

Synthesis of a Linear Symmetrically Fused Tröger's Base Analogue

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Degree Project in Organic Chemistry, 2025
Department of Chemistry
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Popular science summary

Tröger's base (TB) is a molecule discovered by Julius Tröger in the late 1800s. It is its three-dimensional structure that provides this molecule with very interesting properties and uses. Tröger's base has a rigid V-shape that allows for a perfect building block in structures with a particular geometry. Nowadays, TB is especially useful in various fields of chemistry and biochemistry such as drug delivery, molecular recognition and DNA binding.

This work is aimed at synthesising a new version of Tröger's base combining three of the units, resulting in a linear and symmetric TB analogue. The goal of the project in which this work is part of, is to build a larger circular structure with seven of the TB units which when achieved, it will allow for safe transport of drugs through cell membranes.

To get to the new version of a TB, a new synthetic path was developed and tested starting from a commercially available chemical. In the process, it was tried to separate different complementary molecules (isomers) in order to try and improve other synthetic paths. However, that proved to be a major challenge and required more advanced purification techniques.

Eventually, a simpler three-unit Tröger's base was created. Unfortunately, a pure molecule was not achieved, and the full potential of this new TB was not tested due to time constraints. Nonetheless, the obtention of said molecule has been proved and therefore laid important groundwork for future research.

In conclusion, this work has contributed and lead one step closer to the final objective of the project, which was designing an artificial tunnel for drugs to go through cell membranes.

Abstract

Julius Tröger first synthesised 'C₁₇H₁₈N₂' as an unknown product from the condensation of anilines with formaldehyde. From that point on, the history of Tröger's base (TB) has widely evolved as new applications for its characteristic rigid, chiral framework easy to functionalise. It is in the field of supramolecular chemistry that TB has gathered the most attention these last years. This work is framed inside the more ambitious project of developing a heptakis-TB structure aimed at drug delivery through bilayer membranes.

The synthesis begins from a commercially available aniline, leading to a TB analogue through condensations and functionalisation of the TB analogue. During this synthesis, chiral centers are introduced to obtain enantiomerically pure intermediates. However, the separation of diastereomers **22a** and **22b** ends up being a bottleneck due to the troublesome purification conditions and the low yields resulting from it. In order to surmount this obstacle, two strategies are explored: late-stage separation and the synthesis of a simpler linear tris-TB analogue.

For the late-stage separation, intermediates **24** and **24'** are synthesised, but none of them offered improved purification towards to the original step. However, a new synthetic route has been developed to produce **37**, a linear tris-TB, through condensation of the mono-protected 2,8-diamino-TB **36**. Despite preliminary LC-MS data seems to confirm the presence of **37**, further purification and configurational isomers separation methods must be developed.

Overall, this project showcases the synthetic complexity of TB analogues and the need to optimize methodologies for diastereomer handling. Albeit the heptakis-TB analogue remains a future goal, having successfully formed a simpler tris-TB creates new opportunities for the future of modular and functional TB systems.

Keywords: Organic Chemistry, Supramolecular Chemistry, Tröger's base, Diastereomers, Tris-Tröger's Base

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1 List of abbreviations

BINAP ($\pm$)-2,2'-bis(diphenylphosphino)-1,1'-binaphthalene

CC Column Chromatography

DCM Dichloromethane

DCVC Dry Column Vacuum Chromatography

equiv equivalents

HNMR Proton Nuclear Magnetic Resonance

LC-MS Liquid Chromatography – Mass Spectrometry

mw. microwave

PFA Paraformaldehyde

PG Protective Group

rac racemic

R_f Retardation factor

rt. room temperature

TB Tröger's Base

TBAF Tetra-n-butylammonium Fluoride

TFA Trifluoroacetic Acid

THF Tetrahydrofuran

TLC Thin-Layer Chromatography

TMS Trimethylsilyl

XRD X-Ray Diffraction

2 Introduction

2.1 Historical Background

It was in 1887 when Julius Tröger published the first article on the condensation of aromatic amines with formaldehyde [1]. An unknown product ‘C₁₇H₁₈N₂’ was isolated from the reaction of *p*-toluidine and methylal (CH₂(OCH₃)₂) in acidic solution. Despite Tröger’s efforts, he failed to elucidate the structure of his new product, which lead to the assignment of a mediocre grade in Tröger’s doctoral thesis. Although many scientists managed to isolate Tröger’s base during the following years, the real structure remained a mystery for almost fifty years. Finally, Spielman brought to light the structure of the Tröger’s base as *rac*-2,8-dimethyl-6*H*,12*H*-5,11-methanodibenzo[*b,f*][1,5]diazocine (**1**, Figure 2.1) in 1935 [2], same year Wagner disclosed the mechanism for the condensation reaction in which Tröger’s base is formed [3].

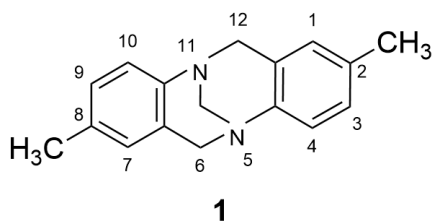


Figure 2.1. Tröger’s base structure and numbering according to its systematic name.

In 1944, Prelog and Wieland managed to separate two TB enantiomers using (+)- α -lactose hydrate on a chiral column chromatography [4]. This achievement allowed in 1991 for the absolute configuration of Tröger’s base to be assigned for each of the possible isomers by X-ray diffraction analysis [5]. Therefore, the absolute configuration for the (+)-TB and (–)-TB results to be (5*S*,11*S*) and (5*R*,11*R*) respectively (Figure 2.2).

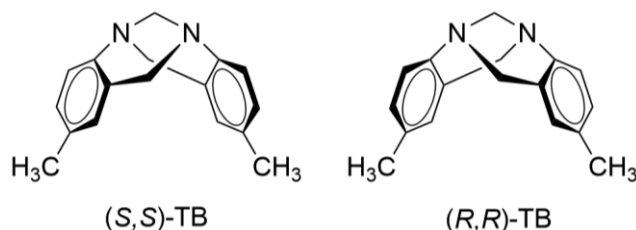


Figure 2.2. Absolute configuration of **TB1** enantiomers.

The 1,5-diazocine bridge connecting the two aromatic rings gives the Tröger’s base its distinctive 90° angle configuration and it only allows the two above mentioned isomers, the (*S,S*) and the (*R,R*). This occurs due to the methylene bridge limiting the pyramidal inversion of the nitrogen in the diazocine bridge.

2.2 Tröger's base condensation and mechanism

The condensation of *p*-toluidine with methylal ($\text{CH}_2(\text{OCH}_3)_2$) in acidic solution was the first ever synthesis of Tröger's base [1]. Despite this being the original method developed by Julius himself, the synthesis of TB analogues is carried out in variations of the original conditions. The most common ones include a methylene synthon, such as formaldehyde or PFA in combination with acidic conditions (Figure 2.3) [6].

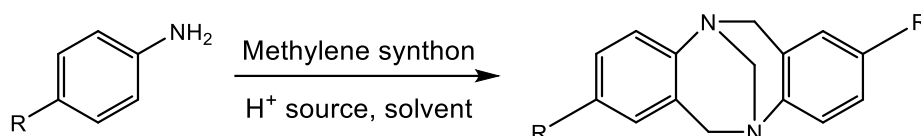


Figure 2.3. Conditions for the TB condensation.

In order to obtain a TB unit and prevent polymerization from happening, the aniline must be *para*-substituted. As well, electron withdrawing groups should be avoided to avoid slow low yielding reactions [7]. However, this obstacle has been bypassed by new conditions of TB condensation.

In 2001, our group developed a protocol which allows the synthesis of halogenated TB analogues [8]. In this conditions, PFA and TFA eliminate the need for electron donor substituted anilines, which allow later functionalisation [9].

As for the mechanism in which **1** is formed, it was Wagner who first proposed a sequence of reactions between formaldehyde and anilines acid-catalysed [3]. However, he later re-examined his work and proposed actualised versions [10, 11] and Farrar made great contributions to it [12]. Therefore, the proposed mechanism for the formation of the 1,5-diazocine bridge is shown in Figure 2.4.

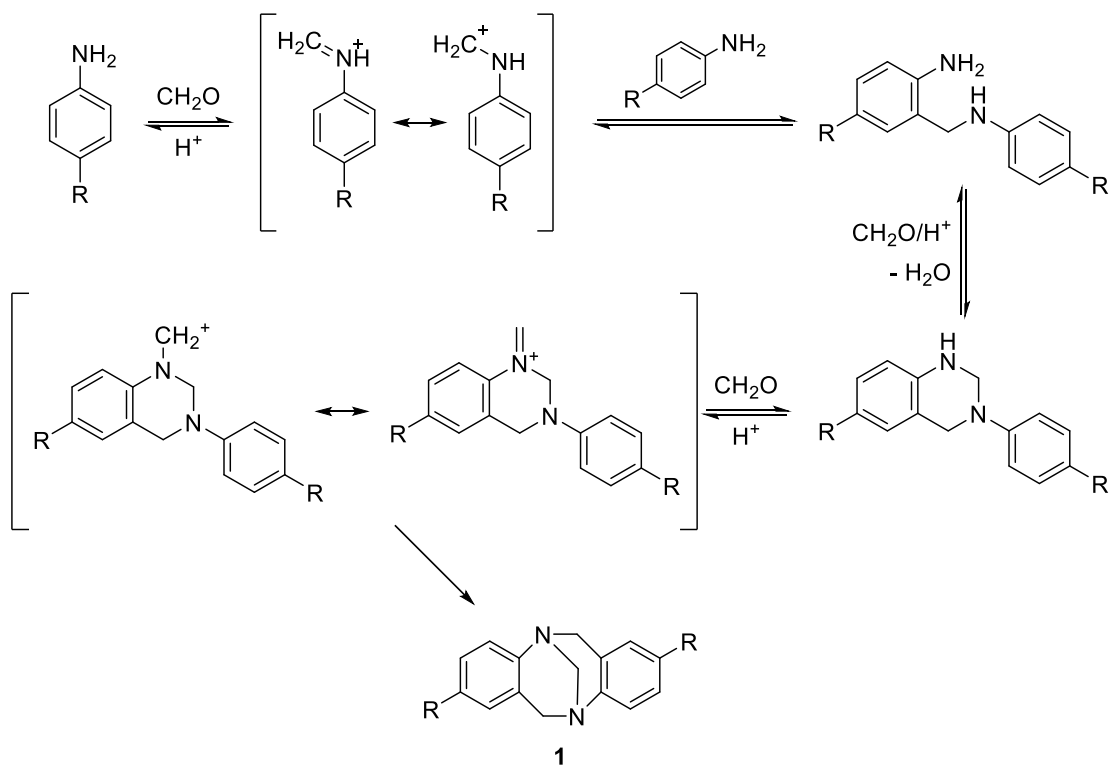


Figure 2.4. The proposed mechanism for the formation of **1** and TB analogues.

2.3 Applications of Tröger's base

After Prelog and Wieland's achievement [4], Tröger's base was only used as a validation substance for newly developed chiral chromatography techniques until the 1980s. Then in 1985 Wilcox made an innovative discover, he managed to synthesise several chiral hosts using TB analogues [6]. Since then and specially in the last two decades, TB analogues and their chemistry have been used throughout a wide range of fields [13-16], as well as being an extensively covered topic in review articles [17, 18].

2.3.1 Tröger's base supramolecular chemistry

After Wilcox's groundbreaking publication on TB analogues, he continued his studies jointly with Adrian. Together they published the synthesis of TB derivatives with carboxylic acid capable of bonding with cyclic urea derivatives or adenine moieties through four hydrogen bonds (**2**; Figure 2.5) [19]. The same TB derivative was later used to resolve the effect water has as a competitive inhibitor on the formation hydrogen-bonding complexes [20].

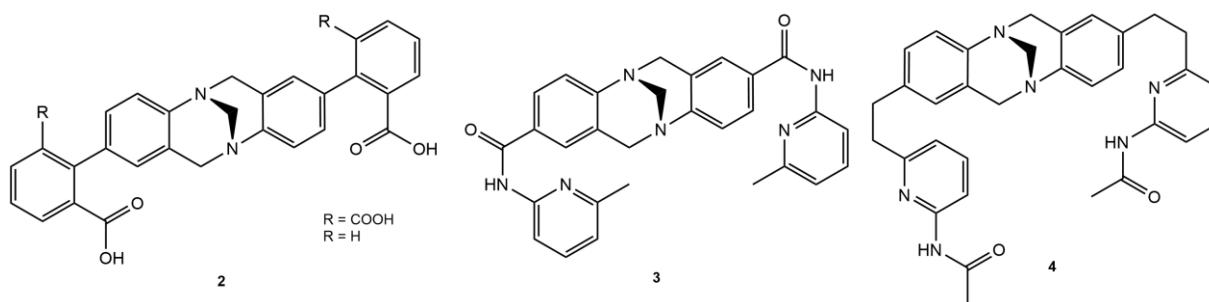


Figure 2.5. TB analogues as hydrogen receptors, diacid (**2**) and aminopyridine (**3,4**) derivatives.

On the same research topic, Goswami and Gosh developed and synthesised two fluorescent aminopyridine TB analogues (**3,4**; Figure 2.5) [21, 22]. The artificial receptors displayed chain-length selectivity for dicarboxylic acids, with compound **3** showing the highest selectivity for suberic acid due to optimal spatial alignment. With the second study [22], multiple aminopyridine analogues of different length were synthesized, of which compound **4** was the longest one, in order to evaluate the relation between the binding properties and the rigidity of the receptor. In the end, it was proved that binding strength and selectivity are enhanced by more rigid and preorganised cavity. The above-mentioned studies were carried out by NMR and fluorescence analyses.

One of our group's multiple TB projects, a bis(18-crown-6) ether TB analogue (**5**; Figure 2.6) was synthesized and used as an artificial receptor for achiral and chiral primary bisammonium salts [23].

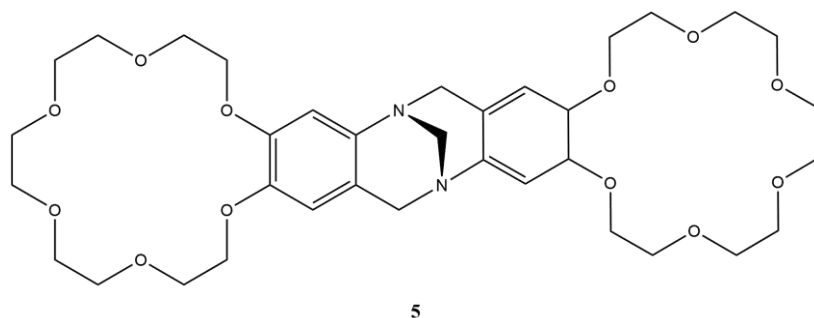


Figure 2.6. TB bis(18-crown-6) ether receptor for bisammonium salts.

The TB analogue was synthesised from a commercially available 18-crown-6 ether aniline in one step. The V-shaped receptor displayed similar selectivity for primary bisammonium salts of 6 to 8 methylene groups, with heptane-1,7-diylbis(ammonium chloride) displaying the

highest affinity. Analogue **5** also exhibited enantiomeric selectivity for L-cystine, forming diastereomeric complexes in a 62:38 ratio distinguishable by HNMR spectra.

The TB bis(18-crown-6) ether receptor was revisited twice by the group in collaboration with other departments. In 2021 the TB analogue was used to study interactions between aromatic cavities and non-polar side chains [24]. Later in 2022 the compound was found to display extremely interesting results when consecutively binding two potassium cations, the enthalpy associated to the binding of the second ion was more negative than the first [25].

On the same line, Crossley synthesised different TB analogues containing two porphyrins originating a chiral cleft molecule [26-28].

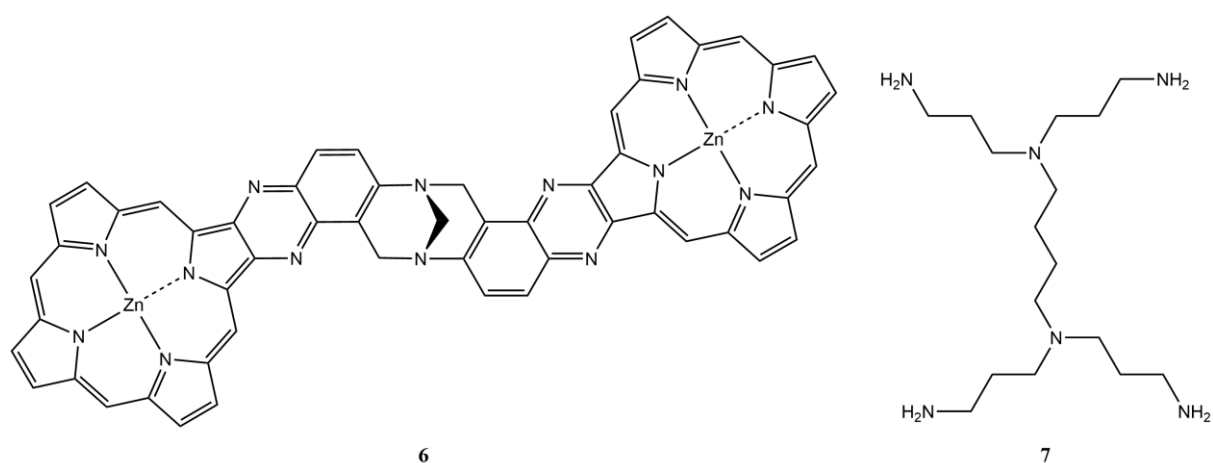


Figure 2.7. Dizinc(II) bis-porphyrin TB receptor (**6**) and tetramine (**7**).

The porphyrin-TB derivatives are known to coordinate to metal ions [26], and when Zn^{2+} is inserted into the porphyrin it displays high affinity towards ditopic diaminoligands [28], and histidine and lysine esters with varying selectivity [27]. Crossley also reported the formation of a molecular capsule as a result of using a dizinc(II) bis-porphyrin TB receptor (**6**; Figure 2.7) and a tetramine dendrimer (**7**; Figure 2.7) [28].

Despite the fall in popularity of Tröger's base, new uses for TB analogues in supramolecular chemistry are still investigated and developed. In 2024, Conghao *et al.* designed and synthesized a new type of macrocycle combining pillararenes and TB (**8a**, **8b**; Figure 2.8) [29]. These macrocycle combinations contain a fixed chirality element (TB) and a conformational chirality element (1,4-alkoxybenzene segment). This combination of fixed and dynamic

stereoelements allow for better understanding and studies of chirality in nature, where the recognition and induction of chirality usually happens together and complement each other.

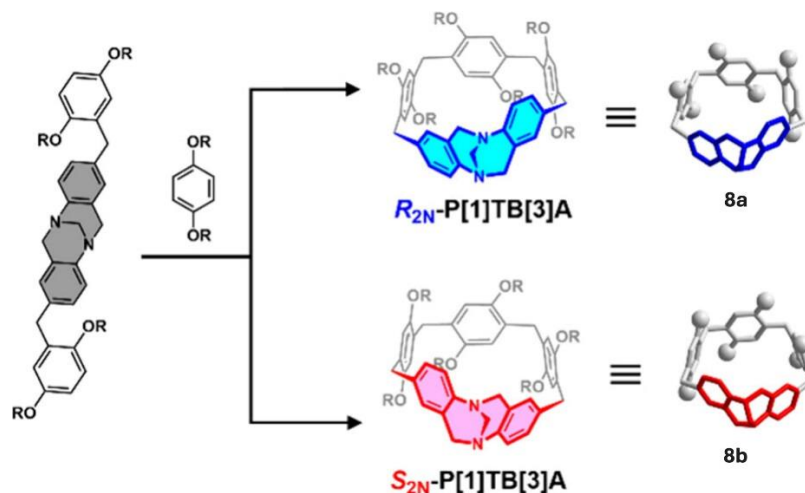
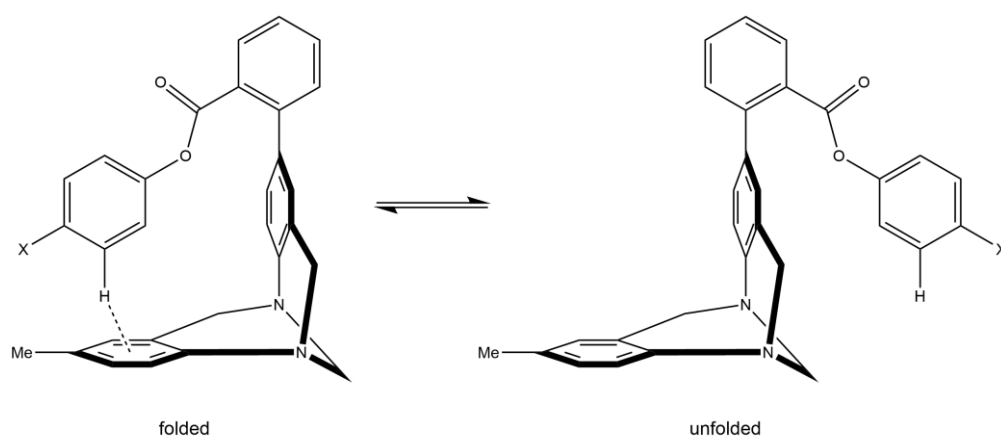


Figure 2.8. Synthetic strategy of P[1]TB[3]A. Estereoisomers of TB-fused pillar[5]arene macrocycles **8a** (R,R) and **8b** (S,S). Image adapted from “Conghao Shi *et al.* Organic Letters 2024 26 (32), 6910-6914”.

2.3.2 Tröger’s base molecular torsion balance

The better understanding of the folding present in proteins has been a known challenge amongst scientists. Therefore, the study of weak forces such as aromatic edge-to-face interactions or CH_3 - π interactions, which play a considerable role in protein folding, is of vital importance.

For this purpose, Wilcox and associates designed a molecular torsion balance with a TB framework (**9**; Figure 2.9) determined the aromatic edge-to-face [30] and CH_3 - π interactions [14]. Further studies by the group, developed an analogue of the previous designed balance which was water soluble and therefore allowed for studies without implementing corrections for the dipole moments between the two conformers of the balance [31]. Further experimentation by Diedrich proved experimentally, the favourable orthogonal dipolar interactions between $\text{C}_{\text{aromatic}}$ -F groups and amide carbonyl groups by using indole analogues of the TB molecular torsion balance [32].



9

Figure 2.9. TB Molecular torsion balance used by Wilcox and coworkers in the determination of aromatic CH₃- π interactions.

2.3.3 Tröger's base interaction with DNA

The use of Tröger's base analogues in conjunction with heterocyclic moieties has proven to have great binding abilities to DNA. Yashima and his group synthesised a 1,10-phenanthroline TB analogue (**10**; Figure 2.10) for the investigation of chiral recognition and interaction with DNA [33]. The TB moiety provides the TB analogues with a favourable orientation to interact with a DNA chain in comparison to the monomeric counterpart (1,10-phenanthroline).

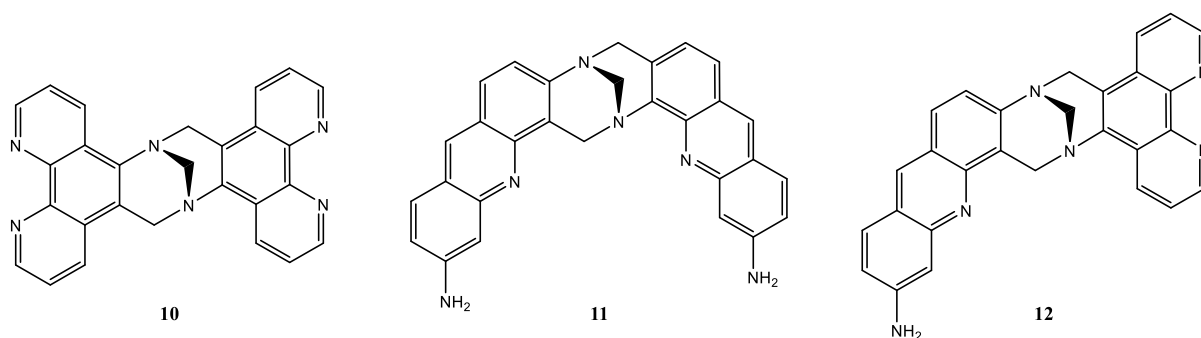


Figure 2.10. Phenanthroline and proflavine TB analogues with DNA binding properties.

Following the steps of Yashima *et al.*, Demeunyk and Lhomme made further investigations on the field, developing multiple TB analogues with binding properties to DNA. With their most notable finds being the proflavine TB analogue **11** and the asymmetric proflavine/phenanthroline TB analogue **12** (Figure 2.10). TB **11** exhibits selectively binding to DNA sequences similarly to calf thymus B-DNA [34]. Whereas TB **12**'s asymmetric nature

contributes to other interesting properties. The acridine ring intercalates between the DNA pairs while the phenanthroline element lies in on DNA groove [35].

Bhattacharya *et al.* described the synthesis of two C_2 -symmetric bisbenzimidazole TB analogues (TBBz1 and TBBz2) (**13**,**14**; Figure 2.11) able to target G-quadruplex DNA (G4DNA) [36]. These ligands have the ability to stabilize the G4DNA complexes, or in some cases, induce a parallel-stranded topology thanks to the discriminatory binding to both groove and end-stacking interactions. Agents capable of inducing DNA into a G4 structure, are potential anticancer drugs and therefore shows the great extent of the TB analogues applications.

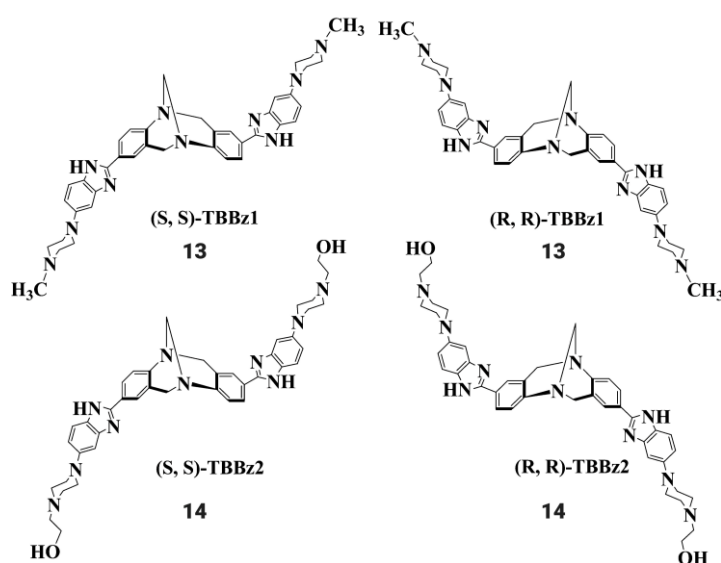


Figure 2.11. The TB analogues TBBz1 and TBBz2 designed and synthesised by Bhattacharya. Image adapted from “Bhattacharya *et al.* J. Med. Chem. 2012, 55, 17, 7460-7471”

2.3.4 Other applications of Tröger’s base

Even after all the other above-mentioned applications, Tröger’s base analogues find uses in many other fields. One example is the TB microporous organic polymer (TB-MOP) designed and synthesised by Lui *et al.* (**15**; Figure 2.12) [37].

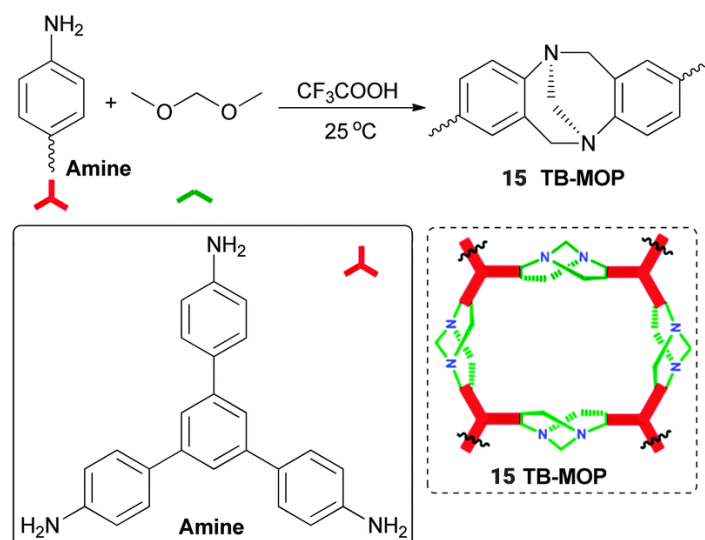


Figure 2.12. Synthetic route and structure of the TB-MOP. Image adapted from “Z. Lui *et al.* Chem. Commun., 2015,51, 1271-1274”.

This TB-MOP excels in CO₂ uptake thanks to its high surface area and has a satisfactory CO₂/N₂ selectivity. When compound **15** coordinates with Ru(III), resulting in TB-MOP-Ru composite, it retains its properties and additionally becomes an active heterogeneous catalyst for the hydrogenation of CO₂ to formic acid. It is thanks to the rigidity of the TB moiety that the polymer gets its key structural properties such as robustness, gas absorption selectivity and catalytic activity.

2.4 Introduction to our group’s Tröger’s base project

The actual Wärnmark’s Tröger’s base project aims for the synthesis of a tubular *heptakis*-TB supramolecular structure for the delivery of drugs through bilayer membranes. Previous research on the topic of artificial ion channels has yielded very interesting results; calixarene, cyclodextrin or peptide based channels have been reported

This ambitious objective is broken down in several steps which help understand the viability and dimensions of the project.

The beginning of the complex synthetic route starts from the commercially available 4-bromo-3-methylaniline (**16**) and goes all the way to the TMS protected TB analogue **17** (Figure 2.13).

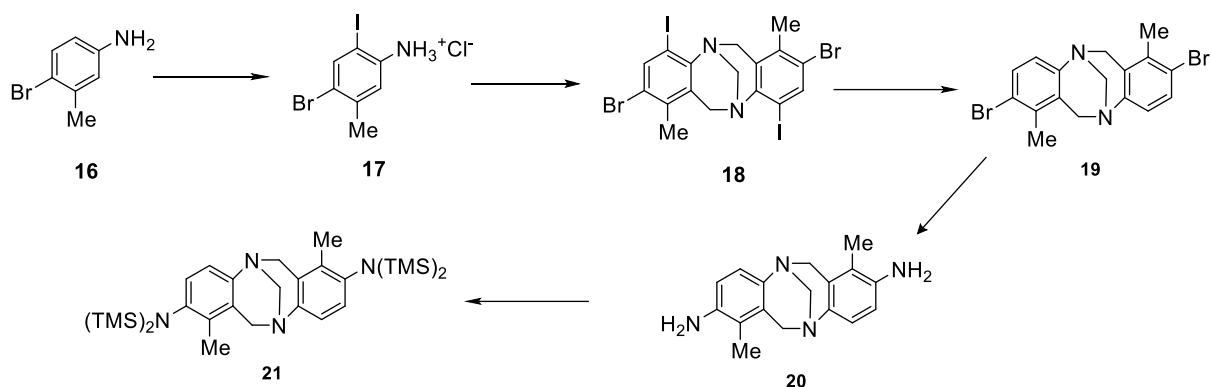


Figure 2.13. The initial route.

In order to obtain a linear and enantiomerically pure *heptakis*-TB, menthyl groups are added to the TB analogue which introduce two chiral centers, thus yielding two diastereomers. Thanks to the chiral centers, it is possible to separate the diastereomeric mixture and continue the designed synthetic routes with the respectively pure TB diastereomers (**22a**, **22b**; Figure 2.14).

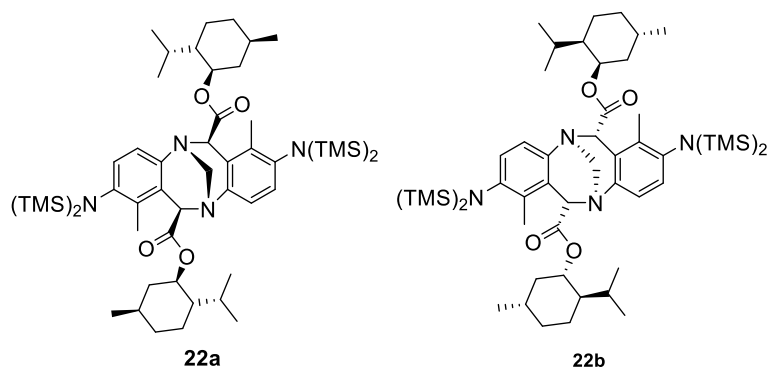


Figure 2.14. The diastereomers created by the introduction of menthyl groups, (+)-**22a** on the left and (–)-**22b** on the right.

This step is also crucial as the menthyl-ester moiety is expected to interact with the bilayer membrane as well as ensuring that when the macrocycle is formed it retains a double radial shape.

The continuation of the synthetic route for **22a** follows very straightforward with the respective protection and condensation until the obtention of an *heptakis*-TB analogue (**28**; Figure 2.15).

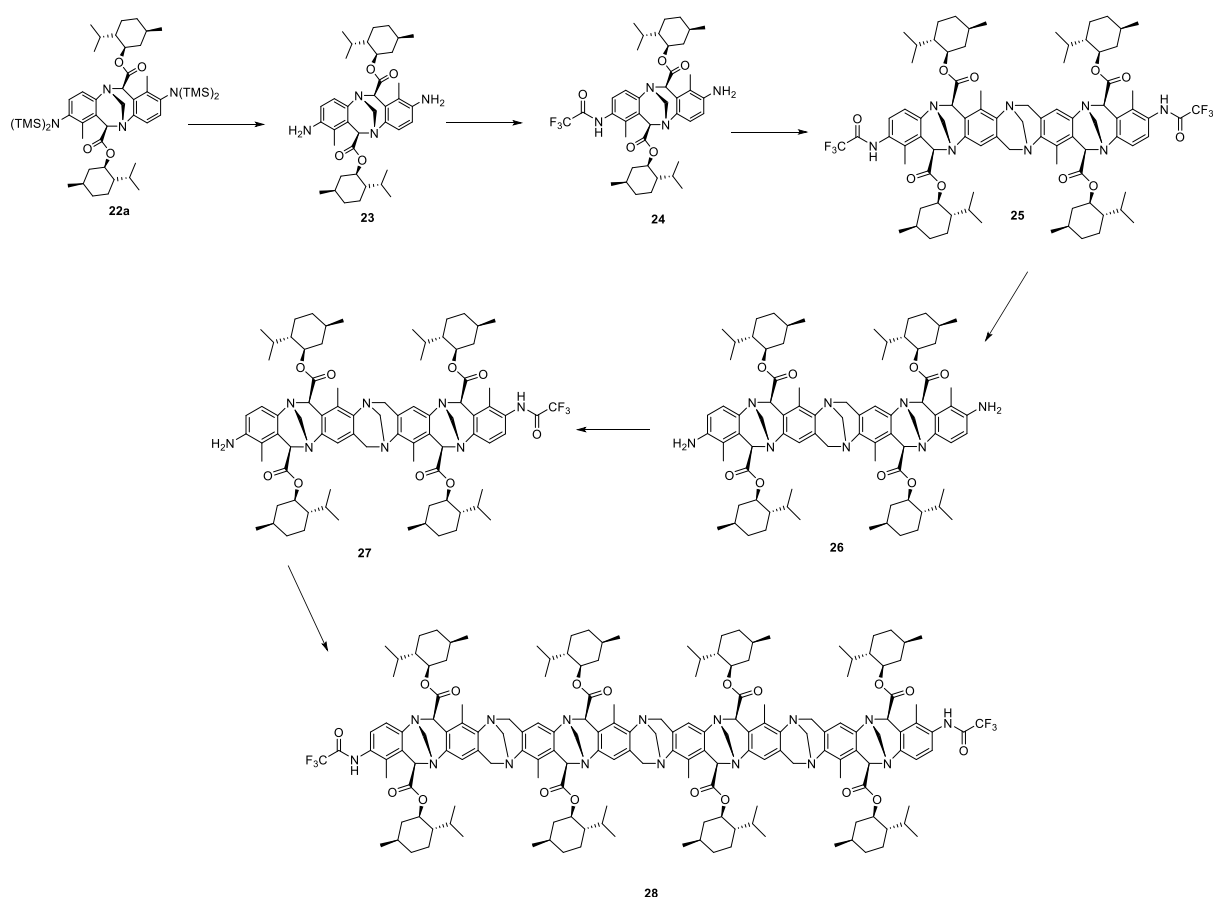


Figure 2.15. The A-route.

The synthesis of **28** was reported by Artacho and Wärnmark in Artacho's PhD thesis [38]. A MALDI-TOF Mass Spectra was run showing promising results on the synthesis of compound **28**.

As for the continuation of the synthetic route for **22b**, the menthyl groups introduced on the last step, are reduced to alcohol groups and subsequently react with dimethyl sulphate yielding **30**. From that point on, the synthetic route follows the same pattern as the A-route, protection of one of the amines, followed by a condensation yielding a new diazonic ring (Figure 2.16). Despite this route showing potential results, it has yet to be explored in depth as the A-route has been the main center of attention in the recent years.

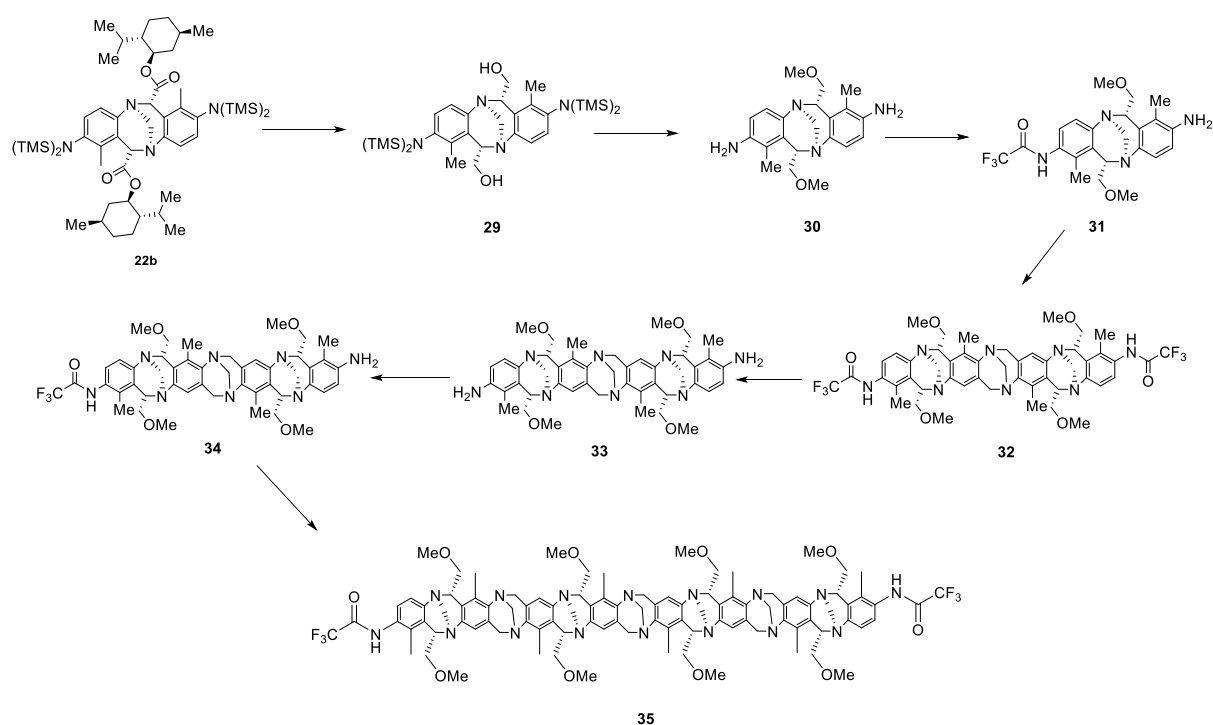


Figure 2.16. The B-route.

If the synthesis of an *heptakis*-TB analogue were to be achieved, the objective of a TB macrocycle that can be functionalised would be closer than ever. These TB tubes would ideally interact with bilayer membranes in order to allow the flux or delivery of drugs, acting as artificial passive carrier protein.

3 Project Outline

The Tröger's base project is focussed on obtaining an *heptakis*-TB analogue. If achieved, it will bring closer than ever the greater objective of a functionalised TB macrocycle expected to interact with bilayer membranes for the delivery of drugs, acting as an artificial passive carrier protein.

It is a complex project, and so, it faces various challenges. The main complication the project is facing right now can be found in step 21 with the separation of diastereomers **22a** and **22b**. Said separation requires a complex column involving poisoning of the silica. Unfortunately, after great efforts by the group to optimize it, the resulting yields for each diastereomer do not surpass 3%. The task assigned for this project is to find a way to reach **25** while going over said bottleneck. After thoroughly discussion, two ideas arise in order to solve the abovementioned problem.

The first one consists of late-stage separation, continuing with a racemic mixture of **22** down the A-route. The separation of diastereomers will be tried with *rac*-**23** and *rac*-**24**. However, purification of *rac*-**25** is deemed not viable since the beginning as the condensation of *rac*-**24** will yield a wide range of products making it difficult to separate the diastereomers, and thus not improving the separation in any way.

The second idea is to condensate **20** in a controlled manner, yielding a simpler *tris*-TB which can be functionalised to obtain an analogue of **25** which can be purified into the respective diastereomers. The suggested synthetic path for the obtention of **37** (Figure 3.1) is based on the A-route and follows the same procedure.

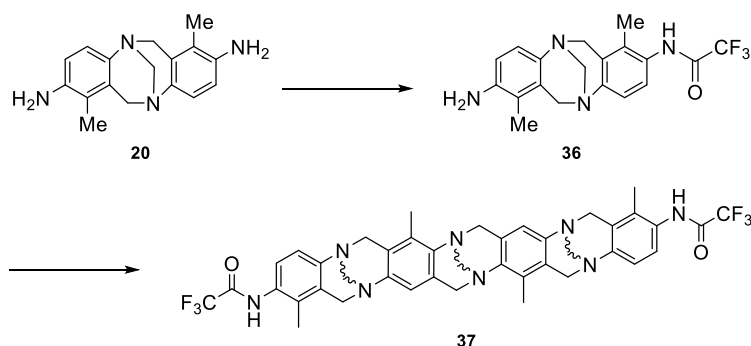


Figure 3.1. Proposed synthetic path for the obtention of **37**.

This option could also solve another big issue with the current synthetic route. XRD of **25** obtained previously by the group (Figure 3.2) shows that the favoured spatial conformation of the molecule is *anti-anti*.

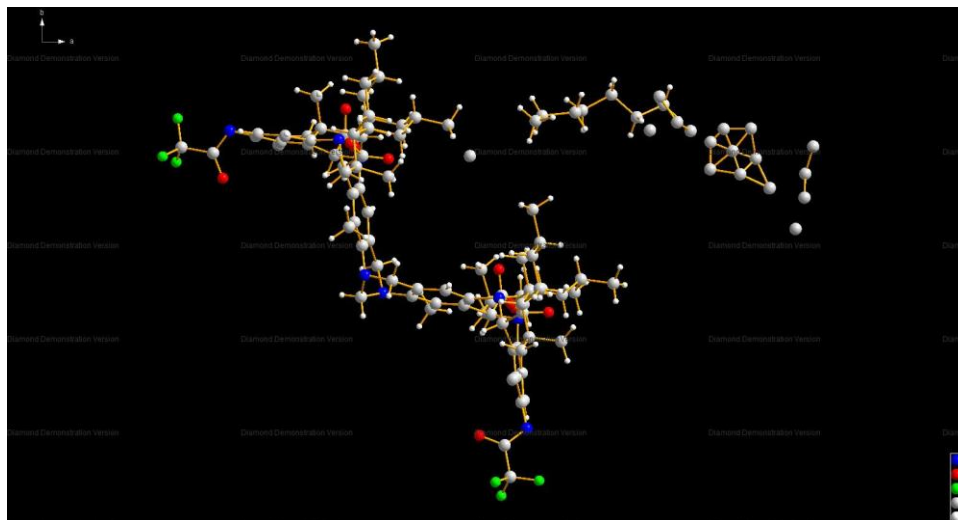


Figure 3.2. XRD characterisation of **25** previously obtained by our group.

For the formation of a cyclic *heptakis*-TB, the *tris*-TB precursor should have a *syn-syn* configuration. Thus, the fact **25** has *anti-anti* configuration poses another problem. However, previous reports by the group show it is possible to synthesise a linear *tris*-TB favouring the formation of cleft isomers and purification of each of the diastereomers formed [39]. Therefore, theoretically both problems would be solved by simply doing a controlled condensation of **20** resulting in **37a**, **37b**, and **37c** (Figure 3.3).

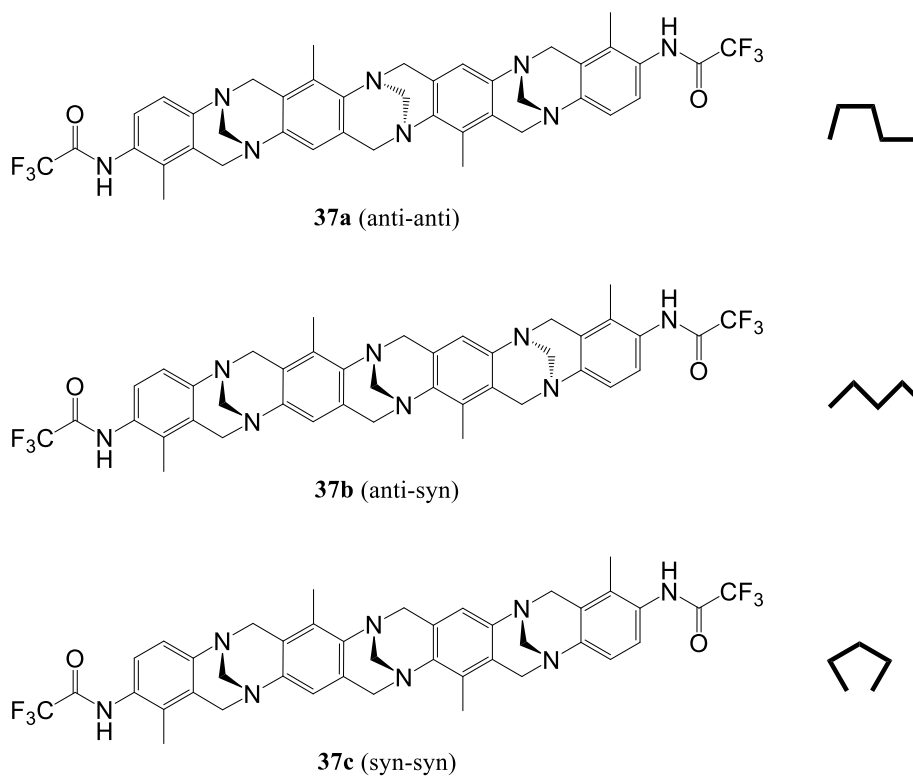


Figure 3.3. All the configuration isomers resulting from the condensation of **36**.

4 Results and discussions

4.1 The initial route

The initial route is the synthetic path to the obtention of **21** from the commercially available 4-bromo-3-methylaniline (Figure 4.1).

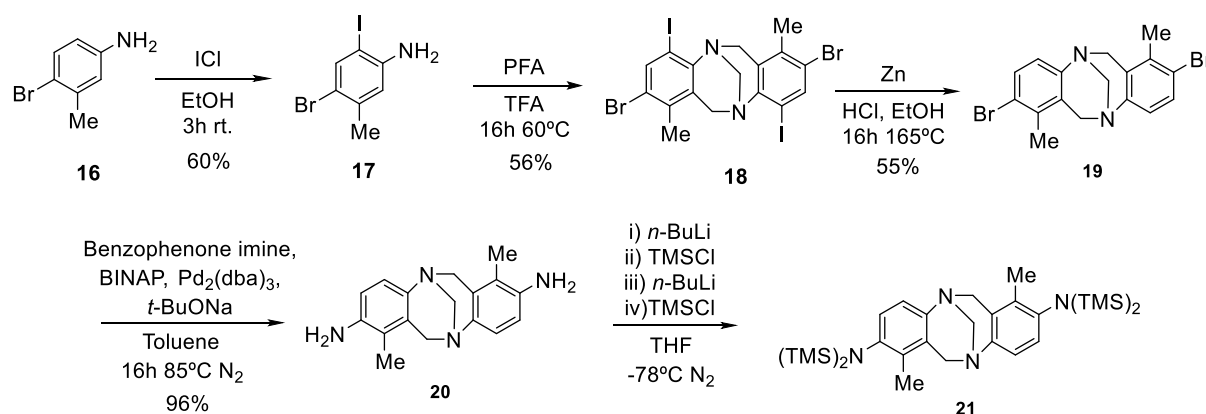


Figure 4.1. Schematic representation of the initial route.

Obtention of **17** is done by halogenation of the para position relative to the methyl (electrophilic aromatic substitution, EAS). Iodine monochloride is used as a reagent due to its strong electrophilic properties. Despite **16** having $C2$ (ortho), $C5$ (meta) and $C6$ (ortho) available positions for EAS, the strongly activating properties of the amine direct the halogenation towards its respective ortho positions ($C2$ and $C6$). Between both ortho positions, $C6$ is more favoured than $C2$ due to the steric impediment of the methyl in position 3. The mechanism of the EAS can be seen in Figure 4.2.

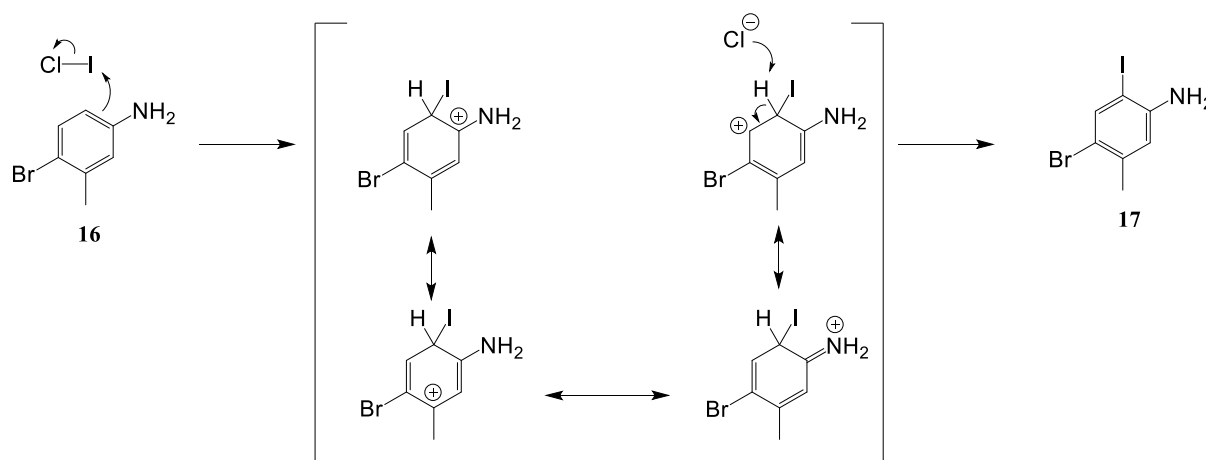


Figure 4.2. Mechanism of halogenation (EAS) for the aniline derivative **16**.

The substituents present in the aniline derivative **17**, are all of crucial importance for the synthesis of the linear-TB. The bromine is present as a support for later functionalization and direction of a new TB unit. The methyl group ensures the linearity of the TB analogues during condensation of the anilines. Finally, the iodization is done to restrict the formation of undesired regioisomers during the condensation of **17**. The obtention of **17** is successfully completed with a 60% maximum yield.

The iodinated aniline is directly condensed, accordingly to the mechanism described in section 2.2 (Figure 2.4), to produce **18** without need of purification of **17**. This reaction can run at rt. but in order to accelerate it, it is run at 60 °C to crack down the PFA. The iodine plays a critical role in reducing the number of side-products this reaction can form. When both ortho positions relative to the amine (C2 and C6) are free at least two side-products result from the intermediates in the TB condensation reported by Wu *et al.* [40]. After purification by DCVC, **18** is obtained with minimal impurities in a 56% maximum yield.

TB analogue **19** is obtained by selective dehalogenation with Zn/HCl. The iodine introduced in the first step have played its role and therefore they are removed in order to avoid undesired side products in the following amination reaction. This reaction usually requires an extensive work up and purification by recrystallization. Instead, the product can be easily obtained by filtration of the precipitate originated after cooling down the solution due to its insolubility in EtOH. This way, **19** is obtained as a white crystalline solid in a 55% maximum yield.

The reductive dehalogenation is followed by an amination of the 2,8-dibromo-TB. The reaction follows a cross-coupling mechanism very similar to a Buchwald-Hartwig reaction (Figure 4.3). The reaction is done according to the previous protocol published by the group [41]. In this case, an imine is used instead of a secondary amine so the resulting imine-TB can be easily hydrolysed into the 2,8-diamino TB **20**.

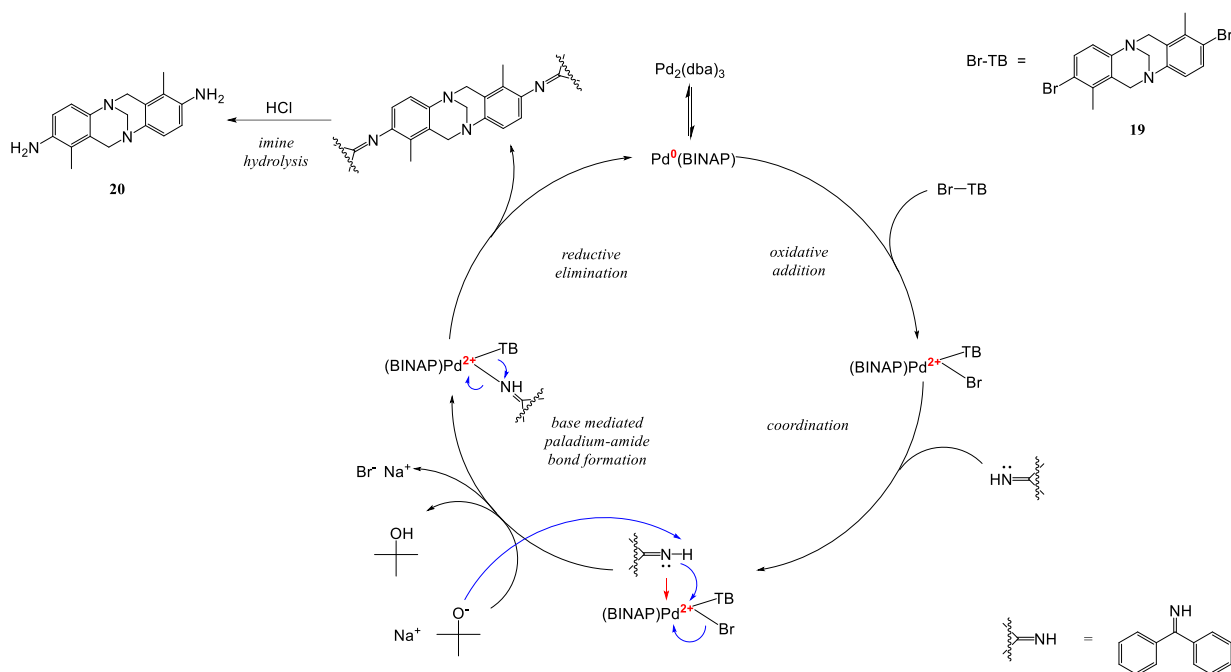


Figure 4.3. Mechanism for the Pd(0) catalysed amination of **19**.

Due to the sensibility of the reagents used, such as the Pd(0) catalyst and *t*-BuONa, the reaction is done under N₂ atmosphere. This same sensibility is what we think is the cause why this reaction failed multiple times, setting back this project. After using brand new reagents, the reaction starts working as usual yielding **20** in over 96% yield.

The initial route finishes with the protection of the primary amines in **20** using TMS chloride (TMSCl). TMSCl is chosen as the protecting group since it can resist the presence of strong bases such as *t*-BuLi, which will be used in the next step of the synthesis. The mechanism for the protection is shown in Figure 4.4. However, the protection of **20** is not done at any point of this work due to the lack of time and the existence of **21** previously synthesised by the group.

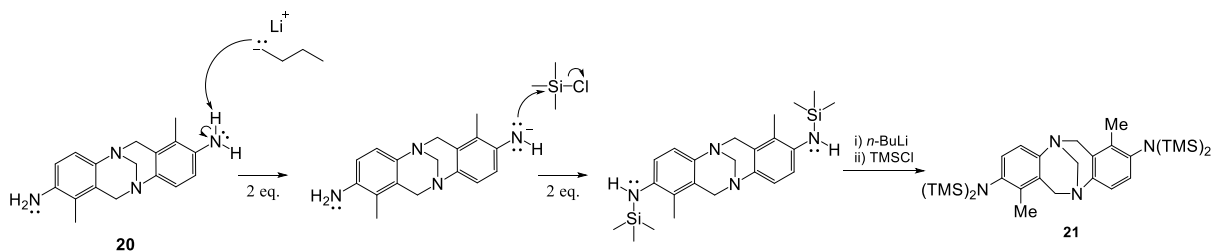


Figure 4.4. Mechanism for the protection of the primary amines in **20** using TMSCl.

4.2 Late-stage separation of diastereomers

As mentioned in section 3, the separation of the diastereomers **22a** and **22b** is a bottleneck in the synthesis of an *heptakis*-TB analogue. A better separation has been tried with the diastereomeric mixtures of **24** and **24'**.

The synthetic route that will be followed in order to try the separation of diastereomers at later stages is very similar to the A-route and the same procedures will be used (Figure 4.5).

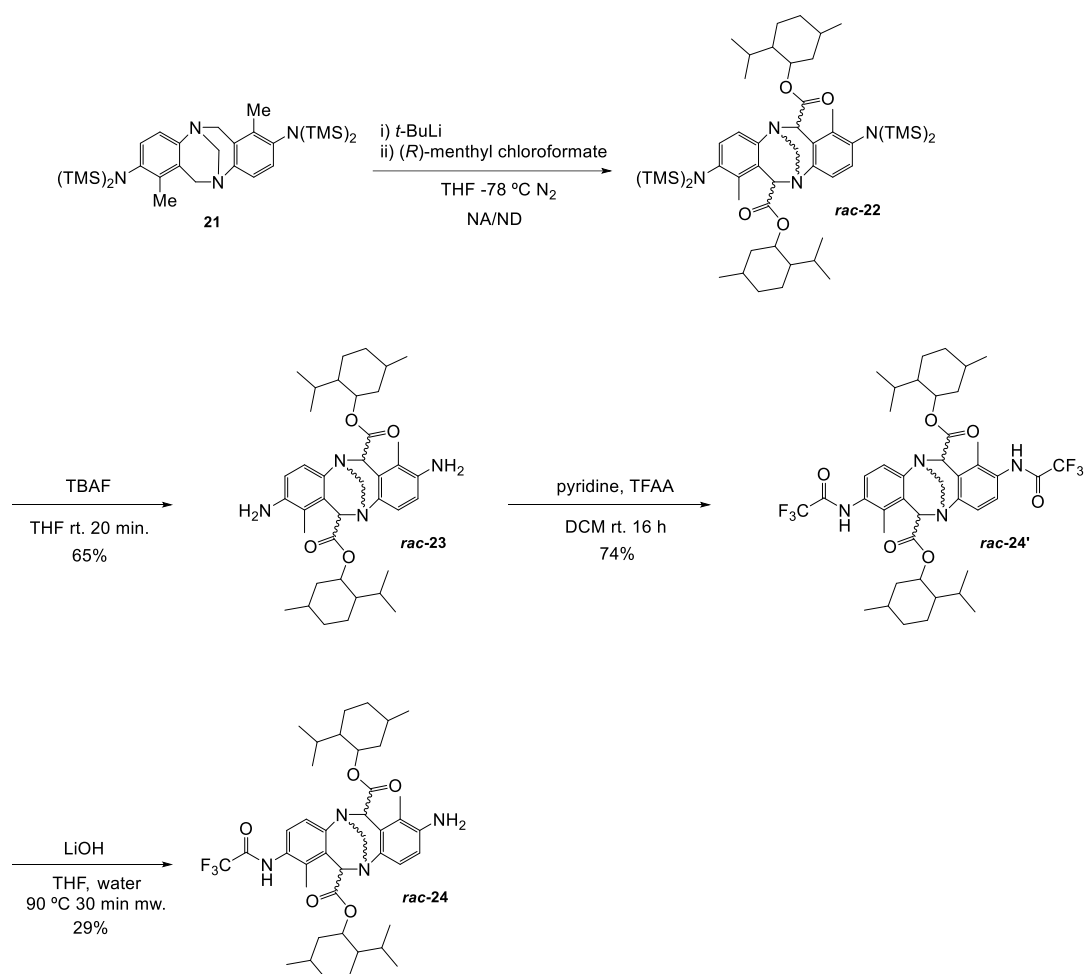


Figure 4.5. Synthetic route proposed for the different compounds in which the late-stage separation will be tried.

The synthesis of **rac-22** is tried from a batch of **21** previously synthesised by the group. The column used for the purification of the crude, is the same one as used for the separation of the diastereomers **22a** and **22b**. It consists of a DCVC with silica poisoned for 1 hour in 1% triethylamine (Et₃N) before packing and a constant 0.2% Et₃N present in every fraction. The

poisoning is done in order to avoid the falling of the amine protecting groups due to the acidic nature of the silica. On the other hand, a constant presence of Et₃N prevents the compound from being completely retained to the silica. Despite the synthesis being successful, **rac-22** is not recovered from the column.

The synthetic route is continued with **rac-22** that had accidentally been synthesized by the group previously. The next step consists in the deprotection of the TMS-protected amines in **rac-22** with TBAF as a fluoride source. The synthesis and purification by DCVC are successful, yielding **rac-23** in a 65% yield.

As the objective of this work is to find an alternative to the already existing purification method for diastereomers **22a** and **22b**, **rac-23** separation is thought to be even more complex than the existing method and therefore is not done. This is due to the interactions between the silica and the amines present in **rac-23**, which greatly increase the retention of the 2,8-diamino TB analogue. Therefore, the synthesis is continued towards the next step.

The synthesis of **rac-24'** is done by addition of a new PG to the amine moieties. TFAA, in presence of pyridine, reacts with the amines introducing a trifluoroacetamide moiety that acts as a PG. The procedure used is the same one the group elaborated for the obtention of **24** from **23** but using double the equivalents of all reagents. This synthesis yields **rac-24'** as the main product in a 74% yield, with **rac-23** and **rac-24** as side products.

With a sample of **rac-24'**, it is evaluated if the separation of the diastereomers is possible by testing different mobile phases for TLC. The results obtained are summarized in Table 4.1.

Sample	Mobile Phase	Separation
1	Hexane:CHCl ₃ 50:50	No
2	Hexane:CHCl ₃ 20:80	No
3	Hexane:CHCl ₃ 0:100	No
4	Hexane:EtOAc 50:50	No
5	Hexane:EtOAc 70:30	No
6	Hexane:EtOAc 80:20	No
7	Hexane:EtOAc 85:15	No
8	Hexane:EtOAc 90:10	No
9	Hexane:EtOAc 95:05	No
10	Hexane:EtOAc 97:03*	No

Table 4.1. Results of the different mobile phases used for the TLC tests with ***rac-24'***. * Mobile phase used in the TLC realized for the separation of **22a** and **22b**.

As seen in Table 4.1, in none of the conditions tried the separation of the diastereomers can be appreciated. Therefore, it is decided to go a “step back” and synthesise ***rac-24*** and try the separation with said compound.

To get ***rac-24*** from ***rac-24'***, only one of the PG must be removed. This is achieved by leaving the reaction for half the time as it is needed to remove both PGs. The reaction is done in a microwave using LiOH as the source of nucleophiles. Synthesis of ***rac-24*** is successful in a 29% yield, with ***rac-24'*** and ***rac-23*** recovered as side products in a 39:20:41 ratio, respectively.

The separation of the diastereomers is this time tried with a sample of ***rac-24***. The results are summarized in Table 4.2.

Sample	Mobile Phase	Separation
1	Heptane:CHCl ₃ 50:50	No
2	Heptane:CHCl ₃ 0:100	No
3	Heptane:EtOAc 50:50	No
4	Heptane:EtOAc 60:40	No
5	Heptane:EtOAc 70:30	No
6	Heptane:EtOAc 75:25	No
7	Heptane:EtOAc 80:20	No

Table 4.2. Results of the different mobile phases used for the TLC tests with ***rac-24***.

The results obtained for this separation are not successful either. No more mobile phases are tried as the whole point of the project is to find better conditions than the DCVC that is currently being used not just find similar conditions at a later stage.

After these unsuccessful tries, the synthesis is not continued with the condensation of ***rac-24*** due to the different configurational isomers that could form: the *anti-anti*, the *syn-anti*, and the *syn-syn* (with two diastereomers each). As this separation would require a lot of effort and time, this part of the work is considered as concluded in order to allocate the remaining time to complete the linear *tris*-TB part of the work which could yield very promising results.

4.3 Synthesis of a linear *tris*-Tröger's base

For this project, a simpler linear *tris*-Tröger's base is synthesised, which would be later functionalised and purified or condensed again to form an *heptakis*-TB. The procedures used will be very similar to the ones of the A-route, maintaining the scale and molar equivalents. A schematic of the proposed synthetic route is shown in Figure 4.6.

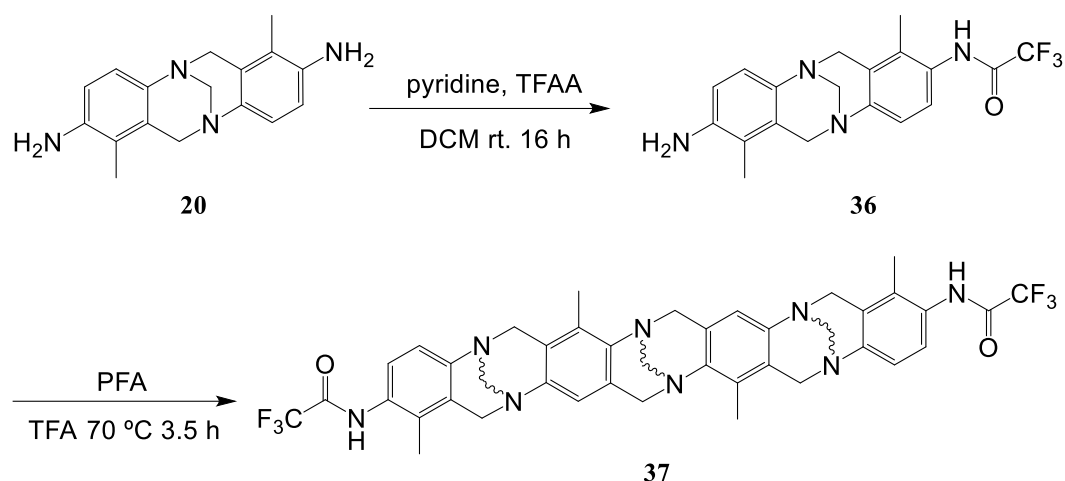


Figure 4.6. Synthetic route proposed for the obtention of the linear *tris*-TB **37**.

To get **36** the same procedure elaborated for the obtention of **24** from **23**, in the A-route, is used. TFAA in presence of pyridine with **20**, yields **36** as a TB analogue with only one amine protected. This allows controlled condensation of a TB to create a new diazocine ring preventing polymerization. With synthesis being successful, a new DCVC has to be designed for this reaction as the one mentioned in the used protocol does not work for the newly synthesised compound. The fractions designed to obtain **36** as a pure compound are as follows: 50% EtOAc in CHCl₃, 80% EtOAc in CHCl₃, 0.1% MeOH in EtOAc, 10% MeOH in EtOAc and 20% MeOH in EtOAc. However, the different compounds elute very closely, and new fractions need to be introduced between each of the previously mentioned ones. Even though the column still has room for improvement, **36** is obtained as a pure compound in a 29% yield.

The synthesis follows with the same procedure as for the condensation of **24** into **25**. To a solution of **36** in TFA, PFA is added in excess and left to react at 70 °C for only 4 hours. An aliquot is taken after 1 hour to see if the reaction is done, as the molecules to be condensate are less voluminous. However, after the proper work up, a lot of starting material can still be identified by HNMR. The reaction is left to react for the 4 hours, and a sample of the crude, after the work up, is taken for LC-MS (normal phased liquid chromatography; Water:MeCN 95:5). Despite many peaks present in the LC-MS spectra, the mass of compound **37** (788.8 g/mol) can be detected ($m/z=789.2$; note that the LC-MS instrument uses electrospray ionization which generates $[M+H]^+$ ions) (Figure 4.7). The corresponding isotopic peaks predicted with ChemDraw ($m/z=100\%$; $m+1/z=44.9\%$; $m+2/z=10.2\%$) match with the isotopic

pattern observed in the MS spectra ($m/z(789.2)=100\%$; $m+1/z(790.2)=44.1\%$; $m+2/z(791.2)=11.7\%$). This evidence further proves the successful synthesis of **37**.

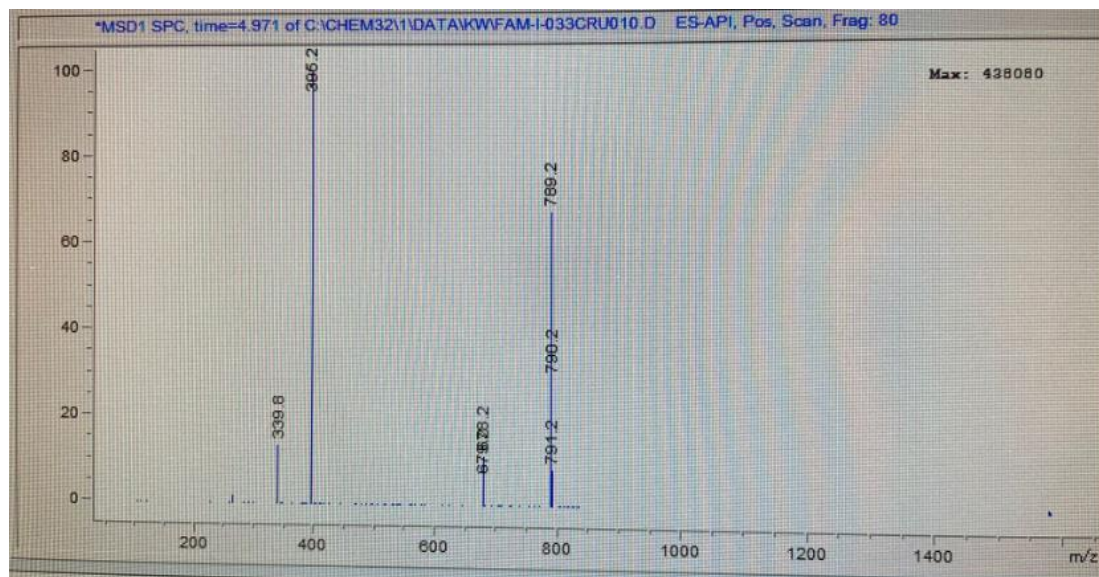


Figure 4.7. Mass spectra at 4.971 minutes retention time. m/z peak at 789.2 matches with compound **37**.

On the same spectra, starting material (**36**; 376.4 g/mol; $m/z=377.2$) and **37** without one of the trifluoroacetamide PG (**37'**; 692.4 g/mol; $m/z=693.2$) can also be identified (Figure 4.8). The peak observed in Figure 4.8 (below), not only matches with **37'**'s m/z value minus the mass of the COF_3 moiety, but it also matches the isotopic pattern observed in Figure 4.7. This observation provides us with credible proof that this peak belongs in fact to **37'**.

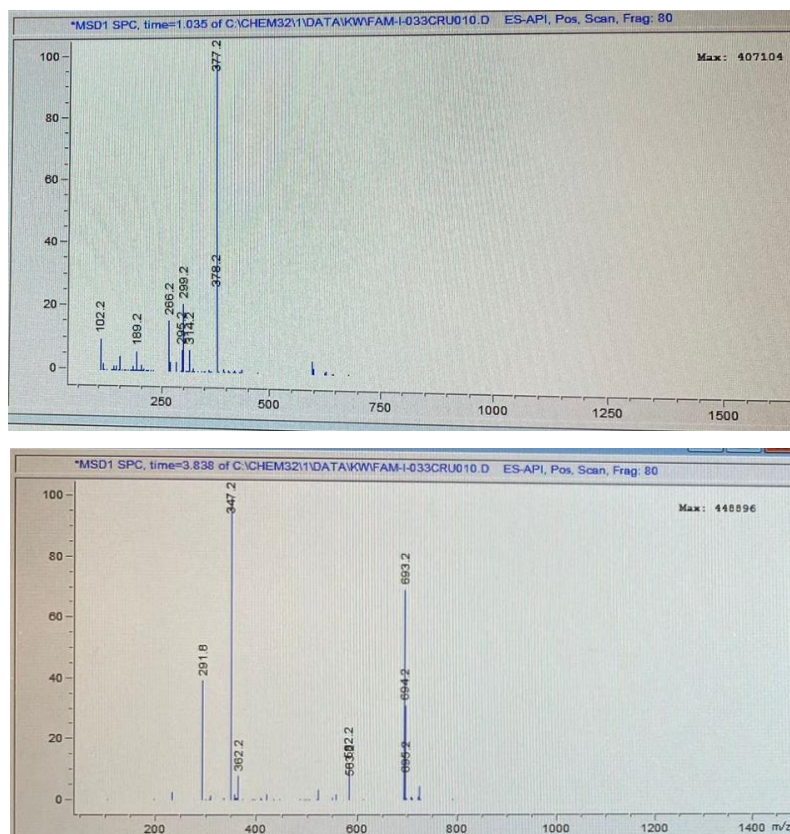


Figure 4.8. MS spectra at 1.035 min (top), where starting material (**36**) can be identified. MS spectra at 3.838 min (below), where compound **37** without a trifluoroacetamide PG.

Due to lack of time, the purification of **37** and later separation of its corresponding configurational isomers has not been achieved.

5 Conclusions

The Tröger's base project has been present in the group for a long time, and therefore many subprojects have been elaborated as well as being in constant evolution. After landing in my hands, here is my contribution to the TB project.

For the late-stage separation of diastereomers, it has been proven how difficult it is to achieve such task. Not only is it an arduous and complex separation the currently used one, but an improved purification of the diastereomers is not possible at later stages in the synthesis. Hopefully the conclusions drawn from this study will allow to explore other options to tackle the introduction and purification of diastereomers in our TB analogue.

Regarding the synthesis of a simpler linear *tris*-TB analogue with options of later functionalisation, not many conclusions can be drawn unfortunately. Despite having some leads towards the confirmation of a successful synthesis of **37**, not enough data has been provided in order to make a sound statement. However, if enough data is gathered, the possibility of separating the different configurations of **37** by CC could be a big breakthrough for the project.

Despite not having extremely favourable results, I'm positive that the information gathered with this study will be of use for the success of the group's Tröger's base project.

On a different note, it must be added that this work has had a huge impact on my chemical knowledge. I have gained expertise in various topics such as total synthesis and inert-Schlenk techniques. The degree of independence given and expected, has provided me with critical skills such as problem solving as well as greater ease and confidence in a laboratory. Lastly, by working in a research group I have had the opportunity to be taught a wide range of tips and tricks by much more knowledgeable people such as my supervisor and examiner.

6 Future aspects

As for the future of the Tröger's base project, there are many new opportunities to explore. However, if the main problems exposed with this study want to be solved, the following topics should be further delved into.

Regarding the topic of diastereomers, reconsideration towards when the chiral centers are introduced to the TB analogue is suggested. Otherwise, new and more adjusted purification methods to the existing setback should be explored.

Respecting the synthesis of a linear *tris*-TB, the future plans are more straightforward. A purification method should be developed for **37**. As well, the possibility of purification of **37a**, **37b** and **37c** by DCVC or preparative TLC should be thoroughly explored. If the separation of the configurational isomers is successful, a big step forward will have been set towards the synthesis of a cyclic *heptakis*-TB analogue.

7 Experimental procedures

All commercial reagents were used as received, unless otherwise stated. Dry DCM and toluene were collected from a dry solvent dispenser (MBRAUN SPS-800) and stored over 4 Å MS. Dry pyridine was distilled from calcium hydride and stored over 4 Å MS. THF was distilled from sodium and benzophenone ketyl and used directly. Reactions run at room temperature are in the range of 20-23 °C. Metal heating mantels were used to achieve required reaction temperatures. Microwave reactions were run using a Biotage Initiator. All dropwise additions were conducted using an NE-4000 programmable 2 channel syringe pump. Dry vacuum column chromatography was carried out on silica gel column (Dry-column silica) (Silica gel 60 (0.015-0.040 mm) for column chromatography. CAS No. 7631-86-9, EC Number 231-545-4, From Merck). Petroleum Ether, PE, for column chromatography is the fraction boiling at 40-60 °C. Analytical thin-layer chromatography (TLC) was carried out on TLC Silica gel 60 F254 and visualized with a UV-lamp (254 nm). ¹H NMR and ¹³C NMR were recorded at rt on a Bruker Avance II at 400.1 MHz. The spectra were recorded in CDCl₃ or MeOH-*d*₄ and the residual solvent signals (7.26/77.16; 3.31/49.00 ppm, respectively) were used as reference. Chemical shifts (δ) expressed in parts per million (ppm) and coupling constants (J) are reported in Hertz (Hz). The following abbreviations are used to indicate apparent multiplicities: s, singlet; br, broad singlet; d, doublet; dd, doublet of doublets; dq, doublet of quartets; t, triplet; td, triplet of doublets, q, quartet, m, multiplet.

4-Bromo-2-iodo-5-methylaniline hydrochloride (**17**).

4-Bromo-3-methylaniline **16** (50.0 g, 0.269 mol) was added to a solution of brand new ICl (15.8 mL, 0.315 mol) in EtOH (99.5%, 550 mL) at 0 °C over 5 minutes. The resulting slurry was stirred for 3h at rt. The reaction mixture was filtrated under vacuum. The filter cake was washed with acetone until a light pink coloured powder was obtained as **16** (51.7 g in total). ¹H NMR (400.1 MHz, MeOH-*d*₄): δ = 8.07 (s, 1H, H-3), 7.22 (s, 1H, H-6), 2.37 (s, 3H, CH₃) ppm.

rac-2,8-Dibromo-4,10-diiodo-1,7-dimethyl-6*H*,12*H*-5,11-methanodibenzo[*b,f*][1,5]diazocine (*rac*-**18**).

Compound **17** (5.00 g, 14.4 mmol) was dissolved in TFA (40 mL) and paraformaldehyde (1.04 g, 34.6 mmol) was added to the solution under stirring. The reaction mixture was heated up to 60 °C under stirring. After 16 h, TFA was removed in vacuo and then water (25 mL) was added. The resulting slurry was made alkaline (pH 8-10) by slow addition of NH₃ (aq, 32%). A few drops of DCM were added to dissolve the so formed solid and the resulting mixture was again made alkaline (pH 8-10). The resulting biphasic solution was then extracted with DCM (4 x 50 mL). The combined red organic phases were washed twice with Na₂S₂O₃ (aq, sat.), followed by brine (50 mL). The resulting organic phase was dried over anhydrous MgSO₄, filtrated and DCM was removed in vacuo resulting in a yellow solid.

Purification method: Up to three batches prepared as above were combined and dissolved in DCM. Dry-column silica (~3 g per batch) was added to the solution, the resulting mixture was evaporated to dryness in vacuo. The resulting solid was added to a dry vacuum column (4x5 cm, PE:CHCl₃ gradient from 30% CHCl₃ to 70% increasing the gradient by 5% every 100ml 2x50mL fractions) to give *rac*-**18** as a colourless solid in a 60% yield from **17** (over one step).

R_f = 0.35 (Hept:CHCl₃, 1:1); ¹H NMR (400.1 MHz, CDCl₃): δ = 7.94 (s, 2H, H-3 and H-9), 4.40 (d, *J* = 17.4 Hz, 2H, H-6x and H-12x), 4.24 (s, 2H, H-13), 4.18 (d, *J* = 17.4 Hz, 2H H-6n and H-12n), 2.18 (s, 6H, Ar-CH₃) ppm; ¹³C NMR (100.4 MHz, CDCl₃): δ = 146.9, 140.4, 135.7, 130.4, 121.8, 94.9, 66.2 (1C), 56.3, 17.8 ppm.

***rac*-2,8-Dibromo-1,7-dimethyl-6*H*,12*H*-5,11-methanodibenzo[*b,f*][1,5]diazocine (*rac*-19).**

To a suspension of *rac*-18 (34.2 g, 52.0 mmol) in EtOH (95%, 2.5 L), zinc dust (68.1 g, 1.04 mol) was added, followed by HCl (aq, 10%, 50 mL) and the resulting mixture was heated to 165 °C under stirring. After 16 h, the mixture was filtrated hot through celite and the filter cake was washed with refluxing EtOH (250 mL), followed by refluxing DCM (250 mL). The filtrate was evaporated in vacuo to yield a slurry which was dissolved in DCM (500 mL) and the resulting solution was washed with water (2 x 200 mL). The combination of aqueous phases was made alkaline (pH 8-10) by slow addition of NaOH (aq, 1.0 M) and extracted with DCM (400 mL). The combination of organic phases from both extractions was washed with water (500 mL), followed by brine (500 mL), dried over MgSO₄, and evaporated to dryness in vacuo. The resulting reddish solid was recrystallized from refluxing EtOAc (250 mL), to give white crystals *rac*-19 in 55% yield.

R_f = 0.17 (Hept:EtOAc 85:15); ¹H NMR (400.1 MHz, CDCl₃): δ = 7.37 (d, *J* = 8.6 Hz, 2H, H-3 and H-9), 6.91 (d, *J* = 8.6 Hz, 2H, H-4 and H-10), 4.54 (d, *J* = 16.8 Hz, 2H, H-6x and H-12x), 4.20 (s, 2H, H-13), 4.14 (d, *J* = 16.8 Hz, 2H, H-6n and H-12n), 2.17 (s, 6H, Ar-CH₃) ppm; ¹³C NMR (400.1 MHz, CDCl₃): δ = 147.5, 135.1, 131.4, 128.0, 124.3, 120.1, 65.5 (1C), 58.3, 18.0 ppm.

***rac*-1,7-Dimethyl-6*H*,12*H*-5,11-methanodibenzo[*b,f*][1,5]diazocine-2,8-diamine (*rac*-20).**

BINAP (0.117 g, 0.180 mmol, 1.5 mol%) and Pd₂(dba)₃ (0.0786 g, 0.0860 mmol, 0.68 mol%) were added to a 100-mL flask, containing *rac*-19 (5.00 g, 12.3 mmol), *t*-BuONa (3.54 g, 36.8 mmol) was added and the flask flushed 5 times with vacuum/nitrogen and dry toluene (50 mL) was added, followed by benzophenone imine (4.6 mL, 27 mmol). The reaction was heated to 85 °C and left stirring overnight under N₂ atmosphere. The resulting brown suspension was concentrated to a slurry in vacuo and THF (250 mL) was added followed by HCl (2 M, 50 mL) and was stirred at rt. for 4 h. Additional HCl (2 M, 25 mL) was added and Hept:EtOAc (2:1, 150 mL) was used to partition the phases. The resulting organic phase was extracted using HCl (2 M, 50 mL). The combined aqueous phases were then washed with Hept:EtOAc (2:1, 200 mL), DCM (200 mL) and Hept:EtOAc (2:1, 2x100 mL). The resulting aqueous phase was made alkaline (pH 10-12) by slow addition of NaOH (aq, 10 M), extracted using CHCl₃ (3x200 mL).

The combined CHCl₃ phases were washed with brine (300 mL), dried over MgSO₄, filtered and evaporated to dryness in vacuo giving **rac-20** as a light yellowish granular solid in >95% yield.

R_f = 0.21 (MeOH:CHCl₃, 5:95); ¹H NMR (400.1 MHz, CDCl₃): δ = 6.91 (d, *J* = 8.5 Hz, 2H, H-4 and H-10), 6.58 (d, *J* = 8.5 Hz, 2H, H-3 and H-9), 4.43 (d, *J* = 16.6 Hz, 2H, H-6x and H-12x), 4.25 (s, 2H, H-13), 4.12 (d, *J* = 16.7 Hz, 2H, H-6n and H-12n), 3.42 (br, 4H, -NH₂), 1.88 (s, 6H, Ar-CH₃) ppm; ¹³C NMR (400.1 MHz, CDCl₃): δ = 140.7, 139.0, 126.1, 122.9, 119.4, 115.3, 65.4 (1C), 57.5, 11.2 ppm.

(+)-Bis((1*R*,2*S*,5*R*)-2-isopropyl-5-methylcyclohexyl) (5*S*,6*R*,11*S*,12*R*)- 1,7-dimethyl-*N*²,*N*²,*N*⁸,*N*⁸-tetrakis(trimethylsilyl)-6*H*,12*H*-5,11-methanodibenzo[*b,f*][1,5]diazocine-6,12-dicarboxylate ((+)-22a) and (-)-bis((1*R*,2*S*,5*R*)-2-isopropyl-5-methylcyclohexyl)(5*R*,6*S*,11*R*,12*S*)- 1,7-dimethyl-*N*²,*N*²,*N*⁸,*N*⁸-tetrakis(trimethylsilyl)-6*H*,12*H*-5,11-methanodibenzo[*b,f*][1,5]diazocine-6,12-dicarboxylate ((-)-22b).

t-BuLi (3.33 mL, 5.3 mmol, 1.6 M) was added over 40 minutes to a stirred solution of **rac-21** (1.00 g, 1.76 mmol) in anhydrous THF (17 mL) at -78 °C under N₂ atmosphere. The reaction was left to stir at -78 °C for 2 h. (1*R*)-(-)-Menthyl chloroformate (1.65 mL, 7.80 mmol) was added to the resulting mixture at -78 °C over 30 minutes and the mixture was allowed to slowly reach rt. in the cooling bath over 16 h under stirring. THF was removed in vacuo and the resulting oil was diluted with EtOAc (50 mL) and washed with water (2x20 mL), brine (20 mL), dried over MgSO₄, and evaporated in vacuo.

Purification method: The reaction crude was dissolved in DCM. Then celite (10 g) was added to the solution and the solvent was evaporated to dryness in vacuo. The resulting powder was loaded on a dry vacuum column (4x7.5 cm, Hept:DCM with a constant 0.2% triethylamine in every fraction. 50 mL fractions were loaded in a gradient of DCM (%) of: 0, 10, 15, 17.5, 20, 22 and then successively increasing 1% up to 35%). The silica gel was deactivated under stirring in 1% triethylamine in heptane for 1 h, packed and washed with heptane (2x50 mL) before loading. The column yields diastereoisomer **(+)-22a** as a (-) solid in (-)% yield, and diastereoisomer **(-)-22b** as a (-) solid in (-)% yield, together with a mixture of both diastereoisomers in (-)% yield.

Tip: 35g of DC-C silica makes a 7.5 cm column.

***rac*-Bis((2-isopropyl-5-methylcyclohexyl)-2,8-diamino-1,7-dimethyl-6*H*,12*H*-5,11-methanodibenzo[*b,f*][1,5]diazocine-6,12-dicarboxylate (*rac*-23).**

To a solution of *rac*-22 (1.02 g, 1.09 mmol) in THF (23 mL) was added TBAF (4.8 mL, 4.8 mmol, 1M in THF) and the resulting solution was vigorously stirred for 20 minutes under rt. THF was removed in vacuo, and the resulting residue was dissolved in EtOAc (15 mL). The resulting solution was washed with water (2x8 mL), brine (10 mL), dried over anhydrous MgSO₄, filtered and evaporated to dryness in vacuo.

Purification method: The reaction crude was dissolved in DCM. Dry-column silica (4 g) was added to the solution, the resulting mixture was evaporated to dryness in vacuo. The resulting solid was added to a dry vacuum column (4 x 5 cm, Hept:EtOAc gradient from 40% EtOAc to 75% increasing the gradient by 5% every 100ml) to yield a colourless solid *rac*-23 in 65% yield.

R_f = 0.42 (Hept:EtOAc 1:1); ¹H NMR (400.1 MHz, CDCl₃): δ = 7.02 (d, *J* = 8.5 Hz, 2H, H-4 and H-10), 6.65 (d, *J* = 8.5 Hz, 2H, H-3 and H-9), 4.81 (td, *J* = 10.8, 4.3 Hz, 2H, -CO₂ CH-R₂), 4.61 (s, 2H, H-6 and H-12), 4.40 (s, 2H, H-13), 3.42 (br, 4H, -NH₂), 2.09-1.97 (m, 4H, menthyl), 1.82 (s, 6H, Ar-CH₃), 1.74-1.68 (m, 4H, menthyl), 1.56-1.46 (m, 4H, menthyl), 1.12-1.02 (m, 4H, menthyl), 0.94-0.84 (m, 14H, menthyl), 0.79 (d, *J* = 7.0, 2H, menthyl) ppm; ¹³C NMR (400.1 MHz, CDCl₃): δ = 170.9, 141.2, 140.8, 124.9, 123.9, 120.0, 116.3, 75.4, 68.9, 58.7 (1C), 46.9, 40.5, 34.4, 31.6, 26.2, 23.2, 22.2, 21.1, 16.0, 12.6 ppm.

***rac*-Bis(2-isopropyl-5-methylcyclohexyl)-1,7-dimethyl-2,8-bis(2,2,2-trifluoroacetamido)-6*H*,12*H*-5,11-methanodibenzo[*b,f*][1,5]diazocine-6,12-dicarboxylate (*rac*-24').**

Anhydrous pyridine (126 μL, 1.54 mmol) was added to a solution of *rac*-23 (879 mg, 1.30 mmol) in anhydrous DCM (204 mL) under inert N₂ atmosphere at rt under stirring. The resulting solution was stirred for 10 minutes before trifluoroacetic anhydride (216 μL, 1.534 mmol) in anhydrous DCM (2.00 mL) was added drop wise under vigorous stirring over 2.5 h. After the addition, the reaction was left stirring for 16 h at rt. under stirring. The DCM was removed in vacuo yielding a brown solid.

Purification method: The reaction crude was dissolved in DCM. Dry-column silica (3.5 g) was added to the solution, the resulting mixture was evaporated to dryness in vacuo. The resulting solid was added to a dry vacuum column (4x5 cm, Hept:EtOAc gradient from 10% EtOAc to

75% increasing the gradient by 5% every 100ml) Bis-trifluoroacetylated ***rac*-24'** was isolated as a colourless solid in 74% yield.

R_f = 0.84 (Hept:EtOAc 50:50)

***rac*-Bis((1R,2S,5R)-2-isopropyl-5-methylcyclohexyl)(5S,6R,11S,12R)-8-amino-1,7-dimethyl-2-(2,2,2-trifluoroacetamido)-6H,12H-5,11-methanodibenzo[b,f][1,5]diazocine-6,12-dicarboxylate (*rac*-24).**

The bis-trifluoroacetylated compound ***rac*-24'** (506 mg, 0.691 mmol) was dissolved in THF (4 mL) in a microwave oven reaction tube. LiOH (28.2 mg, 1.18 mmol) was added to the solution. Water (0.5 mL) was added to the mixture to form a biphasic mixture. The mixture was reacted in a microwave oven at 90 °C for 30 min (400 watts). The reaction mixture was allowed to reach rt. before THF was removed in vacuo and the resulting residue was dissolved in EtOAc (15 mL). The mixture was washed with water (7 mL) and the phases were separated. The aqueous phase was further extracted with EtOAc (2x5 mL) and CHCl₃ (5 mL). The combined organic phases were washed with brine (20 mL), dried over MgSO₄, filtrated and evaporated to dryness in vacuo.

Purification method: The reaction crude was dissolved in DCM. Dry-column silica (3 g) was added to the solution, the resulting mixture was evaporated to dryness in vacuo. The resulting solid was added to a dry vacuum column (4x5 cm, Hept:EtOAc gradient from 10% EtOAc to 75% increasing the gradient by 5% every 100ml) to isolate ***rac*-24** in a 29% yield.

R_f = 0.24 (Hept:EtOAc 70:30); ¹H NMR (400.1 MHz, CDCl₃): δ = 7.74 (s, 1H, -NH-COF₃), 7.44 (d, *J* = 8.7 Hz, 1H, H-4), 7.22 (d, *J* = 8.7 Hz, 1H, H-3), 6.99 (d, *J* = 8.4 Hz, 1H, H-10), 6.64 (d, *J* = 8.4 Hz, 1H, H-9), 4.81 (dq, *J* = 10.8, 4.4 Hz, 2H, -CO₂ CH-R₂), 4.68 (s, 1H, H-12), 4.59 (s, 1H, H-6), 4.45 (d, *J* = 13.3 Hz, 1H, H-13a), 4.35 (d, *J* = 13.4 Hz, 1H, H-13b), 3.20 (s, 2H, -NH₂) 2.07-2.04 (m, 2H, menthyl), 2.02-1.95 (m, 2H, menthyl), 1.92 (s, 3H, H-14), 1.82 (s, 1H, H-15), 1.75-1.68 (m, 4H, menthyl), 1.56-1.51 (m, 4H, menthyl), 1.16-1.05 (m, 4H, menthyl), 0.95-0.88 (m, 14H, menthyl), 0.80 (d, *J* = 7.0 Hz, 3H), 0.80 (d, *J* = 7.0 Hz, 3H, menthyl) ppm; ¹³C NMR (400MHz, CDCl₃): δ = 170.47, 155.58 (q, 2 J_{CF} = 37.0 Hz, 1C), 148.42, 141.55, 140.09, 130.47, 128.48, 126.02, 125.23 (d, *J* = 11.2 Hz), 124.46, 124.28 (d, *J* = 13.3 Hz), 123.77 (d, *J* = 11.1 Hz), 120.01, 116.53, 116.09 (q, ¹J_{CF} = 288.6 Hz), 75.83 (dd, *J*

= 21.8, 16.1 Hz), 68.63 (d, J = 17.1 Hz), 68.62, 58.50, 46.95, 40.49, 34.34, 31.58, 26.37, 26.33, 23.23 (d, J = 3.5 Hz), 22.15, 21.02 (d, J = 2.8 Hz), 16.09, 13.15 (d, J = 3.4 Hz), 12.55 (d, J = 5.2 Hz) ppm.

***rac-N*-(8-amino-1,7-dimethyl-6*H*,12*H*-5,11-methanodibenzo[*b,f*][1,5]diazocin-2-yl)-2,2,2-trifluoroacetamide (*rac*-36).**

Anhydrous pyridine (63 μ L, 0.77 mmol) was added to a solution of *rac*-20 (364 mg, 1.30 mmol) in anhydrous DCM (204 mL) under inert N₂ atmosphere at rt under stirring. The resulting solution was stirred for 10 minutes before trifluoroacetic anhydride (108 μ L, 0.767 mmol) in anhydrous DCM (1.00 mL) was added drop wise under vigorous stirring over 2.5 h. After the addition, the reaction was left stirring for 16 h at rt under stirring. The DCM was removed *in vacuo* yielding a brown solid.

Purification method: The reaction crude was dissolved in DCM. Celite (4 g) was added to the solution, the resulting mixture was evaporated to dryness *in vacuo*. The resulting solid was added to a dry vacuum column (4x5 cm, CHCl₃:EtOAc gradient from 50% EtOAc to 80% increasing the gradient by 30% every 100 ml, EtOAc:MeOH gradient from 0,1% MeOH to 20% increasing the gradient by 10% every 100ml) Mono-trifluoroacetylated *rac*-36 was isolated as a yellowish solid in 25% yield.

R_f = 0.34 (EtOAc 100%)

(*rac*-37).

To a solution of *rac*-36 (100 mg, 0.265 mmol) in TFA (8 mL), paraformaldehyde (35.5 mg, 1.17 mmol) was added in one portion at rt. The resulting mixture was heated to 100 °C and left stirring for 30 min. The resulting mixture was left cooling down to 70 °C while maintaining stirring for 3.5 h. The mixture was allowed to reach rt before TFA was removed *in vacuo*. Water (4 mL) was added to the resulting slurry and was made alkaline (pH 8-10) by slow addition of NH₃ (aq, 32%). A couple of drops of EtOAc were added to redissolve the precipitate/slurry and the biphasic solution was made alkaline again by slow addition of NH₃ (aq, 32%). The alkaline mixture was extracted with EtOAc (4 x 10 mL). The combination of organic phases were washed with water (10 mL), brine (10 mL), dried over anhydrous MgSO₄, filtered and evaporated *in vacuo*.

A purification method for the crude needs to be developed.

8 References

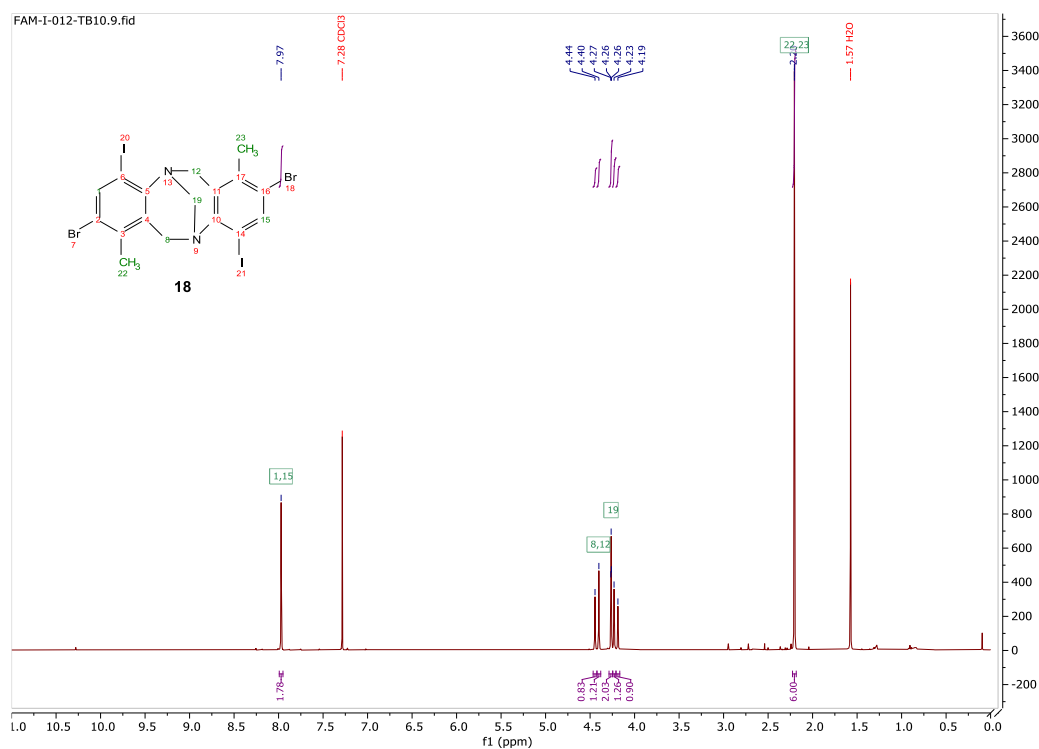
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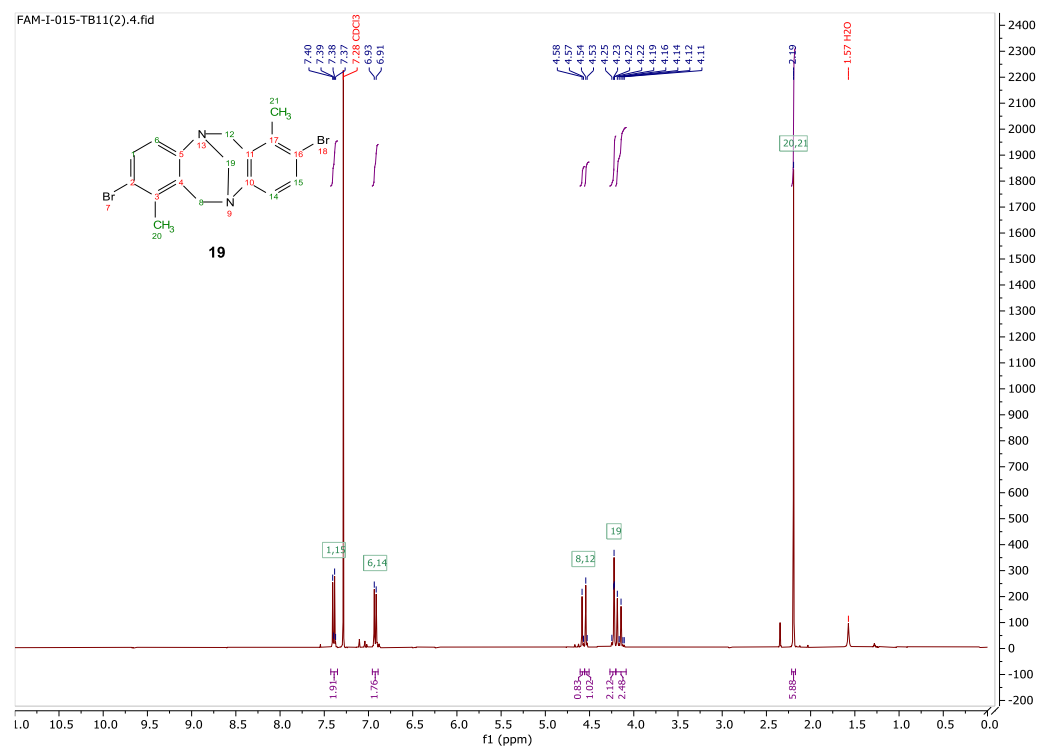
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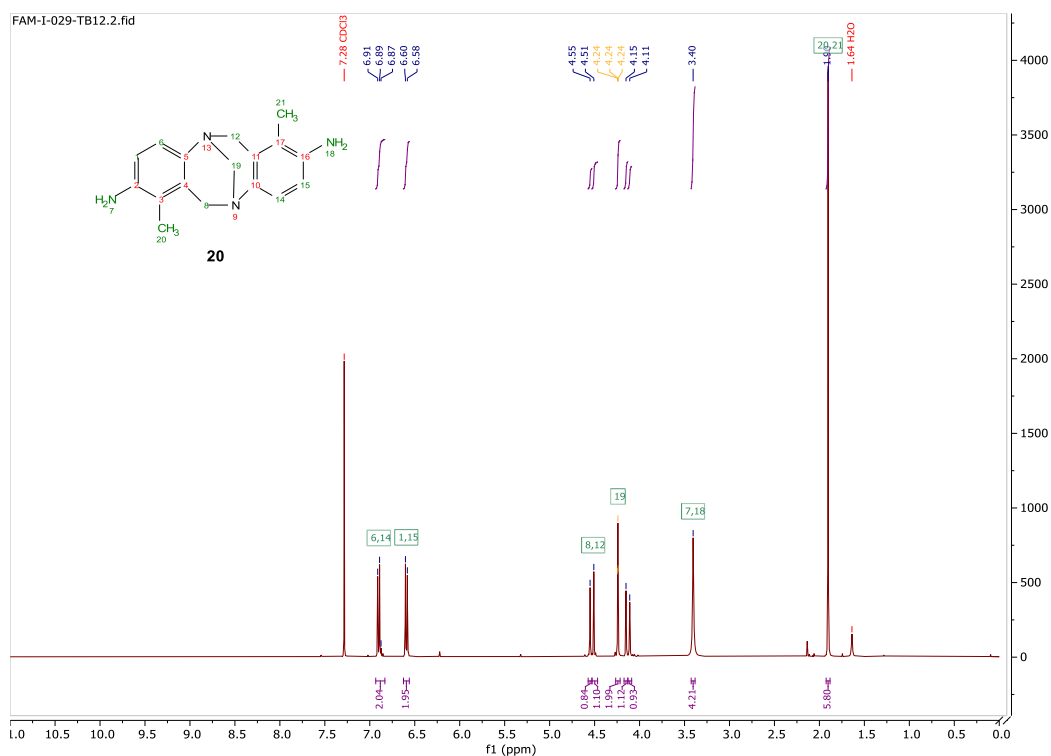
9 Appendix



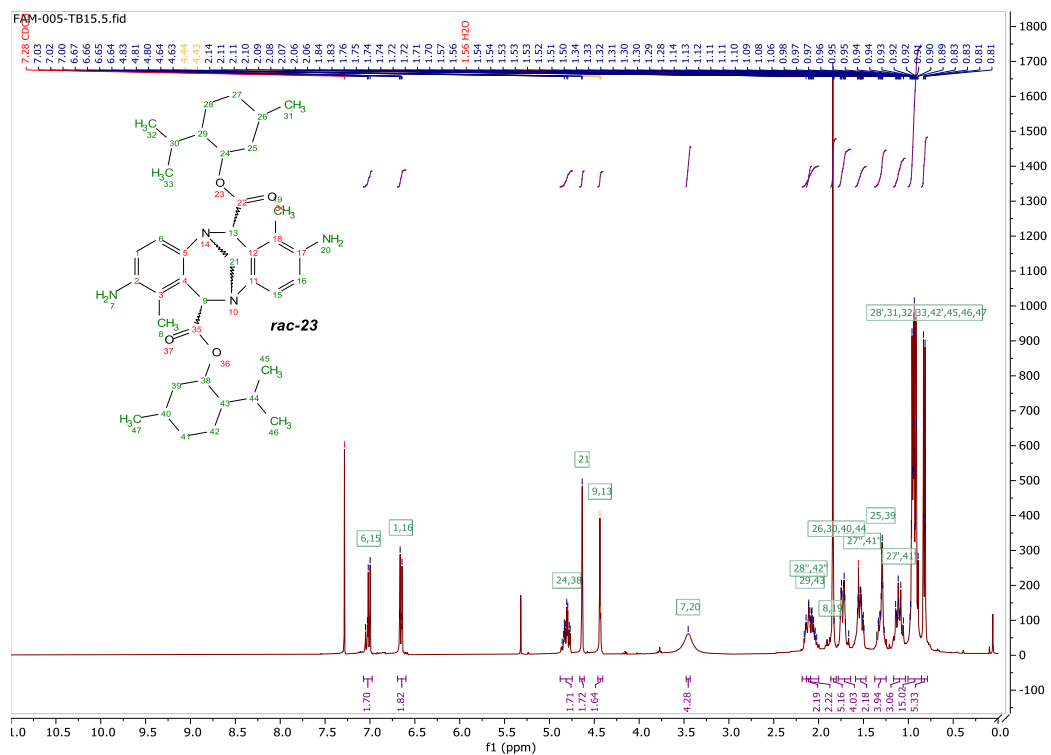
Appendix 1. ¹H NMR of compound 18.



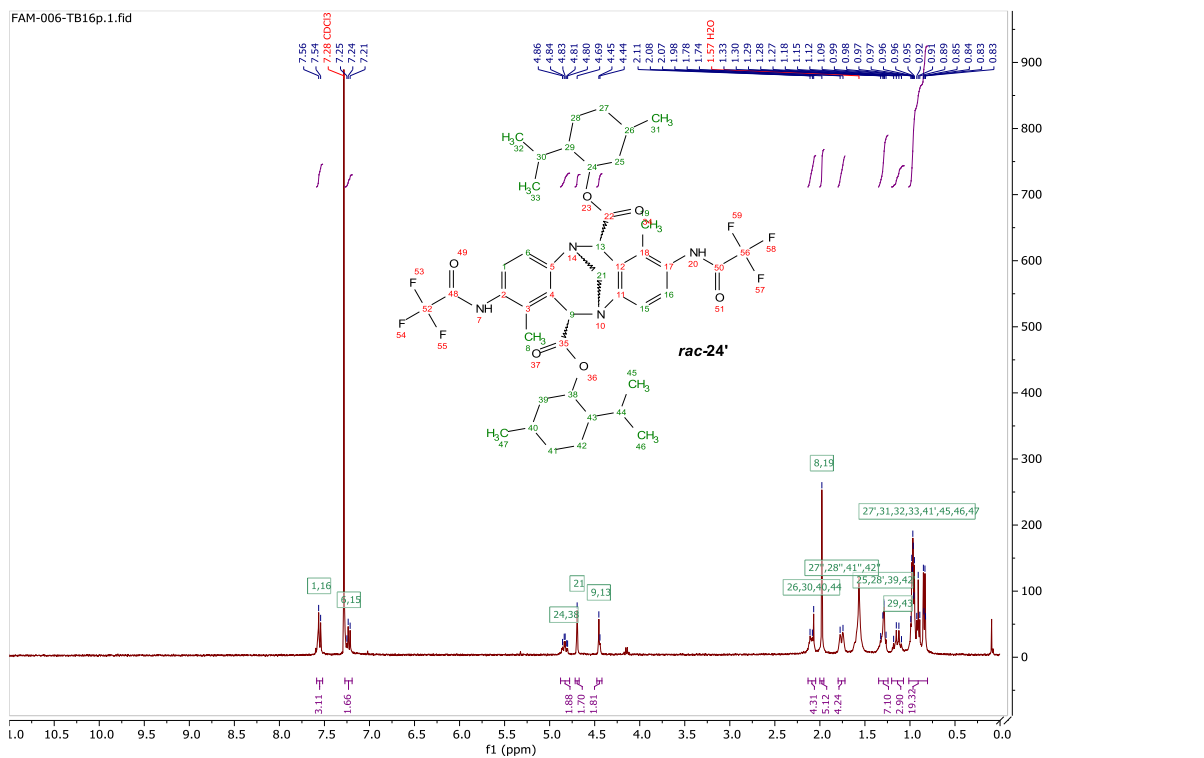
Appendix 2. ¹H NMR of compound 19.



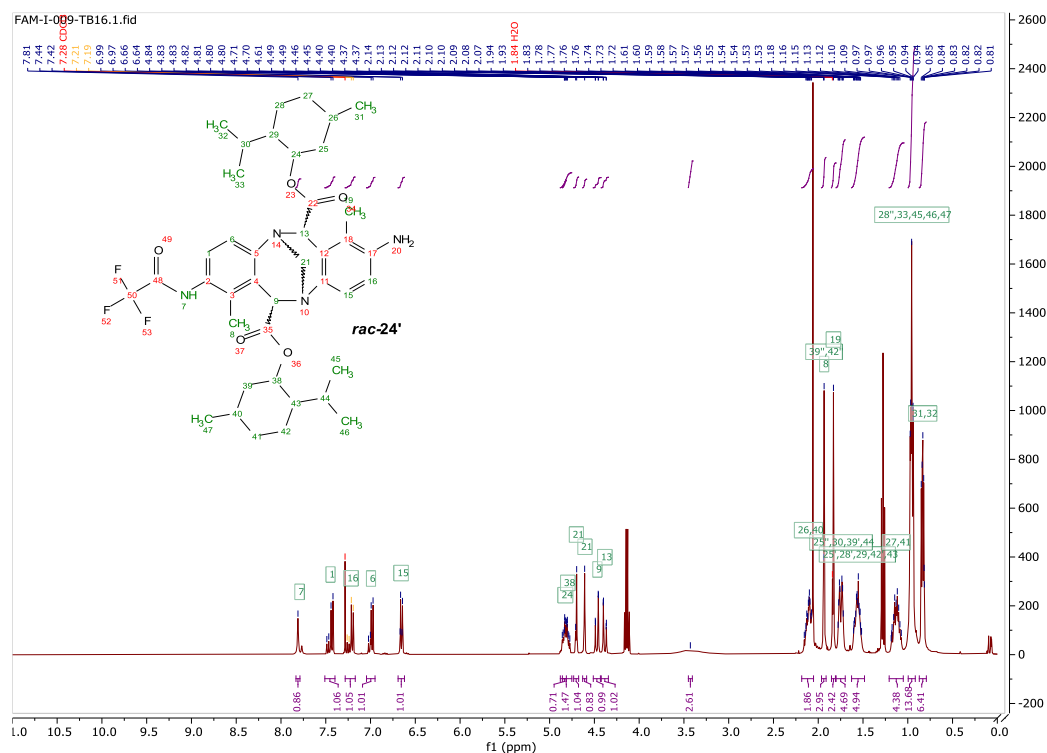
Appendix 3. ¹HNMR of compound **20**.



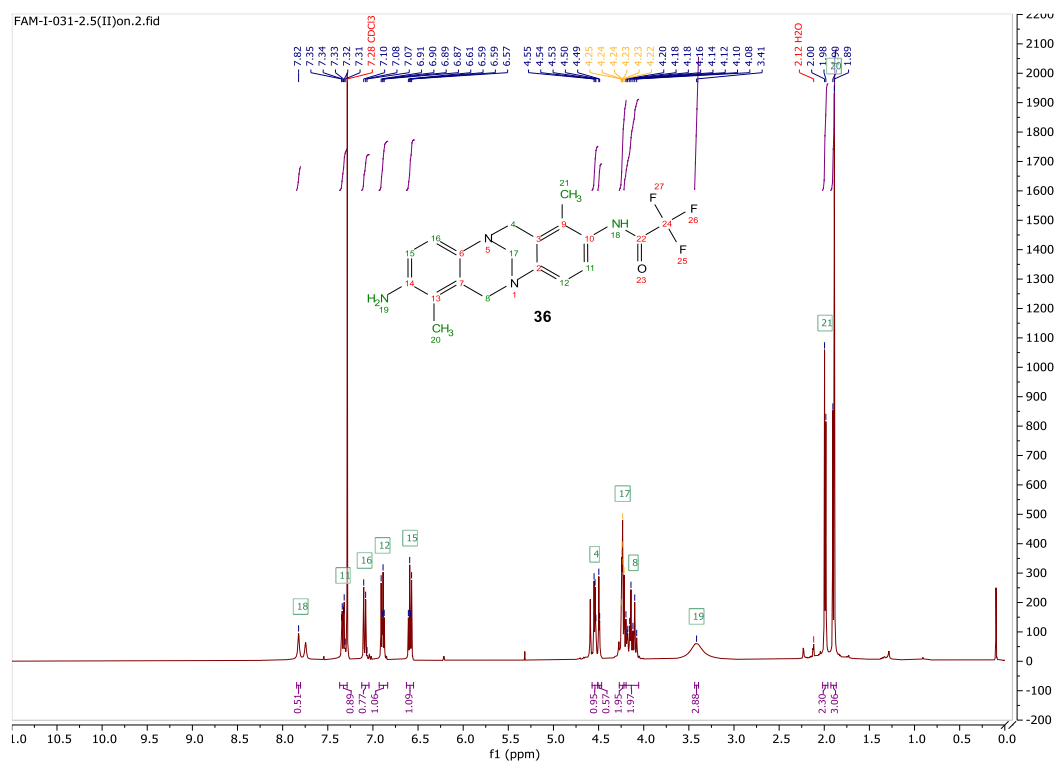
Appendix 4. ¹HNMR of compound **rac-23**.



Appendix 5. ¹HNMR of compound *rac-24'*.



Appendix 5. ¹HNMR of compound *rac-24'*.



Appendix 5. ¹HNMR of compound **36**.