CPBA. Attempts were made to react the resulting oxirane **6** with *p*-nitroaniline, but due to the poor nucleophilicity of the latter combined with steric hindrance no reaction occurred in a 5 M lithium perchlorate diethyl ether (LPDE) solution at 40 °C. When compound **6** was treated with LPEtOAc and the temperature increased to 80 °C, a reaction took place, but not the right one; instead, a lithium-promoted epoxide-carbonyl rearrangement occurred, giving two decanones as the only products. However, treatment of **6** with sodium azide gave an inseparable mixture of the two azidodecanols **7a** and **7b** in a 7:3 ratio as evident from ^{1}H NMR analysis, which upon hydrogenation and subsequent nucleophilic aromatic substitution with 1-fluoro-4-nitrobenzene afforded a mixture of the target compound 3-((4-nitrophenyl)amino)-1-phenyldecan-2-ol (**9a**) and its regioisomer **9b**. The low yield of the desired product, viz. **9a**, was due to overlapping fractions, the yield was only 12%.

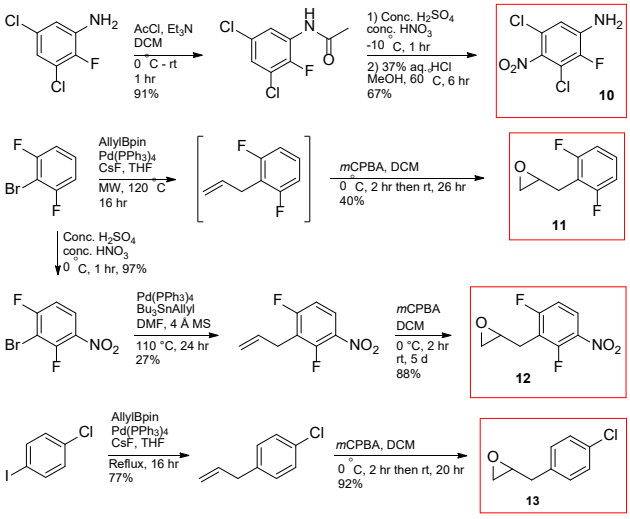


Scheme 4. Synthesis of 3-aminodecan-2-ol derivative 9a.

If aminoalcohol **9a** undergoes degradation just like **5a** when irradiated, octanal should be present in the reaction mixture as should *p*-nitroaniline and benzaldehyde. That was indeed the case (see Figure S1 in supporting information); in addition to the signals from the aromatic compounds, the ^{1}H-NMR spectrum of the hydrolysate showed an aldehyde triplet at 9.76 ppm with a coupling constant of 1.9 Hz and COSY correlations to the alkyl chain. After three days at room temperature, the aldehyde was completely converted to the corresponding carboxylic acid, as evident by a HMBC correlation from the alpha methylene protons at 2.32 ppm to a carbonyl signal at 173.7 ppm and further confirmed by low-resolution mass spectrometry.

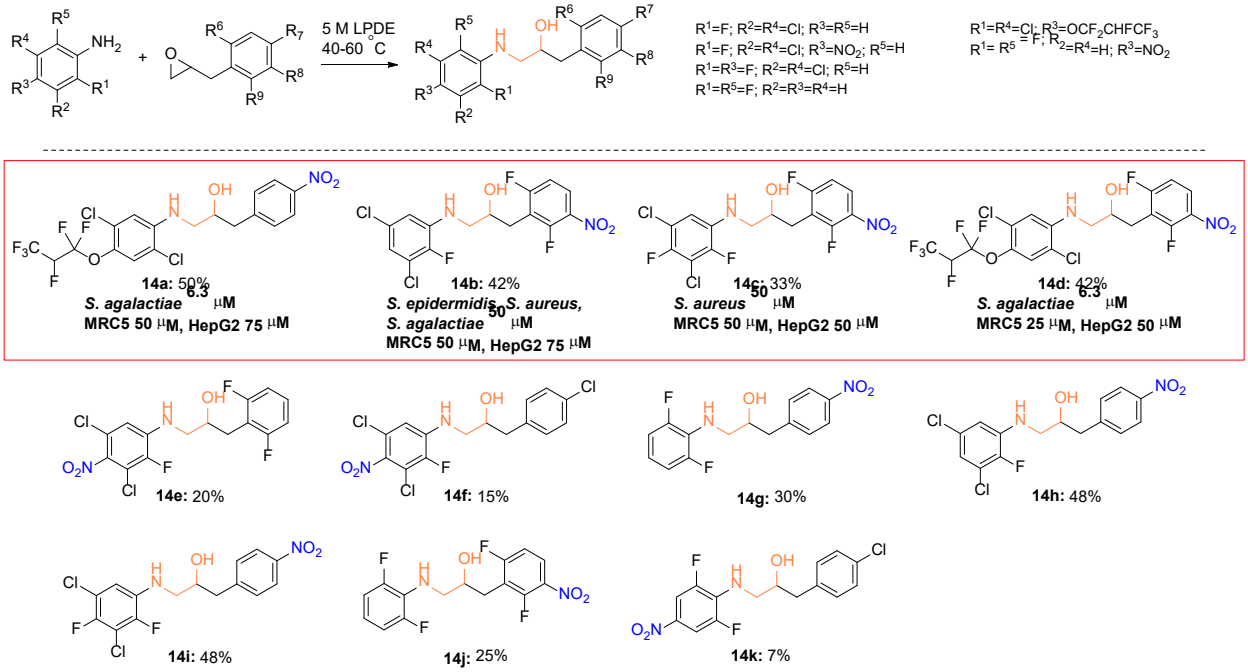
Synthesis of additional ethanolamine derivatives

With the photodecomposition process essentially proved, we started the search for biologically active analogues to **5a** by reacting benzyloxirane derivatives with anilines containing aryl groups that have been found in other molecules showing biological activity. In order to make a range of compounds, some key building blocks, one aniline and three epoxides, were prepared as summarized in Scheme 5.[31] The aniline (**10**) was synthesised from the corresponding trihaloaniline by performing acetylation followed by nitration and acidic hydrolysis, which furnished **10** in 61% overall yield. Two of the epoxides were prepared from 1-bromo-2,6-difluorobenzene. A standard Suzuki-Miyaura cross-coupling with allylboronic acid pinacol ester furnished 2-allyl-1,3-difluorobenzene, which was treated with *m*CPBA to afford 2,6-difluorobenzyloxirane (**11**) in 40% yield over two steps. The other was 2,6-difluoro-3-nitrobenzyloxirane (**12**), which was obtained in 23% overall yield by nitration followed by a Stille cross-coupling with allyltributylstannane and then epoxidation with *m*CPBA. Finally, the conversion of *p*-chloroiodobenzene to epoxide 4-chlorobenzyloxirane (**13**) was achieved in 71% overall yield by the same synthetic route that was used to prepare **11**.



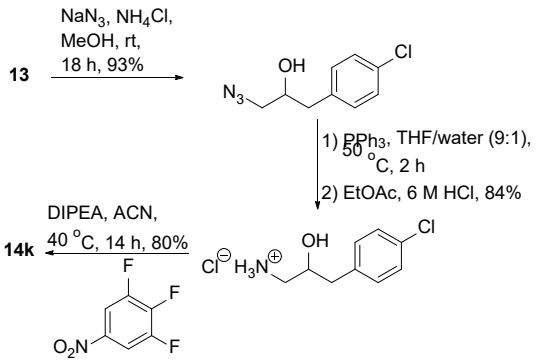
Scheme 5. Synthesis of some key building blocks for the preparation of additional ethanolamine derivatives

With these building blocks at hand, a number of target molecules could be synthesized using a Lewis acid-promoted epoxide ring-opening reaction in a 5 M solution of lithium perchlorate in diethyl ether (LPDE), to which the anilines and epoxides were added (Scheme 6). No product was obtained in better than 50% yield, conceivably because the anilines react sluggishly because they are rather poor nucleophiles due to the electron-withdrawing substituents. The worst case was observed when 2,6-difluoro-4-nitroaniline reacted with oxirane **13** and gave the expected ethanolamine, 3-(4-chlorophenyl)-1-(2,6-difluoro-4-nitrophenyl)aminopropan-2-ol (**14k**) in 7% yield only.



Scheme 6. Preparation of aminols. R¹=F, Cl; R²=Cl, H; R³=NO₂, F, H, OCF₂CHFCF₃; R⁴=Cl, H; R⁵=F, H; R⁶=F, H; R⁷=NO₂, Cl, H; R⁸=NO₂, H, R⁹=F, H.

2,6-Difluoro-4-nitroanline reacted poorly under these conditions, and compound **14k** was therefore prepared in an *umpolung*-type approach, with a reversal of the roles of nucleophile and electrophile (Scheme 7, Method B). Thus, ring-opening of epoxide **13** with sodium azide followed by a Staudinger reduction gave the desired 1,2-amino alcohol, which readily reacted with 1,2,3-trifluoro-5-nitrobenzene to yield the target compound in 80% yield.



Scheme 7. Formation of 14k using Method B.

Screening of biological activity

As already reported, ethanolamine derivatives **14a – 14d** have been screened against selected Gram-positive and Gram-negative bacteria, including *E. Faecalis*, *E. Coli*, *P. Aeruginosa*, *S. Aureus*, *S. Agalactiae*, and *S. Epidermidis* and showed promising antibacterial activity with a minimum inhibitory concentration (MIC) as low as 6.3 μM in the best cases (**14a** and **14d**) (Scheme 6).[31] It was therefore very disappointing when all of the remaining seven analogues were totally inactive (MIC > 100 μM). More compounds have to be prepared and tested before the reason for this difference can be elucidated, but four noteworthy trends have been observed. Firstly, all the active compounds have a nitro group attached to the aryl group closer to the hydroxyl group. Then, none of the compounds with a nitroaryl group attached to the amino substituent (**14e**, **14f** and **14k**) exhibit any activity. Furthermore, only the most active compounds have an alkoxyaryl group attached to the amino substituent. And finally, the biological activity (and lack of activity) is the result of impact from both aryl moieties because even though **14g – 14j** have a nitroaryl motif closer to the OH group, they are totally inactive even at a 100 μM level. On the basis of these observations, additional compounds will be synthesized and tested.

*Photochemical decomposition of aminols **14a - 14d***

The four active compounds, viz. aminols **14a**-**14b**, were subjected to photodecomposition studies at pH 8 and 13. All four compounds degraded completely at pH 13, with the exception of compound **14d**, which displayed 56% conversion. Amino alcohol **14a** decomposed following the same reaction pathway as compound **5a**, yielding 2,5-Dichloro-4-(1,1,2,3,3,3-hexafluoropropoxy)aniline and 4-nitrobenzoic acid (Figure S2 in supporting information). The same is observed for the three other compounds since the expected anilines were detected by ^{1}H NMR. However, stability

studies performed in the dark for compounds **14a**-**14d** at pH 13 revealed that **14b**, **14c**, and **14d** are somewhat unstable in basic environment. An S_NAr reaction with hydroxide occurs, substituting one of the fluorine atoms, which was confirmed by LRMS, ^1H NMR, and ^{19}F NMR. At pH 13, around half of the starting material underwent this unwanted reaction and the rest photodecomposed. However, at pH 8 this was not a problem since all compounds were stable for at least 24 h at room temperature. To our satisfaction we found that compound **14b** and **14d** was completely consumed in the photochemical reaction at pH 8 (Table 2), but they are not following the photo-retro-aldol pathway described for aminol **5a** and confirmed by compound **9a**.

Table 2. Photodecomposition conversions (%) at pH 8 and 13 for compounds 14a-14d.[a]

Compound	Photolysis[a]		Stability[b]	
	pH 8 (%)	pH 13 (%)	pH 13	pH 8
14a	25	100	Yes	Yes
14b	100	100	No	Yes
14c	19	100	No	Yes
14d	100	56	No	Yes

[a]Conversions were estimated by ^1H NMR spectra of the crude reaction mixtures after aqueous workup. [b]Performed for 24 h in the dark at rt.

For aminol **14b**, an intramolecular S_NAr reaction occurs, yielding compound **S1**, as evident from four sharp doublet of doublets appearing in the ^1H NMR spectrum of the crude degradation mixture at 7.76, 7.58, 7.41, and 6.23 ppm (Figure S3 in supporting information). There are only two fluorine atoms present according to ^{19}F NMR (Figure S4B), and the $^3J_{HH}$, $^4J_{HF}$, and $^5J_{HF}$ couplings of 9.3, 8.9, and 1.6 Hz, respectively, confirm this structure. The other possible isomer from an S_NAr reaction, compound **S2**, would have displayed a larger $^3J_{HF}$,[39] but this is not observed (Figure S4B in supporting information). A similar reaction occurred in the photodegradation of compound **14c**, as illustrated by the loss of one fluorine atom in the ^{19}F NMR spectrum of the crude degradation mixture (Figure S4D in supporting information), but the conversion was only 19% at pH 8. The same characteristic signals did not appear in the ^1H NMR spectrum of the degradation of compound **14d**, suggesting that a different mechanism is involved in the degradation of **14d**. However, it was not possible to elucidate any degradation products due to a complicated NMR spectrum with many unresolved multiplets and broad signals indicating that the compound has fully decomposed. Research into this is currently ongoing.

The crude degradation mixtures derived from **14a**-**14d** displayed no activity against the same bacteria strains, as well as no toxicity against MRC5 and HepG2 cell lines.

CONCLUSION

The synthesis of twelve compounds possessing our aminol scaffold that facilitates photodecomposition or photoinitiated S_NAr is described. Four of the compounds possessed antimicrobial activity and their corresponding decomposition products had no antimicrobial activity or toxicity. Through NMR analysis we have elucidated the products derived from the photochemical reactions enabling the establishment of mechanisms for the formation of the photochemical products. Compound **14d**, which was one of the two most active compounds prepared, decomposes totally in the course of 24 hours at pH 8 making this a very interesting lead for further optimalisation of biological activity. Work towards this end is ongoing in these laboratories.

EXPERIMENTAL SECTION

GENERAL INFORMATION

All chemicals were obtained from Sigma Aldrich/Merck or VWR and used as supplied. When specified, solvents were dried by storing over 4 Å molecular sieves. For petroleum ether (pet. ether), the 40−60 °C fraction was used. All reactions were carried out under a nitrogen or argon atmosphere, unless otherwise specified. Microwave-assisted experiments were performed in a CEM Focused Microwave™ Synthesis System, model type Discover, operating at 0-300 W and pressure range of 0-290 psi.

Thin-layer chromatography (TLC) was carried out with silica gel (60 F_{254}) on aluminium sheets with solvent systems consisting of various mixtures of pet. ether, ethyl acetate, and dichloromethane. Visualization was achieved with either UV light (254 and/or 365 nm) or a potassium permanganate stain. Flash chromatography was performed with a hand pump and 230-400 mesh silica gel or an Interchim Puriflash 215 autoflash chromatography system with Biotage Snap Ultra HP-Sphere™ 25 µm silica-gel columns.

IR spectra were recorded on an Agilent Cary 630 FT-IR spectrophotometer equipped with an attenuated total reflectance (ATR) attachment. Samples were analyzed neat on a ZnSe crystal and the absorption frequencies are given in wave numbers (cm^{-1}).

UV-vis spectra were obtained on an Agilent 8453 single-beam UV-vis spectrophotometer with a deuterium-discharge lamp for the UV range and a tungsten lamp for the visible wavelength range. Samples were analyzed in an Agilent open-top UV quartz cell (10 mm, 3.0 mL) with ethanol as solvent. The wavelengths are reported in nm and molar attenuation coefficients in $M^{-1}cm^{-1}$.

NMR spectra were recorded on a Bruker Ascend™ 400 spectrometer (400.13 MHz for ^1H, 100.61 MHz for ^{13}C, 376.46 MHz for ^{19}F, and 161.98 MHz for ^{31}P). Coupling constants (*J*) are given in Hz and the multiplicity is reported as singlet (s), doublet (d), triplet (t), quartet (q), sextet (s), multiplet (m), and broad singlet (bs). Chemical shifts are reported in ppm in the order downfield to upfield and calibration is done using the residual solvent signals for chloroform-*d* (^1H 7.26 ppm; ^{13}C 77.16 ppm), acetonitrile-d_3 (^1H 1.94 ppm; ^{13}C 1.32 ppm), or water-d_2 (^1H 4.79 ppm).[40] Calibration for ^{19}F NMR is done using α,α,α-trifluorotoluene as internal standard in chloroform-*d* (-62.61 ppm) and acetonitrile-d_3 (-63.10 ppm).[41]

High-resolution mass spectra were obtained on a JEOL AccuTOF™ T100GC mass spectrometer. The instrument was operated with an orthogonal electrospray ionization source (ESI), an orthogonal accelerated time of flight (TOF) single stage reflectron mass analyzer and a dual micro channel plate (MCP) detector at the following instrumental settings/conditions; ionization mode: positive, desolvating temperature/ion source temperature = 250 °C, needle

voltage = 3000 V, desolvation gas flow = 2.0 L min^{-1}, nebulizing gas flow = 1.0 L min^{-1}, orifice1 temperature = 120 °C, orifice1 voltage = 24 V, ring lens voltage = 12 V, orifice2 voltage = 6 V, ion guide peak voltage = 800 V, detector voltage = 2300 V, acquisition range = 4-1000 *m/z*, spectral recording interval = 0.5 ns. Mass calibration were performed using the internal standard method and mass drift compensation was performed in each acquisition. Low-resolution mass spectra were recorded on an Advion expression^S compact mass spectrometer (CMS) operated in ESI mode equipped with a Plate Express™ TLC plate reader for sample injection. A solution of ammonium acetate (5.0 mM) and formic acid (0.05%) in acetonitrile and water (95:5) was used as mobile phase for both positive and negative ESI modes.

Synthesis of 1-(4-nitrophenylamino)-3-phenylpropan-2-ol (5a). A sealed reactor tube was charged with 4-nitroaniline (0.28 g, 2.0 mmol), (2,3-epoxypropyl)benzene (0.29 mL, 296 mg, 2.2 mmol), and methanol (0.50 mL). The reaction mixture was irradiated in a microwave reactor at 160-170 °C for 3 h. The mixture was evaporated onto celite and the crude product was isolated by silica-gel flash chromatography (DCM/MeOH 98:2). Concentration of the relevant fractions (R_f = 0.43 in DCM/MeOH 99:1) yielded **5a** as a yellow solid (0.26 g, 47%, mp. 111-112 °C). IR (neat): ν_{max} 3392, 3029, 2921, 1600, 1307; UV-vis: λ_{max} (EtOH) 384 nm (ε 1740 $cm^{-1}M^{-1}$); ^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.08 (d, *J* = 9.2 Hz, 2H), 7.38-7.33 (m, 2H), 7.31-7.27 (m, 1H), 7.24-7.22 (m, 2H), 6.53 (d, *J* = 9.2 Hz, 2H), 4.15-4.09 (m, 1H), 3.41 (dd, *J* = 13.0 Hz, 3.4 Hz, 1H), 3.20 (dd, *J* = 13.0 Hz, 7.8 Hz, 1H), 2.92 (dd, *J* = 13.6 Hz, 5.1 Hz, 1H), 2.83 (dd, *J* = 13.6 Hz, 8.1 Hz, 1H); ^{13}C NMR (100.61 MHz, $CDCl_3$): δ 153.4, 138.3, 137.1, 129.4, 129.0, 127.2, 126.5, 111.5, 71.1, 48.4, 41.9; HRMS: Calcd for $C_{15}H_{15}N_2O_3$ $[M-H]^-$ 271.10827, found 271.10866.

Synthesis of 1-(3-nitrophenylamino)-3-phenylpropan-2-ol (5b). A sealed tube was charged with 3-nitroaniline (0.28 g, 2.0 mmol), (2,3-epoxypropyl)benzene (0.27 mL, 275 mg, 2.0 mmol), and methanol (0.50 mL). The reaction mixture was irradiated in a microwave reactor at 170-180 °C for 90 min. The mixture was evaporated onto celite and the crude product was isolated by flash column chromatography (DCM/pet. ether 7:3). Concentration of the relevant fractions (R_f = 0.45 in DCM/MeOH 99:1) yielded **5b** as a yellow solid (0.39 g, 72%, mp. 74-77 °C). IR (neat): ν_{max} 3545, 3302, 3081, 2915, 1618; UV-vis: λ_{max} (EtOH) 400 nm (ε 1331 $M^{-1}cm^{-1}$); ^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.53 (dd, *J* = 8.0 Hz, 1.9 Hz, 1H), 7.40-7.33 (m, 3H), 7.30-7.23 (m, 4H), 6.88 (dd, *J* = 8.0 Hz, 1.5 Hz, 1H), 4.15-4.09 (m, 1H), 3.36 (dd, *J* = 12.6 Hz, 2.7 Hz, 1H), 3.15 (dd, *J* = 12.6 Hz, 7.8 Hz, 1H), 2.92 (dd, *J* = 13.6 Hz, 4.9 Hz, 1H), 2.87 (dd, *J* = 13.6 Hz, 8.0 Hz, 1H); ^{13}C NMR (100.61 MHz, $CDCl_3$): δ 149.5, 149.0, 137.3, 129.9, 129.5, 129.0, 127.1, 119.4, 112.5, 106.8, 71.1, 49.1, 41.8; HRMS: Calcd for $C_{15}H_{15}N_2O_3$ $[M+H]^+$ 273.1234, found 273.1236.

1-Allyl-4-nitrobenzene. An oven-dried round-bottom flask fitted with a condenser was charged with 1-iodo-4-nitrobenzene (1.00 g, 4.00 mmol), CsF (1.52 g, 10.0 mmol), $Pd(PPh_3)_4$ (0.70 g, 15 mol%), THF (20 mL), and water (2 mL). The mixture was stirred at rt under Ar for 30 min, followed by addition of allylboronic acid pinacol ester (1.36 mL, 7.20 mmol) in THF (8 mL). The reaction mixture was refluxed (oil bath, 95 °C) for 22 h. After cooling to rt the product mixture was evaporated onto celite and purified by silica-gel column chromatography (pet. ether). Concentration of the relevant fractions (R_f = 0.47 in pet. ether/EtOAc 8:2)) gave the title compound (0.35 g, 54%) as a slightly yellow liquid. Spectroscopic data are in accordance with data reported in the literature.[42]

Attempt to prepare 2-(4-nitrobenzyl)oxirane under anhydrous conditions: formation of 3-methyl-2-(4-nitrophenyl)oxirane. An oven-dried round-bottom flask fitted with a condenser was charged with 1-iodo-4-nitrobenzene (996 mg, 4.00 mmol), CsF (2.127 g, 14.0 mmol), $Pd(PPh_3)_4$ (231 mg, 5 mol%), allylboronic acid pinacol ester (1.34 g, 8.00 mmol), and THF (50 mL). The reaction mixture was refluxed (oil bath, 80 °C) for 18 h. After cooling to rt the product mixture was evaporated onto celite and purified by silica-gel flash chromatography (pet. ether). Concentration of the relevant fractions yielded a slightly yellow residue, which was dissolved in DCM (15 mL) and cooled (ice/water bath). *m*CPBA (632 mg, 2.82 mmol) was added and the reaction mixture was stirred at 0 °C for 2 h and rt for 22 h before quenching with 1:1 sat. $NaHCO_3$:10% $Na_2S_2O_3$ (20 mL). The phases were separated, and the aq. layer was extracted with DCM (3 x 15 mL). The combined organic phases were washed with 1:1 sat. $NaHCO_3$:10% $Na_2S_2O_3$ (20 mL), sat. aq. $NaHCO_3$ (2 x 20 mL), water (20 mL), brine (20 mL), dried ($MgSO_4$), filtered, and concentrated in vacuo to yield 3-methyl-2-(4-nitrophenyl)oxirane[43-45] as a white solid (348 mg, 49% over two steps, mp. 79-81 °C; lit.[43] mp 87-88 °C, lit.[45] mp 90-92 °C). R_f = 0.60 in pet. ether/EtOAc 6:4; IR (neat): ν_{max} 3109, 3073, 2977, 2933, 2855, 1601, 1513; ^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.20 (d, *J* = 8.8 Hz, 2H), 7.42 (d, *J* = 8.8 Hz, 2H), 3.67 (d, *J* = 1.9 Hz, 1H), 3.02 (qd, *J* = 5.1 Hz, 1.9 Hz, 1H), 1.50 (d, *J* = 5.1 Hz, 3H); ^{13}C NMR (100.61 MHz, $CDCl_3$): δ 147.9, 145.5, 126.4, 123.9, 60.0, 58.5, 18.0.

Synthesis of 2-(4-nitrobenzyl)oxirane. An oven-dried round-bottom flask charged with 1-allyl-4-nitrobenzene (0.35 g, 2.16 mmol) in anhydrous DCM (12 mL) was cooled (ice/water bath) and stirred for 5 min under Ar followed by addition of *m*CPBA (0.59 g, 2.63 mmol). The reaction mixture was stirred at 0 °C for 2 h, then at rt for 15 h. More *m*CPBA (0.12 g, 0.54 mmol) was added and stirring continued for another 31 h before quenching the reaction with aq. NaOH solution (1 M, 10 mL). The phases were separated, and the aq. phase was extracted with DCM (3 x 15 mL). The combined organic phases were washed with water (20 mL), brine (20 mL), dried ($MgSO_4$), filtered, and concentrated in vacuo. The product was isolated by silica-gel autoflash chromatography (pet. ether/EtOAc/DCM 93:2:5 → 40:55:5) and concentration of the relevant fractions (R_f = 0.26 (pet. ether/DCM 1:1)) yielded 2-(4-nitrobenzyl) oxirane as a yellow oily liquid (0.20 g, 52%). The spectroscopical data were in full accord with the previously reported data.[46]

Synthesis of 3-(4-nitrophenyl)-1-(phenylamino)propan-2-ol (5c). A sealed tube was charged with aniline (0.15 mL, 1.67 mmol), 2-(4-nitrobenzyl)oxirane (0.30 g, 1.67 mmol), and methanol (0.5 mL). The reaction mixture was irradiated in a microwave reactor (170-180 °C, 9.5 bar, 300 W, 5 min ramping) for 5 min. The mixture was evaporated

onto celite and the crude product was isolated by silica-gel autoflash column chromatography (pet. ether/EtOAc/DCM 90:5:5 → 45:50:5). Concentration of the relevant fractions (R_f = 0.20 in DCM/MeOH 99:1) yielded **5c** as a yellow solid (0.29 g, 64%, mp. 90-93 °C). IR (neat): ν_{max} 3326, 3054, 2919, 1598, 1506; UV-vis: λ_{max} (EtOH) 248 nm (ε 15067 $M^{-1}cm^{-1}$); ^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.17 (d, *J* = 8.6 Hz, 2H), 7.42 (d, *J* = 8.6 Hz, 2H), 7.19 (dd, *J* = 8.5 Hz, 7.4 Hz, 2H), 6.77 (t, *J* = 7.4 Hz, 1H), 6.66 (d, *J* = 8.5 Hz, 2H), 4.16-4.10 (m, 1H), 3.32 (dd, *J* = 13.1 Hz, 3.4 Hz, 1H), 3.12 (dd, *J* = 13.1 Hz, 8.1 Hz, 1H), 2.99 (dd, *J* = 13.8 Hz, 4.7 Hz, 1H), 2.92 (dd, *J* = 13.8 Hz, 8.0 Hz, 1H); ^{13}C NMR (100.61 MHz, $CDCl_3$): δ 147.9, 146.9, 146.1, 130.3, 129.5, 123.8, 118.5, 113.5, 70.7, 49.9, 41.2; HRMS: Calcd for $C_{15}H_{15}N_2O_3$ $[M+H]^+$ 273.1234, found 273.1236.

Synthesis of 1-allyl-3-nitrobenzene. A round-bottom flask fitted with a condenser was charged with 1-iodo-3-nitrobenzene (1.99 g, 8.00 mmol), CsF (3.65 g, 24.0 mmol), $Pd(PPh_3)_4$ (1.38 g, 15 mol%), THF (55 mL), and water (15 mL). The mixture was stirred at rt under Ar for 30 min, followed by addition of allylboronic acid pinacol ester (2.72 mL, 14.4 mmol). The reaction mixture was refluxed (oil bath, 95 °C) for 23 h. $Pd(PPh_3)_4$ (0.46 g, 5 mol%), CsF (1.22 g, 8.00 mmol), and allylboronic acid pinacol ester (0.75 mL, 4.00 mmol) were added. THF was removed under reduced pressure and replaced with dioxane (55 mL) followed by reflux (oil bath, 135 °C) for 28 h. After cooling to rt the product mixture was evaporated onto celite and purified by silica-gel autoflash chromatography (pet. ether/DCM 95:5). Concentration of the relevant fractions (R_f = 0.53 in pet. ether/EtOAc 8:2) gave the title compound (0.50 g, 38%) as a slightly yellow liquid. Spectroscopic data are in accordance with data reported in the literatureliterature.[42]

Synthesis of 2-(3-nitrobenzyl)oxirane. A dry 25 mL round-bottom flask charged with 1-allyl-3-nitrobenzene (0.49 g, 3.00 mmol) in dry DCM (15 mL) was cooled (ice/water bath) and stirred for 5 min under Ar followed by addition of *m*CPBA (1.35 g, 6.02 mmol). The mixture was stirred at ambient temperature for 2 hr, then at rt for 26 hr before being quenched with aq NaOH solution (1 M, 20 mL). The phases were separated and the aq phase was extracted with DCM (3 x 15 mL). The combined organic phases were washed with water (20 mL) and brine (20 mL), dried ($MgSO_4$), filtered, and concentrated *in vacuo*. The product was isolated by silica-gel autoflash chromatography (pet. ether/EtOAc/DCM 90:5:5 → 35:60:5) and concentration of the relevant fractions (R_f = 0.28 in pet. ether/EtOAc/DCM 80:15:5) furnished the title compound as a yellowish oil (0.31 g, 58%). IR (neat): ν_{max} 3060, 2989, 2924, 1522, 1348; ^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.13-8.11 (m, 2H), 7.62-7.60 (m, 1H), 7.51-7.48 (m, 1H), 3.21-3.17 (m, 1H), 3.06 (dd, *J* = 14.8 Hz, 4.4 Hz, 1H), 2.92 (dd, *J* = 14.8 Hz, 6.3 Hz, 1H), 2.85-2.83 (m, 1H), 2,55 (dd, *J* = 4.8 Hz, 2.6 Hz, 1H); ^{13}C NMR (100.61 MHz, $CDCl_3$): δ 148.5, 139.3, 135.4, 129.6, 124.0, 122.0, 51.8, 46.8, 38.3; HRMS: Sample did not ionize in ESI mode.

Synthesis of 3-(3-nitrophenyl)-1-(phenylamino)propan-2-ol (5d). A sealed reactor tube was charged with aniline (0.09 mL, 92 mg, 1.00 mmol), 2-(3-nitrobenzyl)oxirane (0.18 g, 1.00 mmol), and MeOH (0.5 mL). The reaction mixture was irradiated in microwave reactor (170-180 °C, 9.5 bar, 300 W, 5 min ramping) for 5 min. The mixture was evaporated onto celite, and the product was isolated by silica-gel autoflash column chromatography (pet. ether/EtOAc/DCM 90:5:5 → 45:50:5). Concentration of the relevant fractions (R_f = 0.52 in DCM/MeOH 95:5) gave **5d** as a brown oily wax (0.25 g, 91%). IR (neat): ν_{max} 3393, 3352, 3053, 3024, 2920, 1602; UV-vis: λ_{max} (EtOH) 248 nm (ε 17019 $M^{-1}cm^{-1}$). ^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.14-8.13 (m, 1H), 8.10 (ddd, *J* = 8.2 Hz, 2.2 Hz, 1.0 Hz, 1H), 7.60-7.58 (m, 1H), 7.50-7.46 (m, 1H), 7.19 (dd, *J* = 8.5 Hz, 7.4 Hz, 2H), 6.76 (tt, *J* = 7.4 Hz, 1.0 Hz, 1H), 6.64 (dd, *J* = 8.5 Hz, 1.0 Hz, 2H), 4.13-4.07 (m, 1H), 3.32 (dd, *J* = 13.1 Hz, 3.4 Hz, 1H), 3.11 (dd, *J* = 13.1 Hz, 8.2 Hz, 1H), 2.98 (dd, *J* = 14.0 Hz, 4.6 Hz, 1H), 2.90 (dd, *J* = 14.0 Hz, 8.2 Hz, 1H); ^{13}C NMR (100.61 MHz, $CDCl_3$): δ 148.5, 148.0, 140.3, 135.8, 129.49, 129.48, 124.3, 121.8, 118.4, 113.5, 70.7, 49.9, 40.9; HRMS: Calcd for $C_{15}H_{15}N_2O_3$ $[M+H]^+$ 272.1234, found 273.1233.

General procedure for the photodecomposition of 5a-5d. A solution of the appropriate compound (≈0.10 mmol) in acetonitrile was added to a photochemical reactor containing distilled water at the appropriate pH to a concentration of ≈0.7 mM and a total volume of either 75 or 150 mL (water/ACN 7:3), depending on the reactor size. The reaction vessel was either purged with N_2 during the reaction or left open to air. The reaction mixture was photolyzed with a 125 W medium-pressure (300-600 nm irradiation) mercury-vapour lamp. After completion, the reaction mixture was transferred to a separatory funnel, saturated with NaCl, and extracted with EtOAc (3 x 50 mL). The aqueous phase was then adjusted to pH ≈2 with HCl (1 M) and the aqueous layer was extracted again with EtOAc (3 x 50 mL). The combined organic fractions were dried ($MgSO_4$), filtered and concentrated in vacuo on a rotary evaporator to yield a residue which was analysed by ^{1}H NMR.

Photolysis of phenylacetaldehyde. A solution of phenylacetaldehyde (110 μL, 11.3 mg, 0.094 mmol) in acetonitrile (45 mL) was added to a photochemical reactor containing distilled water at pH 13. The reaction mixture was photolyzed with a 125 W medium-pressure (300-600 nm irradiation) mercury-vapour lamp for 2 h open to air. The resulting reaction mixture was extracted with DCM (3 x 20 mL). The aqueous phase was then adjusted to pH ≈2 with HCl (1 M) and the aqueous layer was extracted again with DCM (3 x 20 mL). The combined organic fractions were dried ($MgSO_4$), filtered and concentrated in vacuo on a rotary evaporator to yield a residue which was analysed by ^{1}H NMR.

Synthesis of octyltriphenylphosphonium bromide. A solution of triphenylphosphine (1.570 g, 6.00 mmol) and octyl bromide (1.14 mL, 1.275 g, 6.60 mmol) in toluene (20 mL) was refluxed (oil bath, 135 °C) for 4 d. An oily fraction was formed and when the reaction mixture had reached rt, toluene was decanted off. The residue was rinsed with toluene (3 x 10 mL) to remove excess octyl bromide, and this gave the title compound as a colourless syrup (2.70 g, 99%). IR (neat): ν_{max} 3390, 3051, 2923, 2853, 1586, 1436; ^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.90-7.84 (m, 6H), 7.81-7.76 (m, 3H), 7.72-7.67 (m, 6H), 3.90-3.83 (m, 2H), 1.64-1.61 (m, 4H), 1.25-1.19 (m, 10H), 0.83 (t, *J* = 6.9 Hz, 3H); ^{13}C NMR (100.61 MHz, $CDCl_3$): δ 135.0 (d, *J* = 3.0 Hz), 133.9 (d, *J* = 10.0 Hz), 130.6 (d, *J* = 12.5 Hz), 118.7 (d, *J* = 85.7 Hz), 31.8, 30.5

(d, J = 15.4 Hz), 29.4, 29.0, 23.2, 22.8 (d, J = 4.5 Hz), 22.7, 14.2; ^{31}P NMR (161.98 MHz, $CDCl_3$): δ 24.6; HRMS: Calcd for $C_{26}H_{32}P$ $[M\text{-}Br]^+$ 375.22361, found 375.22380.

Synthesis of 1-phenyldec-2-ene. To a stirred solution of octyltriphenylphosphonium bromide (2.70 g, 5.92 mmol) in dry THF (30 mL) was added sodium hydride as a 60% suspension in mineral oil (237 mg, 5.92 mmol). After 2 h of stirring at rt, the solution was cooled (ice/water bath) and phenylacetaldehyde (0.69 mL, 745 mg, 5.92 mmol) was added. The reaction mixture was stirred for 1 h at bath temperature and then for 48 h at rt. THF was removed under reduced pressure, water (20 mL) was added, and the hydrolysate was extracted with DCM (3 x 15 mL). The combined organic layers were concentrated onto celite, and the title compound was isolated by silica-gel flash chromatography (pet. ether). Concentration of the relevant fractions (R_f = 0.56 in pet. ether) yielded a 94:6 mixture of *Z*:*E* isomers of the title compounds as a colourless liquid (617 mg, 48%). IR (neat): ν_{max} 3011, 2923, 2853, 1602, 1453; ^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.31-7.27 (m, 2H), 7.21-7.17 (m, 3H), 5.59-5.49 (m, 2H), 3.41 (d, J = 6.0 Hz, 2H), 2.18-2.13 (m, 2H), 1.42-1.28 (m, 10H), 0.89 (t, J = 6.9 Hz, 3H); ^{13}C NMR (100.61 MHz, $CDCl_3$): δ 141.4, 131.2, 128.52, 128.49, 128.1, 125.9, 33.6, 32.0, 29.9, 29.5, 29.4, 27.4, 22.8, 14.3; HRMS: Sample sent for analysis.

Synthesis of *cis*-2-benzyl-3-heptyloxirane (6). A stirred solution of (*Z*)-dec-2-en-1-ylbenzene/(*E*)-dec-2-en-1-ylbenzene 94:6 (616 mg, 2.85 mmol) in DCM (10 mL) under Ar was cooled (ice/water bath), followed by addition of *m*CPBA (766 mg, 3.42 mmol). The reaction mixture was stirred at 0 °C for 2 h and rt for 22 h, before quenching with 1:1 sat. $NaHCO_3$:10% $Na_2S_2O_3$ (20 mL). The phases were separated, and the aq. layer was extracted with DCM (3 x 15 mL). The combined organic phases were washed with 1:1 sat. $NaHCO_3$:10% $Na_2S_2O_3$ (20 mL), sat. aq. $NaHCO_3$ (2 x 20 mL), water (20 mL), brine (20 mL), dried ($MgSO_4$), filtered, and concentrated *in vacuo* to afford a 94:6 mixture of the *cis*:*trans* isomers of **6** as a colourless liquid (456 mg, 92%). R_f = 0.33 in pet. ether/EtOAc 95:5; IR (neat): ν_{max} 3028, 2955, 2923, 2854, 1604; ^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.35-7.30 (m, 2H), 7.28-7.22 (m, 3H), 3.17 (td, J = 6.2 Hz, 4.2 Hz, 1H), 3.00 (ddd, J = 6.7 Hz, 5.5 Hz, 4.2 Hz, 1H), 2.92 (dd, J = 14.7 Hz, 6.4 Hz, 1H), 2.81 (dd, J = 14.7 Hz, 6.2 Hz, 1H), 1.70-1.60 (m, 2H), 1.58-1.23 (m, 10H), 0.89 (t, J = 7.0 Hz, 3H); ^{13}C NMR (100.61 MHz, $CDCl_3$): δ 138.2, 128.9, 128.7, 126.7, 57.6, 57.5, 34.5, 31.9, 29.7, 29.4, 28.2, 26.8, 22.8, 14.2; HRMS: Calcd for $C_{16}H_{24}ONa$ $[M+Na]^+$ 255.17194, found 255.17201.

Synthesis of 3-azido-1-phenyldecan-2-ol (7a). To a stirred solution of *cis*-2-benzyl-3-heptyloxirane (**6**) (654 mg, 2.81 mmol) in MeOH (6.3 mL) and water (0.7 mL), was added NaN_3 (548 mg, 8.43 mmol) and NH_4Cl (301 mg, 5.62 mmol) at rt. The reaction mixture was stirred at 50 °C for 48 hr. MeOH and water was removed under reduced pressure and the residue was purified by silica-gel flash chromatography (pet. ether/EtOAc 95:5 → 90:10) to yield a 7:3 mixture of regioisomers of the title compound as a colourless oily liquid (624 mg, 81%). R_f = 0.34 in pet. ether/EtOAc 9:1; IR (neat): ν_{max} 3438, 2039, 2925, 2856, 2101, 1604, 1455; ^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.35-7.31 (m, in a ratio 7:3, 2H, **7a**/**7b**), 7.28-7.22 (m, in a ratio 7:3, 3H, **7a**/**7b**), 3.84-3.79 and 3.54-3.50 (2 x m, in a ratio of 7:3, 1H, **7a**/**7b**), 3.49-3.45 (m, 1H, b), 3.23-3.19 (m, 1H, a), 3.03 (dd, J = 13.8 Hz, 6.1 Hz, 1H, b), 2.94 (dd, J = 13.8 Hz, 8.3 Hz, 1H, b), 2.90-2.81 (m, 2H, a), 1.77-1.61 (m, 2H, a/b), 1.60-1.23 (m, in a ratio 7:3, 10H, **7a**/**7b**), 0.90-0.86 (m, in a ratio 7:3, 3H, **7a**/**7b**); ^{13}C NMR (100.61 MHz, $CDCl_3$): δ 137.75 (**7a**), 137.67 (**7b**), 129.49 (**7a**), 129.47 (**7b**), 128.9 (**7a**), 128.8 (**7b**), 127.0 (**7b**), 126.9 (**7a**), 74.6 (**7a**), 72.7 (**7b**), 68.1 (**7b**), 65.6 (**7a**), 41.0 (**7a**), 37.5 (**7b**), 34.7 (**7b**), 31.90 (**7b**), 31.88 (**7a**), 31.0 (**7a**), 29.6 (**7b**), 29.5 (**7a**), 29.31 (**7b**), 29.26 (**7a**), 26.4 (**7a**), 25.8 (**7b**), 22.8 (**7a**/**7b**), 14.2 (**7a**/**7b**); HRMS: Calcd for $C_{16}H_{25}N_3ONa$ $[M+Na]^+$ 298.18898, found 298.18898.

Synthesis of 3-amino-1-phenyldecan-2-ol (8a). A mixture of regioisomers **7a**:**7b** (7:3) (620 mg, 2.25 mmol) dissolved in EtOAc (7 mL) was added 10% Pd/C (38 mg, 10 mol%). The reaction mixture was purged with hydrogen gas (1 atm, balloon) for 10 min before the flask was sealed and left stirring under a hydrogen atmosphere for 24 h. Pd/C was removed by filtering through a 0.45 µm PP syringe filter and concentration of the filtrate yielded a 7:3 mixture of regioisomers **8a** and **8b** (543 mg, 97%). R_f = 0.09 in DCM/MeOH 99:1; IR (neat): ν_{max} 3287, 3112, 3029, 2919 2852, 1602; ^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.35-7.29 (m, in a ratio 7:3, 2H, **8a**/**8b**), 7.25-7.18 (m, in a ratio 7:3, 3H, **8a**/**8b**), 3.58-3.53 (m, 0.7H, **8a**), 3.37-3.33 (m, 0.3H, **8b**), 2.95-2.86 (m, 0.6H, **8b**), 2.84 (dd, J = 13.7 Hz, 4.2 Hz, 0.7H, **8a**), 2.72 (dd, J = 13.7 Hz, 8.1 Hz, 0.7H, **8a**), 2.64-2.60 (m, 0.7H, **8a**), 2.48 (dd, J = 13.0 Hz, 9.1 Hz, 0.3H, **8b**), 1.60-1.46 (m, in a ratio 7:3, 2H, **8a**/**8b**), 1.43-1.27 (m, in a ratio 7:3, 10H, **8a**/**8b**), 0.88 (t, J = 6.8 Hz, in a ratio 7:3, 3H, **8a**/**8b**); ^{13}C NMR (100.61 MHz, $CDCl_3$): δ 139.3 (**8b**), 139.0 (**8a**), 129.5 (**8a**), 129.4 (**8b**), 128.7 (**8b**), 128.6 (**8a**), 126.5 (**8b**), 126.4 (**8a**), 74.8 (**8a**), 73.6 (**8b**), 56.8 (**8b**), 54.7 (**8a**), 41.3 (**8b**), 41.1 (**8a**), 34.8(3) (**8b**), 34.8(1) (**8a**), 31.9(9) (**8b**), 31.9(6) (**8a**), 29.9 (**8b**), 29.8 (**8a**), 29.4(4) (**8b**), 29.4(0) (**8a**), 26.4 (**8a**), 26.0 (**8b**), 22.8(1) (**8b**), 22.79 (**8a**), 14.25 (**8b**), 14.24 (**8a**); HRMS: Calcd for $C_{16}H_{28}NO$ $[M+H]^+$ 250.21654, found 250.21677.

Synthesis of 3-((4-nitrophenyl)amino)-1-phenyldecan-2-ol (9a). A solution of amine **8** (249 mg, 1.00 mmol), 1-fluoro-4-nitrobenzene (155 mg, 1.10 mmol), DIPEA (0.52 mL, 3.00 mmol) in DMF (2 mL) was stirred at 80 °C under Ar for 24 h. The product was isolated from a 7:3 mixture of regioisomers **9a** and **9b** by two consecutive purifications by silica-gel column chromatography (pet. ether/DCM 1:1 → 0:1 and pet. ether/DCM 3:7) and concentration of the relevant fractions (R_f = 0.11 in pet. ether/DCM 3:7) yielded **9a** as a yellow waxy solid (44 mg, 12%). The undesired amino alcohol **9b** was not isolated. IR (neat): ν_{max} 3500, 3398, 3061, 2925, 2855, 1597; ^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.08 (d, J = 9.2 Hz, 2H), 7.34-7.30 (m, 2H), 7.28-7.24 (m, 1H), 7.16-7.14 (m, 2H), 6.51 (d, J = 9.2 Hz, 2H), 4.91 (d, J = 9.6 Hz, NH), 4.02 (t, J = 6.6 Hz, 1H), 3.49-3.43 (m, 1H), 2.82 (d, J = 6.9 Hz, 2H), 1.78 (bs, OH), 1.72-1.57 (m, 2H), 1.35-1.24 (m, 10H), 0.86 (t, J = 6.9 Hz, 3H); ^{13}C NMR (100.61 MHz, $CDCl_3$): δ 153.7, 137.8, 137.5, 129.5, 129.0, 127.1, 126.8, 111.3, 73.5, 55.6, 41.2, 32.8, 31.9, 29.7, 29.3, 26.4, 22.7, 14.2; HRMS: Calcd for $C_{22}H_{30}N_2O_3Na$ $[M+Na]^+$ 393.21486, found 393.21499.

Photolysis of 3-((4-nitrophenyl)amino)-1-phenyldecan-2-ol (9a). A solution of compound **9a** (23.6 mg, 0.064 mmol) was photolysed in a 150 mL (0.42 mM) photochemical reactor according to the general procedure with a

125 W medium-pressure mercury-vapour lamp for 4 h at pH 11. The aqueous phase was extracted with DCM (3 x 50 mL). pH of the aqueous phase was adjusted to ≈2 with HCl (1 M) and extracted with DCM (3 x 50 mL). The combined organic fractions were dried ($MgSO_4$), filtered, and concentrated. The resulting residue was used for further analysis.

Synthesis of *N*-(3,5-dichloro-2-fluorophenyl)acetamide. A solution of 3,5-dichloro-2-fluoroaniline (216 mg, 1.20 mmol) in anhydr. DCM (4 mL) was cooled (ice/water bath), followed by dropwise addition of acetyl chloride (140 µL, 1.92 mmol) and Et_3N (270 µL, 1.92 mmol) over a period of 5 min. The reaction mixture was stirred at ambient temperature for 30 min, then at rt. for another 30 min, before quenching with water (10 mL) and sat. aq. $NaHCO_3$ solution (10 mL). The phases were separated, the aq. layer was extracted with DCM (3 x 10 mL), the combined organic layers were concentrated, and the product was isolated by silica-gel column chromatography (pet. ether/EtOAc 8:2). Concentration of the relevant fractions (R_f = 0.24 in pet. ether/EtOAc 8:2) furnished the title compound as a white solid (242 mg, 91%, mp. 168-169 °C). IR (neat): ν_{max} 3293, 3252, 3181, 3116, 3083, 3046, 2990, 2923, 1678, 1606; ^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.35 (dd, *J* = 5.9 Hz, 2.0 Hz, 1H), 7.33 (bs, NH), 7.11 (dd, *J* = 6.2 Hz, 2.6 Hz, 1H), 2.24 (s, 3H); ^{13}C NMR (100.61 MHz, $CDCl_3$): δ 168.3, 145.7, 130.1 (d, *J* = 4.6 Hz), 128.4 (d, *J* = 11.0 Hz), 124.5, 121.3 (d, *J* = 17.7 Hz), 119.9, 24.9; ^{19}F NMR (376.46 MHz, $CDCl_3$): δ -135.2; HRMS: Calcd for $C_8H_5NOCl_2F$ $[M-H]^-$ 219.97377, found 219.97361.

Synthesis of *N*-(3,5-dichloro-2-fluoronitrophenyl)acetamide. A stirred solution of *N*-(3,5-dichloro-2-fluorophenyl)acetamide (520 mg, 2.34 mmol)in conc. sulfuric acid (6.5 mL) was cooled to -10 °C (ice/salt bath), followed by dropwise addition of an ice cold mixture of conc. sulfuric acid (6.5 mL) and 65% nitric acid (8.4 mL) over a period of 15 min. The reaction mixture was stirred at ambient temperature for 1 h and then poured in to a beaker with ice. DCM (20 mL) was added, the phases were separated, and the aq. layer was extracted with DCM (3 x 15 mL). The combined organic layers were washed with water (20 mL), dried ($MgSO_4$), filtered, and concentrated under reduced pressure to yield a 62:38 mixture of *p*- and *o*-nitrated products as an off-white solid (500 mg, 80%). The next step was performed without further purification. R_f = 0.47 (*p*-NO_2) and 0.51 (*o*-NO_2) in pet. ether/EtOAc 6:4; IR (neat): ν_{max} 3263, 3194, 3112, 3073, 1706, 1682; ^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.63 (d, *J* = 6.7 Hz, 1H, *p*-NO_2), 7.52 (d, *J* = 6.3 Hz, 1H, *o*-NO_2), 7.60 (bs, NH, *p*-NO_2), 7.42 (bs, NH, *o*-NO_2), 2.28 (s, 3H, *p*-NO_2), 2.21 (s, 3H, *o*-NO_2); ^{13}C NMR (100.61 MHz, $CDCl_3$): δ 168.9 (*p*-NO_2), 168.7 (*o*-NO_2), 152.4 (d, *J* = 256.7 Hz, *o*-NO_2), 146.4 (d, *J* = 248.3 Hz, *p*-NO_2), 144.5 (*o*-NO_2), 143.5 (*p*-NO_2), 129.8 (*o*-NO_2), 129.5 (d, *J* = 10.8 Hz, *p*-NO_2), 125.9 (d, *J* = 18.5 Hz, *o*-NO_2), 122.4 (d, *J* = 4.6 Hz, *p*-NO_2), 122.1 (d, *J* = 5.1 Hz, *o*-NO_2), 121.1 (d, *J* = 17.8 Hz, *o*-NO_2), 120.2 (*p*-NO_2), 115.1 (d, *J* = 21.8 Hz, *p*-NO_2), 24.9 (*p*-NO_2), 23.2 (*o*-NO_2); ^{19}F NMR (376.46 MHz, $CDCl_3$): δ -129.7 (*p*-NO_2), -114.7 (*o*-NO_2); HRMS: Calcd for $C_8H_4N_2O_3Cl_2F$ $[M-H]^-$ 264.95885, found 264.95792.

Synthesis of 3,5-dichloro-2-fluoro-4-nitroaniline (10). A mixture of *o*- and *p*-nitro isomers of acetamide (500 mg, 1.87 mmol) was dissolved in MeOH (25 mL) and 37% hydrochloric acid (3 mL) followed by stirring at 60 °C for 6 hr. The volatiles were removed under reduced pressure and the product was isolated by silica-gel autoflash chromatography (pet. ether/EtOAc 9:1 → 7:3). Concentration of the relevant fractions (R_f = 0.17 in pet. ether/EtOAc 8:2) yielded the title compound **10** (284 mg, 67%, mp. 142-144 °C) as a yellow crystalline solid. IR (neat): ν_{max} 3498, 3394, 3205, 3055, 1623; ^{1}H NMR (400.13 MHz, $CDCl_3$): δ 6.75 (d, *J* = 7.5 Hz, 1H), 4.27 (bs, NH_2); ^{13}C NMR (100.61 MHz, $CDCl_3$): δ 145.1 (d, *J* = 244.9 Hz), 139.2, 137.8 (d, *J* = 13.4 Hz), 122.6 (d, *J* = 4.2 Hz), 116.0 (d, *J* = 20.5 Hz), 114.1 (d, *J* = 3.9 Hz); ^{19}F NMR (376.46 MHz, $CDCl_3$): δ -135.1; HRMS: Calcd for $C_8H_{12}Cl_2FN_2O_4$ $[M+2CH_3OH+H]^+$ 289.01527, found 289.01543.

Synthesis of 2-(2,6-difluorobenzyl)oxirane (11). A 35 mL reactor tube charged with 2-bromo-1,3-difluorobenzene (965 mg, 5.00 mmol), allylboronic acid pinacol ester (1008 mg, 6.00 mmol), $Pd(PPh_3)_4$ (289 mg, 5 mol%), CsF (2.70 g, 17.5 mmol), and anhydr. THF (15 mL) was purged with argon gas and irradiated at 120 °C for 1 h in a microwave reactor. The resulting slurry was filtered with the aid of DCM (100 mL) and the filtrate was evaporated onto celite. 2-Allyl-1,3-difluorobenzene was isolated by silica-gel flash chromatography (pet. ether) and the relevant fractions (R_f 0.65, pet. ether) were concentrated until 5 mL pet. ether remained. Dry DCM (15 mL) was added, and the solution was cooled to 0 °C (ice/water) followed by addition of *m*CPBA (1.35 g, 6.00 mmol). The reaction mixture was stirred at ambient temperature for 2 h, then rt. for 26 h before quenching with 1:1 sat. aq. $NaHCO_3$:10% $Na_2S_2O_3$ solution (30 mL). The phases were separated, and the aq. layer was extracted with DCM (3 x 15 mL). The combined organic layers were washed with 1:1 sat. aq. 1:1 sat. $NaHCO_3$:10% $Na_2S_2O_3$ (30 mL), sat. aq. $NaHCO_3$ solution (30 mL), water (30 mL), dried ($MgSO_4$), filtered, and concentrated to yield **11** as a colorless liquid (340 mg, 40% over two steps). R_f = 0.43 in pet. ether/DCM 5:5; IR (neat): ν_{max} 3056, 2998, 2928, 1626, 1589, 1468; ^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.24-7.16 (m, 1H), 6.90-6.86 (m, 2H), 3.19-3.10 (m, 2H), 2.82 (dd, *J* = 14.0 Hz, 5.8 Hz, 1H), 2.77-2.75 (m, 1H), 2.56 (dd, *J* = 4.9 Hz, 2.4 Hz); ^{13}C NMR (100.61 MHz, $CDCl_3$): δ 161.9 (dd, *J* = 247.4 Hz, 8.5 Hz), 128.6 (t, *J* = 10.2 Hz), 112.6 (t, *J* = 20.5 Hz), 111.3 (dd, *J* = 19.0 Hz, 6.9 Hz), 50.8, 47.1, 25.6 (t, *J* = 2.0 Hz); ^{19}F NMR (376.46 MHz, $CDCl_3$): δ -114.8; HRMS: Sample sent for analysis.

2-(2,6-Difluoro-3-nitrobenzyl)oxirane (12). To a stirred solution of 2-allyl-1,3-difluoro-4-nitrobenzene (194 mg, 0.97 mmol) at 0 °C was added *m*CPBA (425 mg, 1.94 mmol). The reaction mixture was stirred for 2 h at 0 °C and 5 d at rt, followed by addition of a 1:1 sat. $NaHCO_3$:10% $Na_2S_2O_3$ (30 mL). The phases were separated, and the aqueous layer was extracted with DCM (3 x 15 mL). The combined organic phases were washed with a 1:1 sat. $NaHCO_3$:10% $Na_2S_2O_3$ (30 mL), sat. aq. $NaHCO_3$ (30 mL), water (30 mL), dried ($MgSO_4$), filtered and concentrated under reduced pressure to yield 2-(2,6-difluoro-3-nitrobenzyl)oxirane (**12**) (R_f = 0.53 in DCM) as a slightly yellow oily liquid (183 mg, 88%). IR (neat): ν_{max} 3104, 3000, 2926, 1728, 1624; ^{1}H NMR (400.13 MHz, $CDCl_3$): δ 8.05 (ddd, *J* = 9.2 Hz, 8.5 Hz, 5.7 Hz, 1H), 7.07-7.02 (m, 1H), 3.22-3.17 (m, 1H), 3.16-3.11 (m, 1H), 3.03-2.97 (m, 1H), 2.81-2.79 (m, 1H), 2.56 (dd, *J* = 4.8 Hz, 2.5 Hz, 1H); ^{13}C NMR (100.61 MHz, $CDCl_3$): δ 164.5 (dd, *J* = 259 Hz, 8 Hz), 155.5 (dd, *J* = 266 Hz, 9 Hz), 134.5, 126.1

(dd, *J* = 11 Hz, 1 Hz), 115.8 (dd, *J* = 22 Hz, 20 Hz), 111.8 (dd, *J* = 25 Hz, 4 Hz), 50.1, 46.9, 25.8 (t, *J* = 2 Hz); ^{19}F NMR (376.46 MHz, $CDCl_3$): δ -101.5 (d, *J* = 13.6 Hz), -115.7 (d, *J* = 13.6 Hz); HRMS (EI/TOF): Calcd for $C_7H_4F_2NO_2$ $[M-C_2H_2O]^+$ 172.02046, found 172.02080.

Synthesis of 1-allyl-4-chlorobenzene. A mixture of 4-chloro-1-iodobenzene (954 mg, 4.00 mmol), $Pd(PPh_3)_4$ (46 mg, 1 mol%), CsF (2127 mg, 14.0 mmol), allyl boronate pinacol ester (1.51 mL, 8.00 mmol) in anhydr. THF (50 mL) was refluxed (oil bath, 70 °C) under Ar for 16 h. The reaction mixture was cooled to rt and pet. ether (20 mL) and water (20 mL) were added. The phases were separated, and the aq layer was extracted with pet. ether (3 x 15 mL). The combined organic phases were concentrated under reduced pressure onto celite, and the product was isolated by silica-gel column chromatography (pet. ether). Concentration of the relevant fractions (R_f = 0.51 in pet. ether) furnished the title compound as a colorless liquid (471 mg, 77%). Spectroscopic data are in accordance with previously reported data in the literature.[42]

Synthesis of 2-(4-chlorobenzyl)oxirane (13). A stirred solution of 1-allyl-4-chlorobenzene (450 mg, 2.95 mmol) in anhydrous DCM (8 mL) under Ar was cooled (ice/water bath), followed by addition of *m*CPBA (804 mg, 3.59 mmol). The reaction mixture was stirred at ambient temperature for 2 h and rt for 22 h, before being quenched with a 1:1 sat. $NaHCO_3$:10% $Na_2S_2O_3$ (20 mL). The phases were separated, and the aq. layer was extracted with DCM (3 x 15 mL). The combined organic phases were washed 1:1 sat. $NaHCO_3$:10% $Na_2S_2O_3$ (20 mL), sat. aq. $NaHCO_3$ (2 x 20 mL), water (20 mL), and brine (20 mL) and then dried ($MgSO_4$), filtered, and concentrated *in vacuo* to yield **11** as a colourless liquid (456 mg, 92%). Spectroscopic data are in accordance with previously reported data in the literature.[47]

Lewis acid-promoted epoxide ring opening; general procedure for the preparation of 14e-14k. Lithium perchlorate was dried under vacuum for 1 h and dissolved in dry diethyl ether to a 5 M solution. Aniline (~ 0.2 M) and epoxide (1.0 equiv.) were added, and the reaction mixture was stirred at 40 °C under Ar. DCM and water were added, the phases were separated, and the aqueous layer was extracted with DCM (3 x 10 mL). The combined organic phases were evaporated on celite and subjected to silica-gel flash chromatography (pet. ether/DCM 3:7). Concentration of the relevant fractions gave the corresponding propan-2-ol derivative essentially pure according to ^{1}H-NMR analysis.

Synthesis of 1-(3,5-dichloro-2-fluoro-4-nitrophenyl)-amino-3-(2,6-difluorophenyl)propan-2-ol (14e). A dry round-bottom flask was charged with lithium perchlorate (2.52 g, 23.7 mmol) and diethyl ether (5 mL). The solution was stirred for 30 min followed by addition of 3,5-dichloro-2-fluoro-4-nitroaniline (260 mg, 1.16 mmol) and 2-(2,6-difluorobenzyl)oxirane (198 mg, 1.16 mmol). The reaction mixture was stirred at reflux (oil bath, 60 °C) for 3 d before DCM (10 mL) was added followed by dropwise addition of water (10 mL). The phases were separated, and the organic layer was extracted with DCM (3 x 10 mL). Isolation by silica-gel flash chromatography (pet. ether/DCM 1:1) and concentration of the relevant fractions (R_f = 0.16 in pet. ether/DCM 3:7) gave **14e** (91 mg, 20%, mp. 158-159 °C) and recovered 3,5-dichloro-2-fluoro-4-nitroaniline (89 mg, 34%). IR (neat): ν_{max} 3402, 3369, 2968, 2928, 1604; ^{1}H NMR (400.13 MHz, CD_3CN): δ 7.32-7.24 (m, 1H), 7.00-6.93 (m, 1H), 6.83 (d, *J* = 7.5 Hz, 1H), 5.45 (bs, NH), 4.05-3.98 (m, 1H), 3.37-3.31 (m, 1H), 3.31 (d, *J* = 5.3 Hz, OH), 2.87-2.85 (m, 2H); ^{13}C NMR (100.61 MHz, CD_3CN): δ 162.8 (dd, *J* = 245 Hz, 9 Hz), 116.0 (d, *J* = 244 Hz), 141.3 (d, *J* = 12 Hz), 137.3, 129.5 (t, *J* = 10 Hz), 123.4 (d, *J* = 4 Hz), 115.1 (d, *J* = 20 Hz), 115.1 (t, *J* = 20 Hz), 112.1 (d, *J* = 26 Hz), 111.0 (*J* = 4 Hz), 69.8, 49.1, 28.7; ^{19}F NMR (376.46 MHz, CD_3CN): δ -115.7, -136.8; HRMS: Calcd for $C_{15}H_{10}N_2O_3Cl_2F_3$ $[M-H]^-$ 393.00261, found 393.00245.

Synthesis of 3-(4-chlorophenyl)-1-((3,5-dichloro-2-fluoro-4-nitrophenyl)amino)propan-2-ol (14f). 3,5-Dichloro-2-fluoro-4-nitroaniline (77 mg, 0.34 mmol) and 2-(4-chlorobenzyl)oxirane (57 mg, 0.34 mmol) were reacted following the general procedure for 20 h. The target compound was isolated by two consecutive purifications by silica-gel flash chromatography (pet. ether/DCM 2:8 and pet. ether/EtOAc 7:3). The relevant fractions were concentrated and pet. ether (2 mL) and EtOAc (3 drops) was added to the residue. The mixture was heated to 50 °C and the liquid was decanted off, leaving a yellow solid containing 80% pure product. This mixture was washed with a solution of pet. ether (10 mL) and EtOAc (15 drops), followed by a final rinse with EtOAc (10 mL) to yield aminol **14f** as a yellow crystalline solid (20 mg, 15%, mp. 148-149 °C). R_f = 0.46 in pet. ether/EtOAc 6:4; IR (neat): ν_{max} 3380, 3091, 2918, 2859, 1603; ^{1}H NMR (400.13 MHz, CD_3CN): δ 7.31 (d, *J* = 8.5 Hz, 2H, ArH), 7.25 (d, *J* = 8.5 Hz, 2H, ArH), 6.79 (d, *J* = 7.5 Hz, 1H, ArH), 5,44 (bs, NH), 3.99-3.91 (m, 1H), 3.29 (ddd, *J* = 13.7 Hz, 6.3 Hz, 3.9 Hz, 1H), 3.19 (d, *J* = 5.1 Hz, OH), 3.17-3.11 (m, 1H), 2.82 (dd, *J* = 13.8 Hz, 4.9 Hz, 1H), 2.70 (dd, *J* = 13.8 Hz, 8.0 Hz, 1H); ^{13}C NMR (100.61 MHz, CD_3CN): δ 146.0 (d, *J* = 244 Hz), 141.3 (d, *J* = 12 Hz), 138.7, 137.3, 132.5, 132.2, 129.2, 123.5 (d, *J* = 4 Hz), 115.2 (d, *J* = 21 Hz), 111.0 (d, *J* = 4 Hz), 71.2, 49.1, 41.1; ^{19}F NMR (376.46 MHz, CD_3CN): δ -136.7; HRMS: Calcd for $C_{15}H_{11}Cl_3FN_2O_3$ $[M-H]^-$ 390.98139, found 390.98148.

Synthesis of 1-(2,6-difluorophenyl)amino-3-(4-nitrophenyl)propan-2-ol (14g). 2,6-Difluoroaniline (43 mg, 0.33 mmol) and 2-(4-nitrobenzyl)oxirane (59 mg, 0.33 mmol) were reacted according to the general procedure for 6 hr to yield aminol **14g** as a white solid (31 mg, 30%, mp. 86-87 °C) along with 43% recovery of epoxide. R_f = 0.15 in DCM; IR (neat): ν_{max} 3308, 3080, 2946, 2886, 2855, 1600; ^{1}H NMR (400.13 MHz, CD_3CN): δ 8.13 (d, *J* = 8.7 Hz, 2H), 7.43 (d, *J* = 8.7 Hz, 2H), 6.91-6.81 (m, 2H), 6.73-6.65 (m, 1H), 4.27 (bs, NH), 3.98-3.90 (m, 1H), 3.46-3.40 (m, 1H), 3.20-3.14 (m, 1H, overlapping with OH), 3.17 (d, *J* = 5.3 Hz, OH), 2.95 (dd, *J* = 13.7 Hz, 4.4 Hz, 1H), 2.80 (dd, *J* = 13.7 Hz, 8.4 Hz, 1H); ^{13}C NMR (100.61 MHz, CD_3CN): δ 154.3 (dd, *J* = 240 Hz, 8 Hz), 148.4, 147.6, 131.5, 127.0 (t, *J* = 14 Hz), 124.2, 118.5 (t, *J* = 10 Hz), 112.5 (dd, *J* = 16 Hz, 7 Hz), 71.9, 52.4 (t, *J* = 4 Hz), 41.7; ^{19}F NMR (376.46 MHz, CD_3CN): δ -130.0; HRMS: Calcd for $C_{15}H_{15}F_2N_2O_3$ $[M+H]^+$ 309.10453, found 309.10467.

Synthesis of 1-(3,5-dichloro-2-fluorophenyl)amino-3-(4-nitrophenyl)propan-2-ol (14h). 3,5-Dichloro-2-fluoro-aniline (59 mg, 0.33 mmol) and 2-(4-nitrobenzyl)oxirane (59 mg, 0.33 mmol) were reacted according to the general procedure for 20 h to yield aminol **14h** as a white solid (57 mg, 48%, mp. 113-114 °C) along with 32% recovery of epoxide. R_f = 0.31 in DCM; IR (neat): ν_{max} 3447,

3351, 3263, 3117, 3085, 2950, 2919, 2904, 2850, 1604; ^{1}H NMR (400.13 MHz, CD_3CN): δ 8.13 (d, *J* = 8.8 Hz, 2H), 7.47 (d, *J* = 8.8 Hz, 2H), 6.68-6.64 (m, 2H), 4.92 (bs, NH), 4.03-3.96 (m, 1H), 3.28-3.22 (m, 1H, overlapping with OH), 3.24 (d, *J* = 5.1 Hz, OH), 3.12-3.05 (m, 1H), 2.97 (dd, *J* = 13.7 Hz, 4.4 Hz, 1H), 2.82 (dd, *J* = 13.7 Hz, 8.4 Hz, 1H); ^{13}C NMR (100.61 MHz, CD_3CN): δ 148.3, 147.6, 146.8 (d, *J* = 239 Hz), 139.9 (d, *J* = 12 Hz), 131.5, 130.5 (d, *J* = 4 Hz), 124.2, 121.3 (d, *J* = 16 Hz), 116.4 (d, *J* = 2 Hz), 111.5 (d, *J* = 3 Hz), 70.9, 49.5, 41.6; ^{19}F NMR (376.46 MHz, CD_3CN): δ -141.6; HRMS: Calcd for $C_{15}H_{14}Cl_2FN_2O_3$ $[M+H]^+$ 359.03600, found 359.03598.

Synthesis of 1-(3,5-dichloro-2,4-difluorophenyl)-amino)-3-(4-nitrophenyl)propan-2-ol (14i). 3,5-Dichloro-2,4-difluoroaniline (65 mg, 0.33 mmol) and 2-(4-nitrobenzyl)oxirane (59 mg, 0.33 mmol) was reacted according to the general procedure for 18 h to yield aminol **14i** as a white solid (59 mg, 48%, mp. 124-125 °C) along with 24% recovered epoxide. R_f = 0.29 in DCM; IR (neat): ν_{max} 3386, 3293, 3116, 3080, 2927, 2850, 1601; ^{1}H NMR (400.13 MHz, CD_3CN): δ 8.14 (d, *J* = 8.8 Hz, 2H), 7.48 (d, *J* = 8.8 Hz, 2H), 6.76 (dd, *J* = 8.5 Hz, 7.2 Hz, 1H), 4.72 (bs, NH), 4.04-3.96 (m, 1H), 3.26-3.21 (m, 1H, overlapping with OH), 3.21 (d, *J* = 5.0 Hz, OH), 3.10-3.04 (m, 1H), 2.97 (dd, *J* = 13.7 Hz, 4.4 Hz, 1H), 2.82 (dd, *J* = 13.7 Hz, 8.4 Hz, 1H); ^{13}C NMR (100.61 MHz, CD_3CN): δ 148.3, 147.7, 146.9 (dd, *J* = 242 Hz, 2 Hz), 146.3 (dd, *J* = 237 Hz, 2 Hz), 135.7 (dd, *J* = 12 Hz, 3 Hz), 131.5, 124.2, 117.3 (dd, *J* = 18 Hz, 4 Hz), 111.2 (dd, *J* = 22 Hz, 20 Hz), 111.1 (d, *J* = 4 Hz), 70.9, 49.8, 41.6; ^{19}F NMR (376.46 MHz, CD_3CN): δ -134.2 (d, *J* = 4.4 Hz), -136.9 (d, *J* = 4.4 Hz); HRMS: Calcd for $C_{15}H_{13}Cl_2F_2N_2O_3$ $[M+H]^+$ 377.02658, found 377.02648.

Synthesis of 3-(2,6-difluoro-3-nitrophenyl)-1-((2,6-difluorophenyl)amino)propan-2-ol (14j). 2,6-Difluoroaniline (39 mg, 0.30 mmol) and 2-(2,6-difluoro-3-nitrobenzyl)oxirane (65 mg, 0.30 mmol) were reacted according to the general procedure for 19 h to yield aminol **14j** as an off-white solid (26 mg, 25%, mp. 74-76 °C) along with 26% recovery of epoxide. R_f = 0.23 in DCM; IR (neat): ν_{max} 3346, 3256, 3100, 2933, 1621; ^{1}H NMR (400.13 MHz, CD_3CN): δ 8.04 (ddd, *J* = 9.2 Hz, 8.6 Hz, 5.7 Hz, 1H), 7.12 (ddd, *J* = 9.2 Hz, 8.6 Hz, 1.8 Hz, 1H), 6.91-6.81 (m, 2H), 6.72-6.66 (m, 1H), 4.31 (bs, NH), 3.98-3.90 (m, 1H), 3.50-3.44 (m, 1H), 3.31-3.20 (m, 2H, overlapping OH), 2.95-2.84 (m, 2H); ^{13}C NMR (100.61 MHz, CD_3CN): δ 165.5 (dd, *J* = 256 Hz, 8 Hz), 156.2 (dd, *J* = 263 Hz, 10 Hz), 154.3 (dd, *J* = 239 Hz, 8 Hz), 135.4, 126.9 (t, *J* = 14 Hz), 126.6 (dd, *J* = 12 Hz, 1 Hz), 118.7 (dd, *J* = 21 Hz, 19 Hz), 118.5 (t, *J* = 10 Hz), 112.7-112.5 (m), 112.5 (dd, *J* = 16 Hz, 7 Hz), 70.3, 52.3 (t, *J* = 4 Hz), 29.1; ^{19}F NMR (376.46 MHz, CD_3CN): δ -103.2 (d, *J* = 14.0 Hz, 1F), -117.7 (d, *J* = 14.0 Hz, 1F), -130.1 (s, 2F); HRMS: Calcd for $C_{15}H_{13}F_4N_3O_3$ $[M+H]^+$ 345.08568, found 345.08577.

Synthesis of 3-(4-chlorophenyl)-1-(2,6-difluoro-4-nitrophenyl)aminopropan-2-ol (14k). *Method A: Lithium perchlorate-promoted epoxide ring opening:* 2,6-Difluoro-4-nitroaniline (272 mg, 1.56 mmol) and 2-(4-chlorobenzyl)oxirane (220 mg, 1.30 mmol) were reacted according to the general procedure for 24 h, and isolation by silica-gel flash column chromatography (pet. ether/DCM 4:6) yielded the target compound **14k** (31 mg, 7%, R_f 0.17, DCM).

Synthesis of 1-azido-3-(4-chlorophenyl)propan-2-ol. To a stirred solution of 2-(4-chlorobenzyl)oxirane (**13**) (214 mg, 1.27 mmol) in MeOH (2.7 mL) and water (0.3 mL), was added NaN_3 (248 mg, 3.81 mmol) and NH_4Cl (136 mg, 2.54 mmol) at rt. The reaction mixture was stirred at rt for 18 h. MeOH was removed under reduced pressure and water (5 mL) and EtOAc (5 mL) were added. The phases were separated, and the aq. layer was extracted with EtOAc (3 x 10 mL). The combined organic layers were dried ($MgSO_4$), filtered, and concentrated under reduced pressure gave the title compound as a colorless oily liquid (250 mg, 93%), which was essentially pure based on ^{1}H NMR. R_f = 0.59 in pet. ether/EtOAc 1:1; IR (neat): ν_{max} 3415, 2922, 2096, 1491; ^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.30 (d, *J* = 8.5 Hz, 2H), 7.15 (d, *J* = 8.6 Hz, 2H), 4.00-3.94 (m, 1H), 3.39 (dd, *J* = 12.5 Hz, 3.7 Hz, 1H), 3.29 (dd, *J* = 12.5 Hz, 6.8 Hz, 1H), 2.79-2.77 (m, 2H); ^{13}C NMR (100.61 MHz, $CDCl_3$): δ 135.7, 132.9, 130.8, 129.0, 71.6, 56.1, 40.2; HRMS:.

Synthesis of 1-amino-3-(4-chlorophenyl)propan-2-ol hydrochloride. 1-Azido-3-(4-chlorophenyl)propan-2-ol (520 mg, 2.46 mmol) and PPh_3 (708 mg, 2.70 mmol) in a mixture of THF (9 mL) and water (1 mL) were stirred at 50 °C under Ar for 2 h. THF was removed under reduced pressure, and EtOAc (20 mL) and 6 M aq. hydrochloric acid (20 mL) was added. The phases were separated, and the organic layer was extracted with water (2 x 10 mL). The combined aq. phases were then washed with Et_2O (40 mL) and concentrated under reduced pressure. Traces of water was azeotropically removed by adding toluene (5 mL) followed by evaporation under reduced pressure. Three repetitions of this process gave the title compound as sharp white needles (459 mg, 84%, mp. 188-190 °C). R_f = 0.17 as freebase in DCM/EtOH/NH_3 (20%) 89:10:1; IR (neat): ν_{max} 3452, 3209, 2913, 1600; ^{1}H NMR (400.13 MHz, D_2O) δ 7.30 (d, *J* = 8.4 Hz, 2H), 7.21 (d, *J* = 8.4 Hz, 2H), 4.10-4.04 (m, 1H), 3.17 (dd, *J* = 13.1 Hz, 2.5 Hz, 1H), 2.93 (dd, *J* = 13.0 Hz, 10.2 Hz, 1H), 2.84 (dd, *J* = 14.0 Hz, 4.9 Hz, 1H), 2.72 (dd, *J* = 14.0 Hz, 8.4 Hz, 1H); ^{13}C NMR (100.61 MHz, D_2O) δ 135.8, 131.9, 130.9, 128.5, 68.6, 44.1, 39.7; HRMS: Calcd for $C_9H_{13}NOCl$ $[M+H]^+$ 186.06802, found 186.06890.

Synthesis of 3-(4-chlorophenyl)-1-(2,6-difluoro-4-nitrophenyl)aminopropan-2-ol (14k). *Method B: Nucleophilic aromatic substitution:* A solution of 1-amino-3-(4-chlorophenyl)propan-2-ol hydrochloride (222 mg, 1.00 mmol), 3,4,5-trifluoronitrobenzene (128 µL, 194 mg, 1.10 mmol), DIPEA (700 µL, 519 mg, 4.00 mmol) in ACN (6 mL) was stirred at 40 °C under Ar for 14 h. The product was isolated by silica-gel column chromatography (DCM) and concentration of the relevant fractions (R_f = 0.18 in DCM) yielded **14k** as a yellow crystalline solid (273 mg, 80%, mp. 130-132 °C). IR (neat): ν_{max} 3491, 3294, 3095, 3023, 2897, 1610; ^{1}H NMR (400.13 MHz, $CDCl_3$): δ 7.78 (dd, *J* = 8.1 Hz, 2.1 Hz, 2H), 7.31 (d, *J* = 8.4 Hz, 2H), 7.16 (d, *J* 8.4 Hz, 2H), 4.80 (bs, NH), 4.07-4.01 (m, 1H), 3.75 (d, *J* = 13.4 Hz, 1H), 3.43 (dd, *J* = 13.4 Hz, 8.2 Hz, 1H), 2.87 (dd, *J* = 13.7 Hz, 4.5 Hz, 1H), 2.74 (dd, *J* = 13.7 Hz, 8.5 Hz, 1H); ^{13}C NMR (100.61 MHz, $CDCl_3$): δ 150.1 (dd, *J* = 243.6 Hz, 9.0 Hz), 135.9 (t, *J* = 10.7 Hz), 135.5, 133.1, 132.7 (t, *J* = 12.6 Hz), 130.8, 129.1, 109.0 (dd, *J* = 18.2 Hz, 9.6 Hz), 71.8, 50.1 (t, *J* = 4.5 Hz), 40.9; ^{19}F NMR (376.46 MHz, $CDCl_3$): δ -128.8; HRMS: Calcd for $C_{15}H_{12}N_2O_3ClF_2$ $[M-H]^-$ 341.05100, found 341.05100.

General procedure for photodecomposition of 14a-14d. A solution of the appropriate compound (≈0.10 mmol) in acetonitrile was added to a photochemical reactor containing distilled water at the appropriate pH to a concentration of ≈0.7 mM and a total volume of either 75 or 150 mL (7:3, water:ACN), depending on the reactor size. The reaction vessel was either purged with N_2 during the reaction or left open to air. The reaction mixture was photolyzed with either a 6 W low-pressure mercury-vapour lamp (mainly 254 nm irradiation). After completion, the reaction mixture was transferred to a separatory funnel, saturated with NaCl, and extracted with EtOAc (3 x 50 mL). The pH was adjusted to ≈2 and the aqueous layer was extracted again with EtOAc (3 x 50 mL). The combined organic layers were dried ($MgSO_4$), filtered and concentrated in vacuo on a rotary evaporator to yield a residue which was analysed by ^{1}H NMR.

Photolysis of 1-((2,5-Dichloro-4-(1,1,2,3,3,3-hexafluoropropoxy)phenyl)amino)-3-(4-nitrophenyl)propan-2-ol (14a) A solution of compound **14a** (26.6 mg, 0.052 mmol) was photolyzed in a 75 mL (0.70 mM) photochemical reactor according to the general procedure with a 6 W medium-pressure mercury-vapour lamp for 24 h at pH 13 and 8. The resulting reaction mixture was worked up according to the general procedure.

Photolysis of 1-((3,5-Dichloro-2-fluorophenyl)amino)-3-(2,6-difluoro-3-nitrophenyl)propan-2-ol (14b) A solution of compound **14b** (20.4 mg, 0.052 mmol) was photolyzed in a 75 mL (0.69 mM) photochemical reactor according to the general procedure with a 6 W medium-pressure mercury-vapour lamp for 24 h at pH 13 and 8. The aqueous layer was extracted with EtOAc. The resulting reaction mixture was worked up according to the general procedure.

Photolysis of 1-((3,5-Dichloro-2,4-difluorophenyl)amino)-3-(2,6-difluoro-3-nitrophenyl)propan-2-ol (14c) A solution of compound **14c** (21.0 mg, 0.051 mmol) was photolyzed in a 75 mL (0.68 mM) photochemical reactor according to the general procedure with a 6 W medium-pressure mercury-vapour lamp for 24 h at pH 13 and 8. The aqueous layer was extracted with EtOAc. The resulting reaction mixture was worked up according to the general procedure.

Photolysis of 1-((2,5-Dichloro-4-(1,1,2,3,3,3-hexafluoropropoxy)phenyl)amino)-3-(2,6-difluoro-3-nitrophenyl)propan-2-ol (14d) A solution of compound **14d** (26.8 mg, 0.049 mmol) was photolyzed in a 75 mL (0.66 mM) photochemical reactor according to the general procedure with a 6 W medium-pressure mercury-vapour lamp for 24 h at pH 13 and 8. The aqueous layer was extracted with EtOAc. The resulting reaction mixture was worked up according to the general procedure.

ASSOCIATED CONTENT

The Supporting Information is available free of charge at http://pubs.acs.org.
Figure S1-S4 and NMR spectra of all new compounds.

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Notes

The authors declare no competing financial interests.

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