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The Synthesis of Dendrimer-Bound Catalysts and Their Use in MacMillan-Type Reactions

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Dendrimers—highly branched macromolecules—present an attractive option for use as a catalytic framework because of their large size and their availability for functionalization. In addition, the use of organocatalysts to form asymmetric products has become an increasingly studied field in the pharmaceutical industry and cancer research. Organocatalyst terminally functionalized dendrimers present the possibility of both catalytic utility and increased recovery in MacMillan-type asymmetric reactions. Terminal sites of generations 2.0, 3.0, and 4.0 PAMAM dendrimers have been functionalized with the MacMillan group's (2S, 5S)-5-benzyl-2-tert-butyl-3-methyl-imidazolidin-4-one catalyst. These functionalized PAMAM dendrimers were characterized by NMR and MALDI-TOF MS analysis. The organocatalyst functionalized dendrimers show promising yield, enantioselectivity, and recoverability in MacMillan-type organocatalytic reactions.

INTRODUCTION

Organo-catalysts are a classification of catalyst that are comprised of mostly carbon, hydrogen, oxygen, and nitrogen atoms. Organic molecules used for catalysis have a long history, but recent interest in organo-catalysis has been turned to catalysts that can produce enantioselective and diasteroselective (optically active) products. Relatively recently, in the 1970s, a revolutionary reaction called the Hajos-Parrish-Eder-Sauer-Wiechert reaction was carried out by using a naturally occurring chiral reagent, proline, as a catalyst to promote the synthesis of a chiral aldol reaction product.[1] After this asymmetric reaction catalyzed by an organo-catalyst was reported and shown to be extremely enantioselective, the field of organo-catalysis had a huge increase in the amount of attention it drew. This gain in asymmetric catalysis sparked more research into different classes of organocatalyst. During this gain in attention, another influential class of catalysts, imidazolidinone catalysts, were developed by David MacMillan[2] by modifying the naturally occurring chiral phenylalanine. The MacMillan research group at Princeton University in New Jersey currently works to develop different organo-catalysts to aid in a wide range of reactions, some of which are prevalent in the pharmaceutical industry and also utilized in the total synthesis of natural products.[3]

The organo-catalysts developed by the MacMillan group are important for a variety of organic syntheses. These syntheses include high enantioselectivity for various commonly observed asymmetric organocatalytic reactions (diels-alder, aldol addition, and nucleophilic addition) as well as cascade catalysis.[4] The catalysts developed for these reactions produce enantioselective and diastereoselective products, which are of interest to many organizations and research efforts. Most commonly, optically active molecules are extremely important in pharmaceutical applications where different stereoisomers can play very different roles in the body.[5] Because of their wide variety of uses and the procedure used in synthesizing the organo-catalysts, the organo-catalysts developed by MacMillan are relatively expensive to purchase.[6] As of now, the catalysts are difficult to recover and purify for reuse, leading to disposal and therefore making any synthesis involving the catalyst expensive and inefficient.

By modifying how MacMillan's catalysts are introduced into a reaction and building multiple catalytic units onto a large organic framework instead of introducing them into a reaction as discrete molecules, the effective size of the catalytic unit increases. This increase in size leads to more recoverability options for the catalysts, and can make syntheses involving MacMillan's catalysts more efficient since the catalysts can be recovered and reused. This project focuses on using dendrimers as an organic scaffold onto which the catalyst can be synthesized.

Dendrimers are synthetic polymers that have a treelike structure—whose "branches" emanate from a single point called a core. PAMAM (poly(amido amine)) dendrimers are a common class of dendrimer used in many different materials science and biotechnology applications due to their physical properties. Dendrimers can be synthesized with different cores and terminal end groups. The synthetic route being taken in this project utilizes specific generations of PAMAM dendrimers, which have an ethylene diamine core, branches that contain amide groups, and terminate in amine groups (Figure 1). PAMAM dendrimers are symmetrical and adopt a relatively spherical shape. The terminal amine ends of the "branches," or end groups, are relatively monodisperse over the molecule's spherical surface. These amine groups are reactive as nucleophiles and therefore present a unique opportunity as a site for functionalization (addition of molecules to the amine end groups) of the PAMAM end groups. PAMAM dendrimer solubility also typically mimics the solubility of their terminal groups, which is important for keeping reaction conditions relatively consistent when investigating the use of dendrimers in catalysis. PAMAM dendrimers can be purchased or synthesized in multiple generations as well—Generations 2.0, 3.0, and 4.0 PAMAM dendrimers will be used for this project, which have 16, 32, and 64 amine-

terminated end groups available for functionalization with the desired organo-catalyst, respectively.[7]

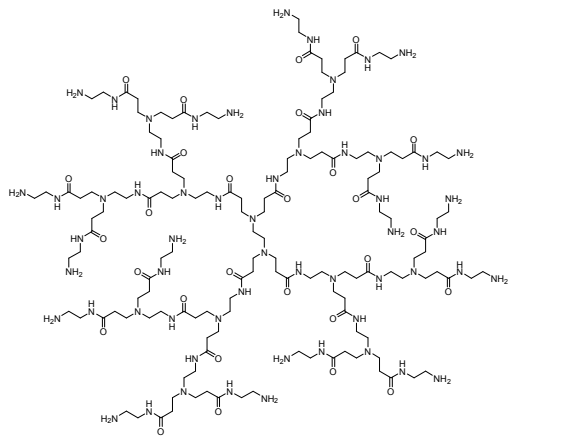


Figure 1: A Generation 2.0 PAMAM Dendrimer

Dendritic catalysis has become an increasingly important and studied field. Three structural motifs that utilize the dendritic macromolecule as a catalyst delivery system have been investigated (Figure 2). In the first motif, the core of the dendrimer is the single catalytic unit in dendrimer **1**.[8] The second, the "dendritic box" motif **2**, utilizes pockets inside the dendrimer formed by the placement of its branches to capture and deliver the catalytic unit.[9] The third motif **3**, incorporating the largest number of catalytic units, covalently binds catalytic units to the terminal ends of the dendrimer.[10-14] Using the motif in **3** and covalently binding the catalyst to the amine end groups of the PAMAM dendrimer framework is ideal for our method of dendritic catalysis because it allows separation of the catalyst with any method of separation that could be utilized with dendrimers or other similar macromolecules.

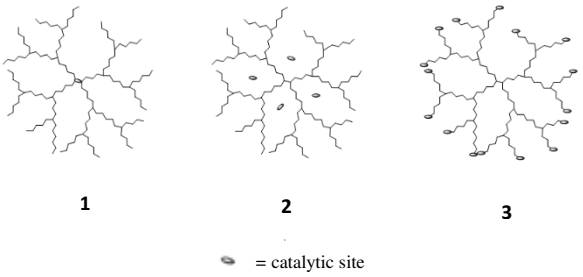


Figure 2: Varying Dendrimer Catalyst Delivery Motifs

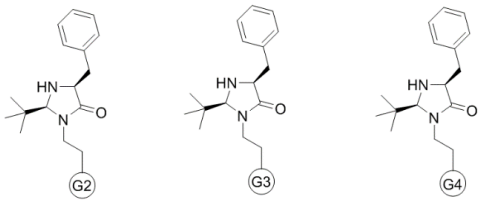
PAMAM dendrimers were chosen in particular for this work due to their properties for an ideal platform for catalysis. Their spherical shape, solubility properties, organic structure, and monodisperse end groups make them attractive as a framework for organocatalysis. The spherical shape and monodisperse end groups of a PAMAM dendrimer are an optimal way to deliver a catalyst to a system because the catalytic site is on the surface of the molecule, leading to ready accessibility to reagents for catalysis. Solubility properties of PAMAM dendrimers typically mimic those of the end groups, especially in higher generations. This provides the opportunity to keep solvent reaction conditions consistent between dendrimer-bound catalysis and catalysis using free, unbound catalysts. The organic structure of the PAMAM catalysts provides the option of covalently binding molecules to the terminal end-groups.

By exploiting the reactive amino end groups, functionalizing the PAMAM dendrimers with MacMillan's (2S, 5S)-5-(1-benzyl-1H-indol-3-yl)-2-tert-butyl-3-methyl-imidazolidin-4-one catalyst (therefore increasing the effective size of the catalytic delivery framework) provides a route for catalyst purification. The size of the organo-catalyst functionalized dendrimers theoretically introduce the opportunity for recovery and reuse by filtration, dialysis, or size exclusion chromatography, which makes the MacMillan group's procedures more economically feasible and environmentally friendly. Recovery and reuse of this catalyst could impact the fields of green chemistry as well as the pharmaceutical industry.

DISCUSSION

Catalyst Functionalization of Dendrimers

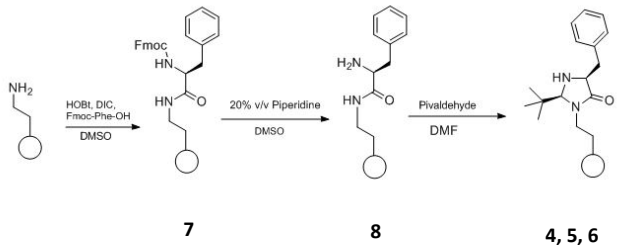
A set of three PAMAM dendrimers **4-6** were functionalized with a MacMillan-type imidazolidinone catalyst known to perform enantioselective catalysis (Figure 3). Although the dendrimer bound catalysts illustrated only show one catalytic unit per dendrimer, these schematics symbolize fully mono-functionalized dendrimers with the number of end groups functionalized respective to their generation.



4 **5** **6**

Figure 3: Dendrimer generations 2.0-4.0 Functionalized with MacMillan's (2S, 5S)-5-(1-benzyl-1H-indol-3-yl)—tert-butyl-3-methyl-imidazolidin-4-one catalyst.

Dendrimers **4-6** were functionalized using an approach adapted from a related functionalization by Koskinen and coworkers[15] in order to develop the synthetic scheme to produce the desired imidazolidinone catalyst (Scheme 1). Coupling of *N*- fluorenylmethyloxycarbonyl (fmoc) protected L-phenylalanine with the free amine surface of each generation of PAMAM dendrimer was completed under hydroxybenzotriazole and diisopropyl-carbodiimide conditions. Deprotection of the resulting material **7** was accomplished with piperidine to reveal the phenylalanine-functionalized dendrimer **8.** Treating the dendrimer with pivaldehyde in dimethylformamide resulted in the mono-functionalized dendrimers **4-6.**



Scheme 1

The method that was adapted for the PAMAM dendrimer functionalization in Scheme 1 was not initially intended or optimized for varying generations of PAMAM dendrimer. Work on this project also included optimization of reagents, reaction conditions, and purification methods for the synthesis in Scheme 1. Reagent amounts were scaled appropriately for the number of amino end groups on the dendrimer, instead of a linear molar ratio. Reaction times were increased in order to allow reagents to navigate through the dendritic framework and react with terminal amino end groups. The pH of reactions were adjusted to be compatible with PAMAM dendrimer structure (pH 4-9). The first two steps in Scheme 1 were found to be able to be completed in dimethylsulfoxide (DMSO) instead of dimethylformamide (DMF) in order to minimize hazardous chemical exposure and wear on lab instruments.[16,17] Pi-stacking of excess fmoc-phenyalanine with dendrimer-functionalized fmoc-phenylalanine inside the dendritic framework required that steps one and two in Scheme 1 were completed as a one-pot synthesis in order to drive all fmoc from the dendritic framework at once with an excess of piperidine in the deprotection step. This one-pot synthesis streamlined separation of fmoc from the phenylalanine-functionalized dendrimer **8.** Completing steps one and two in Scheme 1 as a one-pot synthesis also maximized recovery of **8** due to no loss of **7** in an unnecessary and time consuming purification step.

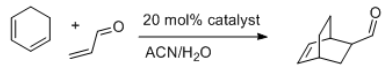
Functionalized dendrimer **8,** as well as functionalized dendrimers **4,5,6** were purified via dialysis in DMSO. Since DMSO was found to be trapped inside of the dendritic material, an additional step to drive solvent from the material was needed. When DMSO from dialysis is being removed via lyophilization, millipore water is added to the mixture to drive DMSO from the internal dendritic framework as well as creating diluted solution of DMSO and water that aides in the removal of solvent from the dendrimer material, as water has a higher vapor pressure (17.54 mmHg)[18] than DMSO (0.42 mmHg)[19]. This results in the DMSO almost completely leaving the internal dendritic framework by lyophilization to provide more accurate yields by weight as well as not interfering with any characterization methods. Additionally, in the process of developing the parameters shown in Scheme 1, it was found that cyclization of the catalyst in step 3 of Scheme 1 was sometimes not completely successful due to excess solvent, not enough reagent, or not long enough of a reaction time. In these cases, it was found that if the material was re-submitted to the reaction conditions in step 3 of Scheme 1, additional

cyclization would occur, further functionalizing the dendrimer with the desired catalyst. During the course of this work the parameters for step 3 of Scheme 1, including amounts of solvent, reagent, and reaction time, were optimized. The methods used to cyclize the catalyst are now efficient enough to produce quantitative yields of dendrimer functionalized with cyclized catalyst in one ring-closing reaction. For the purification of the resultant fully functionalized dendrimer-bound catalyst, DMF must be evaporated via lyophilization before submitting the material to dialysis, since DMF will degrade the dialysis tubing (made of regenerated cellulose) that is compatible with DMSO.

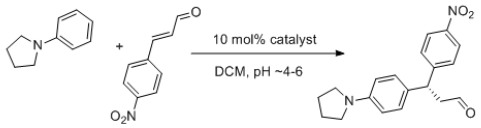
The resultant new dendrimers **4-6** were characterized using ^{1}H NMR and ^{13}C NMR spectroscopy techniques as well as matrix assisted laser desorption ionization time-of-flight (MALDI-TOF) mass spectrometry. Multiple matrices used for MALDI-TOF-MS were investigated in order to optimize dendrimer characterization, including *trans*-3-indoleacrylic acid (IAA) and 2,5-dihydroxybenzoic acid (DHB). Optimal MALDI-TOF-MS results were obtained using DHB as a matrix.

Catalytic Activity of Catalyst-Functionalized Dendrimers

Functionalized PAMAM dendrimers **4-6** were evaluated for their ability to promote enantioselective catalysis using selected reactions catalyzed by the MacMillan-type imidazolidinone catalyst that was built onto the dendritic framework (Schemes 2, 3).[20,21]



Scheme 2



Scheme 3

The reactions in Scheme 2 and 3 were chosen to demonstrate the catalytic utility of the dendrimer because they were within the range of what is compatible with the framework of a PAMAM dendrimer. The reactions in Schemes 2 and 3 were run at room temperature. This is advantageous as low temperatures cannot be supported for more than 8 hours in our lab, and temperatures over 30°C can degrade the framework of the dendrimer. The reactions chosen were also performed in a pH range that was adequate for the dendrimer framework, as the framework can also degrade over pH of 9, and at pHs lower than 3, the amine groups inside the dendrimer are ionized and the dendrimer can precipitate out of solution as a salt. Since these are screening reactions, the reactions in Scheme 2 and 3 were also chosen due to their readily available or relatively inexpensive reagents.

In order to gather sufficient evidence to support our dendrimer-bound catalyst's utility, reactions

shown in Scheme 2 and 3 were run under only acidic conditions (no catalyst), the original conditions published (free catalyst), and with the dendrimer-bound catalysts **4, 5, 6.** The time until reaction completion, percent yield, optical activity, and catalyst recovery were compared to values reported by the MacMillan group. Enantiomeric excess was determined via polarimetry in chloroform.

Solvent Studies and Other Notable Observations

Although dendrimer solubility generally mimics the solubility of their end groups, lower generations of PAMAM dendrimers can exhibit different solubility properties in certain organic solvents because of their less spherical shape (compared to higher generations) and exposed internal structure. A series of control reactions for the aniline addition reaction in different solvents were used in order to determine catalyst functionality in differing solvent environments in order to broaden possible reaction conditions.

During these solvent studies, multiple things were discovered about the practical process of the procedures in Scheme 2 and 3 with dendrimer-bound material. In the reaction shown in Scheme 3, an amber vial (preferably subsequently covered in tinfoil) must be used in order to prevent light degradation of the 1-phenylpyrrolidine starting material. In the reaction shown in Scheme 3, there is also a cooling step to -10°C. When running the reaction in Scheme 3 with DMSO as a solvent, the cooling step cannot be employed because of the freezing point of DMSO at 19°C.[22] Though the reaction cannot be cooled, it was found that it had no noticeable effect on the yield or enantioselectivity of the reaction. If the solvent used in the nucleophilic addition reaction is DCM, parafilm should not be used in order to prevent DCM from leeching the hard-to-separate parafilm into the reaction mixture.

Since the nucleophilic addition reaction in Scheme 3 is run in acidic conditions, there is also a reaction acidifying step. The amount of acid in the original reaction is the amount needed to acidify all of the catalyst material. However, in a system using the dendrimer-bound catalyst, some of the internal framework of the dendrimer is protonated before the catalytic groups on the surface of the dendrimer. It was found that the amount of acid needed to be increased in a dendrimer-bound catalyst system in order to protonate the catalytic groups and run the reaction successfully. However, because of the chance of the dendrimer framework being completely protonated and precipitating out of solution, the reaction must be kept above a pH of 3.

Dialysis was used to purify the dendrimer material from the reaction mixture, since the dendrimer material should stay in the dialysis tubing as the reaction mixture diffuses outside of the dialysis tubing. Dialysis in DMSO was initially tried, but the lyophilization procedure from evaporating DMSO was time consuming.

Dialysis in DCM was also attempted, but it was found that DCM in large volumes degraded the dialysis tubing and leeched cellulose into the reaction mixture. Removing the cellulose from the reaction mixture added an extra chromatography purification step (in addition to the chromatographic method to purify product from reagents), so DCM was discarded as a potential solvent for dialysis and the use of DMSO had evidence of being more efficient than DCM in these conditions.

Purification of the desired products from the reactants was performed via silica gel chromatography. It was found that chromatography using a Chromatotron (a centrifugal thin-layer silica gel purification system)[23] with a hand-held UV lamp was more efficient, used less solvent, and provided better separation and yields than classic flash or gravity chromatography with silica gel.

RESULTS

Three generations of dendrimer-bound (2S, 5S)-5-benzyl-2-*tert*-butyl-3-methylimidazolidin-4-one were successfully synthesized with good functionalization yield. Phenylalanine functionalization was completed in near quantitiative yield (Table 1, Steps 1 and 2 to produce **8** in Scheme 1). Cyclization into the complete catalyst (Step 3 to produce **4,5,6** in Scheme 1) was completed in high yield when characterized by NMR, but MALDI-TOF-MS data shows a lower yield of cyclization (Table 1).

Table 1: Percent functionalization for 8 and 4,5,6 by both NMR and MALDI-TOF-MS

GENERATION	PAMAM 8		PAMAM 4,5,6	
	MALDI	NMR	MALDI	NMR
G(2.0)	Quant.	Quant.	26%	86%
G(3.0)	91%	Quant.	32%	90%
G(4.0)	97%	Quant.	26%	93%

Because of the internal consistency of percent functionalization found via both MALDI-TOF-MS and NMR spectroscopy, respectively, and due to the nature of the high-powered laser used in MALDI-TOF-MS, it is theorized that the laser fragments the fragile fully cyclized catalyst functionalized dendrimer. Further investigation is currently being conducted to determine another method of characterization to determine the mass of the catalyst functionalized dendrimers to confirm this theory.

Two reactions published by MacMillan using free (2S, 5S)-5-benzyl-2-*tert*-butyl-3-methylimidazolidin-4-one were investigated by substituting free catalyst with dendrimer-bound catalyst, and evidence shows that the dendrimer-bound catalyst both catalyzes these reactions and produces the same rotation in chirality in the desired product. (Table 2).

Table 2: Comparison of Free Catalyst and Dendrimer-Bound Catalyst Activity

		FREE CATALYST	DENDRIMER-BOUND CATALYST
ANILINE ADDITION (DCM)	Yield	89%	32%
	Optical Rotation	-2.77°	-0.4° (observed rotation)
DIELS ALDER (ACN/WATER)	Yield	4%	24%
	Optical Rotation	-536°	-460°

Since both control and on-dendrimer catalyzed reactions ran to completion via NMR monitoring, the discrepancy in percent yield is attributed to purification error. The optical rotation for all products produced by dendrimer-bound catalyst is concurrent with the chirality observed with free catalyst, which is promising for future work because the optical rotation provides evidence the dendrimer-bound catalyst works with similar enantioselectivity to the free catalyst.

Attempts to recover the dendrimer-bound catalyst were initially successful but are currently being optimized. Purification of the dendrimer-bound catalyst by filtration was initially seemed promising because of limited dendrimer solubility in acetonitrile and water, but NMR analysis showed high levels of starting reagents trapped inside the dendrimer framework after washing of the filtrate. Dialysis against DMSO using regenerated cellulose dialysis tubing with a MWCO of 3500 kDa proved a more efficient way to purify dendrimer-bound catalyst. (Table 3).

Table 3: Recovery of dendrimer-bound catalyst using dialysis

TRIAL NUMBER	GENERATION	PERCENT RECOVERY
1	G (3.0)	14
2	G (3.0)	6
3	G (3.0)	43
4	G (2.0)	28

There has not yet been any evidence of a correlation between dendrimer generation used and yield or recovery.

CONCLUSIONS

Three generations (2.0, 3.0, and 4.0) of dendrimer-bound (2S, 5S)-5-benzyl-2-*tert*-butyl-3-methylimidazolidin-4-one catalyst have been successfully synthesized. The dendrimer-bound catalysts demonstrate similar yield and optical activity to free (2S, 5S)-5-benzyl-2-*tert*-butyl-3-methylimidazolidin-4-one catalyst as reported by MacMillan. The dendrimer-bound catalysts also demonstrate higher potential for recovery of catalyst than free catalyst. Recovery methods are currently being optimized, most promisingly through dialysis techniques. The efficiency of the dendrimer-bound catalyst after a number of uses and recoveries is also being investigated. Future work will include optimizing catalytic conditions and developing orthogonal catalysts for multistep or cascade catalysis, as well as attempting different catalytic delivery and recovery methods through the use of linking the

dendrimer-bound catalyst to a gold surface for solid-support catalysis.

ACKNOWLEGEMENT

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EXPERIMENTALS

Generation 2.0 PAMAM-based nitrogen-linked phenylalanine dendrimer (8):

An aqueous solution of amine-terminated Starburst PAMAM dendrimer (Generation 2.0, 12.57% (w/w)) was lyophilized. The resulting residue (124.8 mg, 0.0383 mmol, 1 eq) was dissolved in DMSO (1.0 mL). In a separate scintillation vial, a solution of 9-fluorenylmethoxycarbonyl-L-phenylalanine (0.9442 g, 2.43 mmol, 64 eq) and hydroxybenzotriazole (0.3375 g, 2.49 mmol, 64 eq) in DMSO (4.0 mL) was added to a solution of N,N'-diisopropylcarboimide (382 uL, 2.43 mmol, 64 eq) in DMSO (0.25 mL) and allowed to stir for five minutes. The two solutions were combined and stirred (2.5 h) to form **7**. This product was dialysed against DMSO (MW cutoff 1 kDa). The solution was lyophilized to give **7**. This product was used without further purification.

Resulting crude product **7** was deprotected in a 20% (v/v) solution of piperidine/ DMSO by dissolving in DMSO (7.0 mL) and adding piperidine (1.75 mL). After 110 minutes of stirring, the reaction mixture was dialysed against DMSO (MW cutoff 1 kDa). The solution was lyophilized using water to drive out DMSO to give title compound (**8**) as a fluffy, white solid (136.6 mg). ^{1}H NMR (500MHz, CD_3OD) δ (ppm) 2.31 (m, 61H, PAMAM-x), 2.54 (m, 28H, PAMAM-x), 2.76 (m, 61H, PAMAM-x), 2.97, 3.01 (m, 16H, Ph*CH*$_2$CH), 3.21 (m, 96H, PAMAM-x), 3.50 (m, 16H, PhCH$_2$C*H*), 7.20, 7.28 (m, 80H, Ph) ppm; ^{13}C NMR (500MHz, CD_3OD) δ 34.7, 38.6, 39.9, 40.4, 42.5, 51.1, 53.4, 57.9, 127.9, 129.7, 130.5, 139.0, 175.1, 176.9 ppm; MALDI-TOF-MS (pos) 5628.

Generation 2.0 PAMAM-based nitrogen-linked (2S,5S)-(-)-2-*tert*-butyl-3-methyl-5-benzyl-4-imidazolidinone dendrimer (4):

Phenylalanine functionalized G (2.0) PAMAM (**8**) (126.6 mg, 0.2256 mmol, 1 eq) was dissolved in dimethylformamide (1.5 mL) and pivaldehyde was added (490 uL, 4.51 mmol, 200 eq). N, N'-diisopropylethylamine was added until the reaction mixture reached a pH of 9. The reaction mixture was stirred for approximately 40 hours. This product was lyophilized to remove DMF, re-dissolved in DMSO and dialysed against DMSO (MW cutoff 1 kDa). The solution was lyophilized using water to drive out DMSO to give title compound **4** as a pale yellow glassy solid (89.2 mg). $[\alpha]_D$= -131.5; ^{1}H NMR (500MHz, CD_3OD) δ 0.85 (s, 123H, *tert*-butyl) 2.31 (m, 60H, PAMAM-x) 2.52 (m, 28H, PAMAM-x), 2.74 (m, 61H, PAMAM-x) 3.21 (m, 96H, PAMAM-x) 3.65, 3.69 (dd, 16H, PhCH$_2$C*H*) 6.88, 7.06, 7.21 (m, 80H, Ph) ppm; ^{13}C NMR (500MHz, CD_3OD) δ (ppm) 27.2, 34.8, 37.3, 38.7, 40.1, 40.5, 41.8, 42.6, 51.2, 53.6, 58.0, 76.1, 127.7, 129.4, 129.7, 130.6, 131.2, 138.9, 174.6,

174.7, 175.1, 175.5, 176.5, 177.0 ppm; MALDI-TOF-MS (pos) 6118.

Generation 3.0 PAMAM-based nitrogen-linked phenylalanine dendrimer (8):

An aqueous solution of amine-terminated Starburst PAMAM dendrimer (Generation 3.0, 7.01% (w/w)) was lyophilized. The resulting residue (116.8 mg, 0.0169 mmol, 1 eq) was dissolved in DMSO (2.0 mL). In a separate scintillation vial, a solution of 9-fluorenylmethoxycarbonyl-L-phenylalanine (0.8398 g, 2.17 mmol, 128 eq) and hydroxybenzotriazole (0.2950 g, 2.18 mmol, 128 eq) in DMSO (3.0 mL) was added to a solution of N,N'-diisopropylcarboimide (340 uL, 2.18 mmol, 128 eq) in DMSO (1.0 mL) and allowed to stir for five minutes. The two solutions were combined and stirred (2.5 h) to form **7**. This product was dialysed against DMSO (MW cutoff 3.5 kDa). The solution was lyophilized to give **7**. This product was used without further purification.

Resulting crude product **7** was deprotected in a 20% (v/v) solution of piperidine/ DMSO by dissolving in DMSO (5.5 mL) and adding piperidine (1.4 mL). After 60 minutes of stirring, the reaction mixture was dialysed against DMSO (MW cutoff 3.5 kDa). The solution was lyophilized using water to drive out DMSO to give title compound **8** as a fluffy, white solid (122.2 mg). ^{1}H NMR (300MHz, CD_3OD) δ (ppm) 2.33 (m, 124H, PAMAM-x), 2.56 (m, 57H, PAMAM-x), 2.77 (m, 119H, PAMAM-x), 2.96 (m, 61H, PhCH_2CH), 3.21 (m, 182H, PAMAM-x), 3.51 (m, 32H, PhCH$_2$C*H*), 7.21, 7.28 (m, 169H, Ph); ^{13}C NMR (500MHz, CD_3OD) δ (ppm) 34.8, 28.6, 40.0, 40.4, 42.5, 128.0, 129.6, 130.6, 139.1, 174.6, 175.1, 176.9 ppm; MALDI-TOF-MS (pos) 10606.

Generation 3.0 PAMAM-based nitrogen-linked (2S,5S)-(-)-2-*tert*-butyl-3-methyl-5-benzyl-4-imidazolidinone dendrimer (5):

Phenylalanine functionalized G (3.0) PAMAM **8** (122.2 mg, 0.012 mmol, 1 eq) was dissolved in dimethylformamide (2.0 mL) and pivaldehyde was added (228 uL, 2.05 mmol, 200 eq). N, N'-diisopropylethylamine was added until the pH of the reaction mixture reached 9. The reaction mixture was stirred for approximately 40 hours. This product was lyophilized to remove DMF, re-dissolved in DMSO and dialysed against DMSO (MW cutoff 3.5 kDa). The solution was lyophilized using water to drive out DMSO to give title compound **5** as a pale yellow glassy solid (126.1 mg). ^{1}H NMR (500MHz, CD_3OD) δ 0.89 (s, 252H, *tert*-butyl), 2.35 (m, 120H, PAMAM-x), 2.57 (m, 56H, PAMAM-x) 2.78 (m, 121H, PAMAM-x), 3.25 (m, 185H, PAMAM-x), 3.71, 3.74 (dd, 31H, PhCH$_2$C*H*), 6.93, 7.09, 7.24 (m, 163H, Ph) ppm; ^{13}C NMR (500MHz, CD_3OD) δ ppm 25.1, 25.8, 33.4, 35.9, 37.3, 38.7, 39.1, 40.4, 41.2, 49.8, 52.2, 56.6, 74.6, 126.3, 128.0, 128.3, 129.2, 129.8, 137.5, 173.7, 174.1, 175.1; (500MHz,

CD_3OD) δ ppm; MALDI-TOF-MS (pos) 11372.

Generation 4.0 PAMAM-based nitrogen-linked phenylalanine dendrimer (8):

An aqueous solution of amine-terminated Starburst PAMAM dendrimer (Generation 4.0, 13.12% (w/w)) was lyophilized. The resulting residue (123.7 mg, 0.0087 mmol, 1 eq) was dissolved in DMSO (2.0 mL). In a separate scintillation vial, a solution of 9-fluorenylmethoxycarbonyl-L-phenylalanine (0.8622 g, 2.23 mmol, 256 eq) and hydroxybenzotriazole (0.3067 g, 2.27 mmol, 256 eq) in DMSO (4.0 mL) was added to a solution of N,N'-diisopropylcarboimide (346 uL, 2.22 mmol, 256 eq) in DMSO (0.5 mL) and allowed to stir for five minutes. The two solutions were combined and stirred (2.5 h) to form **7.** This product was dialysed against DMSO (MW cutoff 3.5 kDa). The solution was lyophilized to give **7.** This product was used without further purification.

Resulting crude product **7** was deprotected in a 20% (v/v) solution of piperidine/ DMSO by dissolving in DMSO (7.0 mL) and adding piperidine (1.75 mL). After 90 minutes of stirring, the reaction mixture was dialysed against DMSO (MW cutoff 3.5 kDa). The solution was lyophilized using water to drive out DMSO to give title compound **8** as a fluffy, white solid (161.4 mg). ^{1}H NMR (500MHz, CD_3OD) δ (ppm) 2.29 (m, 240H, PAMAM-x), 2.52 (m, 112H, PAMAM-x), 2.73 (m, 243H, PAMAM-x), 2.98 (m, 63H, Ph*CH*$_2$CH), 3.21 (m, 368H, PAMAM-x), 3.51 (64H, m, PhCH$_2$C*H*), 7.19,7.25 (m, 319H, Ph) ppm; ^{13}C NMR (500MHz, CD_3OD) δ (ppm) 34.8, 38.7, 40.0, 40.5, 42.6, 51.3, 53.6, 57.8, 127.9, 129.6, 130.6, 139.0, 175.2, 176.8 ppm; MALDI-TOF-MS (pos) 21164.

Generation 4.0 PAMAM-based nitrogen-linked (2S,5S)-(-)-2-*tert*-butyl-3-methyl-5-benzyl-4-imidazolidinone dendrimer (6):

Phenylalanine functionalized G (4.0) PAMAM **8** (151.8 mg, 0.0066 mmol, 1 eq) was dissolved in dimethylformamide (1.5 mL) and pivaldehyde was added (143 uL, 1.32 mmol, 200 eq). N, N'-diisopropylethylamine was added until the pH of the reaction mixture reached 9. The reaction mixture was stirred for approximately 40 hours. This product was lyophilized to remove DMF, re-dissolved in DMSO and dialysed against DMSO (MW cutoff 3.5 kDa). The solution was lyophilized using water to drive out DMSO to give title compound **6** as a pale yellow glassy solid (99.3 mg). $[\alpha]_D$= -47.4; ^{1}H NMR (500MHz, CD_3OD) δ 0.89 (s, 521H, *tert*-butyl), 2.35 (m, 239H, PAMAM-x), 2.56 (m, 119H, PAMAM-x), 2.78 (m, 239H, PAMAM-x), 3.25 (m, 366H, PAMAM-x), 3.71 (dd, 62, PhCH$_2$C*H*), 6.92, 7.08, 7.22 (m, 320H, Ph) ppm; ^{13}C NMR (500MHz, CD_3OD) δ 25.4, 26.4, 27.4, 35.0, 37.4, 40.2, 40.3, 40.7, 42.0, 51.3, 53.8, 58.1, 76.2, 127.9, 129.6, 129.9, 131.4, 139.0, 139.3, 174.7,

174.8, 175.2, 176.6, 177.0 ppm; MALDI-TOF-MS (pos) 22700.

Acid-Catalyzed in Dichloromethane 3-(4-Nitrophenyl)-3-(4-pyrolidin-1-yl-phenyl)-propionaldehyde

To a 5 mL amber scintillation vial equipped with a magnetic stir bar was added 4-nitrocinnamaldehyde (13.7 mg, 0.077 mmol, 1.0 equiv) dichloromethane (0.2 mL), and HCl (as a 4N solution in 1,4-dioxane, 5 uL). The solution was cooled to -10˚C in a salt/ice bath before addition of 1-phenylpyrrolidine (35.1 uL, 0.244 mmol, 3.0 equiv). After 48 h, the crude reaction mixture was analyzed by NMR and product was found to be formed in a 4-nitrocinnamaldehyde:product ratio of 1:1.2, providing evidence that any additional product formed after the 16 d mark in the on-dendrimer catalyst is due to catalytic activity, and not acid catalysis.

Dendrimer-Bound Catalyst in Dichloromethane (S)-3-(4-Nitrophenyl)-3-(4-pyrolidin-1-yl-phenyl)-propionaldehyde

To a 5 mL amber scintillation vial equipped with a magnetic stir bar was added G(2.0) dendrimer-bound (2S, 5S)-5-benzyl-2-*tert*-butyl-3-methylimidazolidin-4-one (6.5 mg, 9.7E-4 mmol, 0.20 equiv catalyst), dichloromethane (0.5 mL), HCl to acidify the solution to pH ~4 (as a 4N solution in 1,4-dioxane, 20 uL), and 4-nitrocinnamaldehyde (14.5 mg, 0.082 mmol, 1.0 equiv). The solution was cooled to -10˚C in a salt/ice bath before addition of 1-phenylpyrrolidine (34.3 uL, 0.234 mmol, 3.0 equiv). After 16 d, the reaction mixture was transferred into dialysis tubing (MWCO 3500) and dialyzed against DCM. The DCM outside the dialysis tubing was concentrated in vacuo and subjected to silica gel chromatography via chromatotron. Gradient elution with 25-50% EtOAc in hexanes followed by concentration in vacuo afforded the product as a bright orange oil in 32.4% yield (8.6 mg, 0.027 mmol); $[\alpha]_{obs}$= -0.4

Dendrimer-Bound Catalyst in Dimethylsulfoxide (S)-3-(4-Nitrophenyl)-3-(4-pyrolidin-1-yl-phenyl)-propionaldehyde

To a 5 mL amber scintillation vial equipped with a magnetic stir bar was added G(3.0) dendrimer-bound (2S, 5S)-5-benzyl-2-*tert*-butyl-3-methylimidazolidin-4-one (14.0 mg, 1.0 E-3 mmol, 0.20 equiv catalyst), dimethylsulfoxide (0.45 mL), HCl to acidify the solution to pH ~4 (as a 4N solution in 1,4-dioxane, 30 uL), and 4-nitrocinnamaldehyde (24.1 mg, 0.136 mmol, 1.0 equiv). 1-phenylpyrrolidine was then added to the vial (57.3 uL, 0.398 mmol, 2.98 equiv). After 20 d, the reaction mixture was transferred into dialysis tubing (MWCO 3500) and dialyzed against DMSO. The DMSO outside the dialysis tubing was concentrated under reduced pressure in a lyophilizer and subjected to silica gel chromatography via chromatotron. Gradient elution with 25-50% EtOAc in hexanes followed by concentration in vacuo afforded the product as

a bright orange oil in 26.97% yield (11.9 mg, 0.037 mmol); apparent $[\alpha]_D$= -1.89 (c=0.030 mmol, 1 mL $CHCl_3$), 45% ee.

Free Catalyst in Acetonitrile and Water (2R)-Bicyclo[2.2.2]oct-5-ene-2-carboxyaldehyde

To a 5 mL amber scintillation vial equipped with a magnetic stir bar was added (2S, 5S)-5-benzyl-2-*tert*-butyl-3-methylimidazolidin-4-one (24.2 mg, 0.098 mmol), acetonitrile/water (95/5 v/v, 0.13 mL), acrolein (500 uL, 7.48 mmol), and cyclohexadiene (240 uL, 2.52 mmol). The solution was stirred for 24 h, after which time the reaction mixture was diluted with diethyl ether (10 mL) and washed with water (10 mL). The aqueous layer was extracted with diethyl ether (5 mL x 4) and the combined organics were dried (Na_2SO_4) and concentrated. Purification by silica gel chromatography (10% ether/pentane) afforded the title compound as a colorless oil in 4% yield (14.0 mg, 0.10 mmol); apparent $[\alpha]_D$= -535° (c=5.1*10E-6 mmol, 1 mL $CHCl_3$)

Dendrimer-Bound Catalyst in Dichloromethane (2R)-Bicyclo[2.2.2.]oct-5-ene-2 carboxylaldehyde

To a 5 mL amber scintillation vial equipped with a magnetic stir bar was added G(4.0) (2S, 5S)-5-benzyl-2-*tert*-butyl-3-methylimidazolidin-4-one (3.0 mg, 1.07*E-4 mmol), dichloromethane, acrolein (6.5 uL, 0.097 mmol), and cyclohexadiene (5.0 uL, 0.052 mmol). The solution was stirred for 8 d, after which time the reaction mixture was diluted with diethyl ether (10 mL) and washed with water (10 mL). The aqueous layer was extracted with diethyl ether (5 mL x 4) and the combined organics were dried (Na_2SO_4) and concentrated. Purification by silica gel chromatography (10% ether/pentane afforded the title compound as a colorless oil in 24% yield (17.4 mg, 1.3*10E-4 mmol); apparent $[\alpha]_D$ = -460° (c=3.7*10E-2 mmol, 1 mL $CHCl_3$)

Free Catalyst in Dichloromethane (S)-3-(4-Nitrophenyl)-3-(4-pyrolidin-1-yl-phenyl)-propionaldehyde

To a 5 mL amber scintillation vial equipped with a magnetic stir bar was added (2S, 5S)-5-benzyl-2-*tert*-butyl-3-methylimidazolidin-4-one (24.1 mg, 0.099 mmol, 0.20 equiv) dichloromethane (1.0 mL), HCl (as a 4N solution in 1,4-dioxane, 25 uL), and 4-nitrocinnamaldehyde (88.5 mg, 0.500 mmol, 1.0 equiv). The solution was cooled to -10°C in a salt/ice bath before addition of 1-phenylpyrrolidine (216 uL, 1.485 mmol, 3.0 equiv). After 48 h, the reaction mixture was subjected directly to silica gel chromatography. Gradient elution with 25-50% EtOAc in hexanes followed by concentration in vacuo afforded the product as a bright orange oil in 89% yield (162.1 mg, 0.500 mmol); apparent $[\alpha]_D$=-2.77 (c=0.37 mmol, 1 mL $CHCl_3$), 67%ee.

Free Catalyst in Dimethylsulfoxide (S)-3-(4-Nitrophenyl)-3-(4-pyrolidin-1-yl-phenyl)-propionaldehyde

To a 5 mL amber scintillation vial equipped with a magnetic stir bar was added (2S, 5S)-5-benzyl-

2-*tert*-butyl-3-methylimidazolidin-4-one (27.6 mg, 0.112mmol, 0.20 equiv) dimethylsulfoxide (0.5 mL), HCl (as a 4N solution in 1,4-dioxane, 25 uL), and 4-nitrocinnamaldehyde (99.3mg, 0.500 mmol, 1.0 equiv). The solution was cooled to -10˚C in a salt/ice bath before addition of 1-phenylpyrrolidine (241.8 uL, 1.68 mmol, 3.0 equiv). After 48 h, the reaction mixture was subjected directly to silica gel chromatography. Gradient elution with 25-50% EtOAc in hexanes followed by concentration in vacuo afforded the product as a bright orange oil in 69.5% yield (126.4 mg, 0.390 mmol); apparent $[\alpha]_D$= -3.9 (c=0.34 mmol, 1 mL $CHCl_3$), 94%ee

Free Catalyst in Dichloroethane (S)-3-(4-Nitrophenyl)-3-(4-pyrolidin-1-yl-phenyl)-propionaldehyde

To a 5 mL amber scintillation vial equipped with a magnetic stir bar was added (2S, 5S)-5-benzyl-2-*tert*-butyl-3-methylimidazolidin-4-one (25.6mg, 0.104mmol, 0.20 equiv) dichloroethane (1.0yk mL), HCl (as a 4N solution in 1,4-dioxane, 30 uL), and 4-nitrocinnamaldehyde (92.2 mg, 0.512 mmol, 1.02 equiv). The solution was cooled to -10˚C in a salt/ice bath before addition of 1-phenylpyrrolidine (223.0 uL, 1.55 mmol, 3.0 equiv). After 48 h, the reaction mixture was subjected directly to silica gel chromatography. Gradient elution with 25-50% EtOAc in hexanes followed by concentration in vacuo afforded the product as a bright orange oil in 47.6% yield (80.3 mg, 0.248 mmol); $[\alpha]_D$=-2.36 (c=0.13 mmol, 1 mL $CHCl_3$), 57%ee

Characterization Notes

^{1}H NMR and ^{13}C NMR for all functionalized dendrimer characterization was carried out in deuterated methanol. ^{13}C NMR was optimized by running 8,000-9,000 scans on samples with a 3 second relaxation delay.

Optimum MALDI-TOF-MS characterization can be carried out in both *trans*-3-indolacrylic acid (IAA) or 2,5-dihydroxybenzoic acid (DHB) with dimethylformamide (DMF) as a solvent with the concentrations used in Table 4. It was found that DHB provided more consistent success in MALDI-TOF-MS analysis attempts. Myoglobin was used as a standard for calibration purposes.

Myoglobin standards were prepared by adding 10 uL of myoglobin solution (1.6mg/mL H_2O) to 10 uL of DHB solution (20 mg/mL DHB in 80% ACN/H_2O).

Table 4: Optimal Concentration Range for MALDI-TOF-MS Analysis

Desired Concentration (Dendrimer:Matrix)	uL 1mM Dendrimer Solution	uL 1M Matrix Solution	uL 0.1M Matrix Solution
1:250	**4**	-	**10**
1:500	**2**	-	**10**
1:1000	**1**	-	**10**
1:2500	**4**	**10**	-
1:5000	**2**	**10**	-
1:10k	**1**	**10**	-
1:20k	**1**	**20**	-
1:30k	**1**	**30**	-

1M DHB Matrix solution was prepared by adding 0.0231g of DHB to 150 uL of DMF.

0.1M DHB Matrix solution was prepared by adding 20 uL of 1M DHB solution to 180 uL of DMF.

1 mM dendrimer solutions were prepared according to their theoretical mass based on functionalization and generation (Table 5).

Table 5: Preparation of 1 mM dendrimer solutions.

Generation	Concentration Dendrimer **8** in DMF	Concentration Dendrimers **4,5,6** in DMF
G (2.0)	**5.5mg/mL**	**6.7 mg/mL**
G (3.0)	**12 mg/mL**	**14 mg/mL**
G (4.0)	**24 mg/mL**	**28 mg/mL**

REFERENCES

1. Hajos, Zoltan G., and David R. Parrish. "Asymmetric synthesis of bicyclic intermediates of natural product chemistry." *The Journal of Organic Chemistry* 39.12 (1974): 1615-1621.
2. Ahrendt, Kateri A., Christopher J. Borths, and David WC MacMillan. "New strategies for organic catalysis: the first highly enantioselective organocatalytic Diels-Alder reaction." *Journal of the American Chemical Society* 122.17 (2000): 4243-4244.
3. University, Princeton. "Research." *Princeton University*. Trustees of Princeton University © 2014, 22 Dec. 2010. Web. 31 Mar. 2014.
4. Huang, Y., Walji, A. M., Larsen, C. H., MacMillan, D. W. C. "Enantioselective Organo-Cascade Catalysis" J. Am. Chem. Soc. 2005, 127, 1505115053.
5. "History of Thalidomide - Thalidomide." *Www.thalidomide.ca - Thalidomide*. N.p., n.d. Web. 19 Apr. 2015
6. "(2S,5S)-(-)-2-tert-Butyl-3-methyl-5-benzyl-4-imidazolidinone." *Sigma-Aldrich*. Sigma-Aldrich Co. LLC, n.d. Web. 31 Mar. 2014.
7. "PAMAM Dendrimers." *PAMAM Dendrimers*. Dendritech, n.d. Web. 31 Mar. 2014.
8. Lo, C-M.; Chow, H-F. "Structural Effects on the Catalytic, Emulsifying, and Recycling Properties of Chiral Amphiphilic Dendritic Organocatalysts" *J. Org. Chem.* **2009**, *74*, 5181-5191.
9. Jansen, J.F.G.A.; de Brabander-vand der berg, E.M.M; Meijer, E.W. "Encapsulation of Guest Molecules into a Dendritic Box" *Science* **1994**, *266*, 1226- 1228
10. Balogh, L.; Tomalia, D.A. "Poly(amidoamine) dendrimer-templated nanocomposites. 1. Synthesis of zerovalent copper nanoclusters" *J. Am. Chem. Soc.* **1998**, *120*, 7355-7356.
11. Ooe, M.; Murata, M.; Mizugaki, T. Ebitani, K.; Kaneda, K. "Supramolecular Catalysis by Encapsulating Palladium Complexes within Dendrimers" *J. Am. Chem. Soc.* **2004**, *126*, 1604-1605.
12. Pittlekow, M.; Moth-Poulsen, K.; Boas, U.; Christensen, J. B. "Poly(amidoamine)-Dendrimer-Stabilized Pd(0) Nanoparticles as a Catalyst for the Suzuki Reaction" *Lagmuir* **2003**, *19*, 7682-7684.
13. Oh, S.-K.; Niu, Y.; Crooks, R. M. "Size-Selective Catalytic Activity of Pd Nanoparticles Encapsulated within End-Group Functionalized Dendrimers" *Lagmuir* **2005**, *21*, 10209-10213.

14. Narayanan, R.; El-Sayad, M. A. “Effect of Colloidal Catalysis on the Nanoparticle Size Distribution: Dendrimer—Pd vs PVP-Pd Nanoparticles Catalyzing the Suzuki Coupling Reaction” *J. Phys. Chem. B* **2004**, *108*, 8572-8580.
15. Selkala, S. A.; Tois, J.; Pihko, P. M.; Koskinen, A. M. P. “Asymmetric Organocatalytic Diels-Alder Reactions on Solid Support“ *Adv. Synth. Catal.* **2002**, *344*, 941-945
16. Mannocci, Luca, et al. "High-throughput sequencing allows the identification of binding molecules isolated from DNA-encoded chemical libraries." *Proceedings of the National Academy of Sciences* 105.46 (2008): 17670-17675.
17. Fuentes, German E., Christina A. Chantell, Hirendra Patel, and Mahendra Menakuru. *Conventional Synthesis of a Poly-Alanine Peptide on the Symphony and Prelude*. Protein Technologies, Inc., n.d. Web. 19 Apr. 2015.
18. "Vapor Pressure of Water 20-37 C." Rutgers Chemistry, n.d. Web. 19 Apr. 2015.
19. "Dimethyl Sulfoxide and High-Purity Solvents." *Sigma-Aldrich*. Sigma-Aldrich, n.d. Web. 19 Apr. 2015.
20. Ahrendt, Kateri A., Borths, Christopher J., MacMillan, D.W.C. “New Strategies for Organic Catalysis: The First Highly Enantioselective Organocatalytic Diels-Alder Reaction” J. Am. Chem. Soc., 2000, 122, 4245-4244
21. Paras, Nick A, MacMillan, D.W.C. “The Enantioselective Organocatalytic 1,4-Addition of Electron-Rich Benzenes to α,β-Unsaturated Aldehydes” *J. Am. Chem. Soc.*, **2002**, *124* (27), 7894–7895
22. "Dimethyl Sulfoxide MSDS." Science Lab Chemicals and Laboratory Equipment, n.d. Web. 19 Apr. 2015.
23. Chromatotron Centrifugal Thin-Layer Chromatograph. *T Squared Technology*. TSQtech, n.d. Web. 17 Apr. 2015.