

**SYNTHESIS, CHARACTERIZATION AND EVALUATION OF PAMAM
DENDRIMER-GOLD COMPLEX AS AN ANION RECOGNITION
MATERIAL**

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*This report writing is dedicated to my beloved parent
Abdul Malek Mohd Yusop & Jemilah Ahmad and my family members,
to my adorable supervisor, Prof Dr Salasiah Endud, and also
to my fellow friends.*

Thanks for everything...

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In the spirit of knowledge, I hope to provide useful inputs and remarkable insights for the readers in my research area.

ABSTRACT

Dendrimers are highly branched, monodisperse macromolecules and this field of study has increased rapidly from the time they were discovered about twenty years ago. In this study, PAMAM dendrimer was successfully synthesized by a divergent synthesis route using the reagent excess method starting from ethylenediamine (EDA) followed by consecutive Michael addition and ester amidation reaction. Methanol was used as solvent and three dendrimer generations were prepared: G 1.0, G 2.0 and G 3.0. For preparation of PAMAM-thiol functionalized gold nanoparticle *in-situ* reduction of gold from Au^{3+} to Au^0 was achieved through the reaction of nanogold-thiol functionalized PAMAM dendrimer with sodium borohydrate (NaBH_4). The PAMAM dendrimer-gold complex gave light purple solution and was characterized by ^1H nucleus magnetic resonance (NMR), Fourier transform infrared (FT-IR), and ultraviolet-visible (UV-Vis) spectroscopies. Analysis of the UV-Vis spectral analysis of the PAMAM dendrimers showed that the wavelength maximum, λ_{max} significantly shifted from 330.15 nm to 517.28 nm with the addition of gold-thiol nanoparticles due to binding of the thiol functional group to gold particles. Anion recognition ability of the PAMAM-thiol functionalized gold nanoparticle has been studied by treating the PAMAM dendrimer-gold complex with nitrate ion. Based on the UV-Vis spectra, the wavelength maximum of Au(III) was shifted from 526.98 nm to higher wavelength upon binding of the nitrate anion to the surfaces of gold-thiol nanoparticles which the process involved excitation of the electrons from $\pi \rightarrow \pi^*$.

ABSTRAK

Dendrimer adalah makromolekul ekasebar dengan struktur bercabang-cabang dan bidang kajian ini telah berkembang pesat sejak penemuannya hampir dua puluh tahun yang lalu. Dalam kajian ini, dendrimer PAMAM telah berjaya disintesis dengan menggunakan kaedah sintesis divergen dengan menggunakan kaedah reagen berlebihan bermula dengan etilenadamina (EDA) diikuti tindak balas penambahan Michael dan amidasi ester secara berturutan. Metanol telah digunakan sebagai pelarut dan tiga generasi dendrimer telah disediakan iaitu G 1.0, G 2.0 dan G 3.0. Bagi penyediaan PAMAM-tiol berfungisikan nanopartikel emas penurunan *in-situ* emas dari Au^{3+} kepada Au^0 telah dijalankan melalui tindak balas dendrimer PAMAM berfungisikan nanoemas-tiol dengan natrium borohidrat (NaBH_4). Kompleks dendrimer PAMAM-emas tersebut menghasilkan larutan berwarna ungu muda dan telah dicirikan menggunakan spektroskopi Resonan Magnet Nuklear ^1H (RMN), inframerah transformasi Fourier (FT-IR) dan ultralembayung-nampak. Analisis spectrum ultralembayung-nampak dendrimer PAMAM menunjukkan panjang gelombang maksimum, λ_{max} secara signifikan telah beranjak dari 330.15 nm kepada 517.28 nm dengan penambahan partikel nanoemas-tiol akibat penambatan kumpulan berfungsi tiol oleh partikel emas. Sifat pengenalpastian anion PAMAM berfungisikan partikel nanoemas-tiol telah dikaji melalui tindak balas kompleks dendrimer PAMAM-emas dengan ion nitrat. Berdasarkan spectrum UV-vis, panjang gelombang maksimum Au(III) didapati beranjak dari 526.98 nm kepada panjang gelombang yang lebih tinggi apabila terjadi penambatan ion nitrat pada permukaan nanopartikel emas-tiol yang mana proses tersebut melibatkan pengujaan elektron dari orbital $\pi \rightarrow \pi^*$.

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LIST OF ABBREVIATIONS

FTIR	-	Fourierr transform infrared
EDA	-	Ethylenediamine
TEM	-	Transmission electron microscopy
UV-vis	-	Ultraviolet-visible
Au	-	Aurum
PAMAM	-	Poly(amidoamide)
λ_{\max}	-	Maximum wavelength
SH	-	Thiol group
NMR	-	Nuclear magnetic resonance
NaBH ₄	-	Sodium Boro Hydrate
S	-	Sulfur
NO ₃ ⁻	-	Nitrate ion

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CHAPTER 1

INTRODUCTION

1.1 Background of Study

Since the pioneering work of well-defined, three-dimensional structural order macromolecules by Vögtle [1], Tomalia [2,3], and Newkome [4], interest in dendrimers and hyperbranched polymers has been increasing at an amazing rate.

The study of these polymers expands to all areas including theory, synthesis, characterization of structures, and properties, and investigations of potential applications. In the beginning the research on dendrimers focused on the synthesis, characterization, and properties of perfect dendrimers of higher generations. For the synthesis of dendrimers constructed by step-by-step sequences, two fundamentally different strategies, the divergent approach (from the inside out) [2,4] and convergent approach (from the outside in) [5], were employed. In either way, dendrimers can be prepared with high regularity and controlled molecular weights, and the macromolecules consist of a polyfunctional central core covalently linked to layers of repeating units (generations) and a number of terminal groups (Figure 1.1). These units are interdependent and create a unique molecular shape, leading to intrinsic properties such as high solubilities and low viscosity.

Dendrimers free-shaped synthetic macromolecule has garnered a great deal of scientific interest due to their unique molecular nanostructure. Used in a variety of

scientific applications, the use of dendrimers is now widely regarded as a safer, more precise, and more effective way to practice medicine [6].

Nanocomposites are materials that are created by introducing nanoparticulates (often referred to as filler) into a macroscopic sample material (often referred to as the matrix). This is part of the growing field of nanotechnology. After adding nanoparticulates to the matrix material, the resulting nanocomposite may exhibit drastically enhanced properties. For example, adding carbon nanotubes tends to drastically add to the electrical and thermal conductivity. Other kinds of nanoparticulates may result in enhanced optical properties, dielectric properties or mechanical properties such as stiffness and strength.

In general, the nanosubstance is dispersed into the matrix during processing. The percentage by weight (called mass fraction) of the nanoparticulates introduced can remain very low (on the order of 0.5% to 5%) due to the incredibly low filler percolation threshold, especially for the most commonly used non-spherical, high aspect ratio fillers (e.g. nanometer-thin platelets, such as clays, or nanometer -diameter cylinders, such as carbon nanotubes). Dendrimers are known for their three-dimensional, monodispersed, highly branched, macromolecular nanoscopic architecture with a number of reactive end groups [7]. Commercially available PAMAM (poly(amidoamine)) dendrimer prepared by the divergent growth approach of Tomalia et al. are one of the most widely used dendrimer scaffolds in biology. These macromolecules have uniform size and monodispersed. Furthermore, high structural and chemical homogeneity of dendrimer might facilitate quality control of their drug conjugates in production.

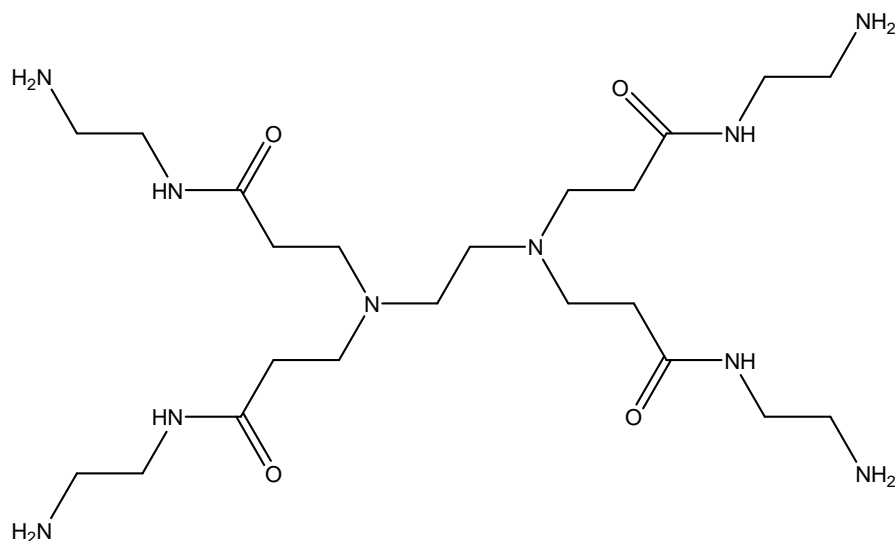


Figure 1.1: Generation 1.0 of PAMAM dendrimer with ethylenediamine core and amine surface group.

The divergent route to dendrimer synthesis is based on the construction of a molecular superstructure starting with a focal point or core and progressing outward to the periphery, as illustrated in Figure 1.2. Dendrimers are built in layers, or generations, upon a defined core that possesses a specific number of active sites, to which the successive tiers are, for the most part, covalently attached. The number of active sites on the core determines their n-directionality and limits the number of building blocks that can be added to form the next generation. This trend is repeated (iterative synthesis) as the reactive sites on the periphery of the previous generation are revealed for the assembly of the next generational growth layer.

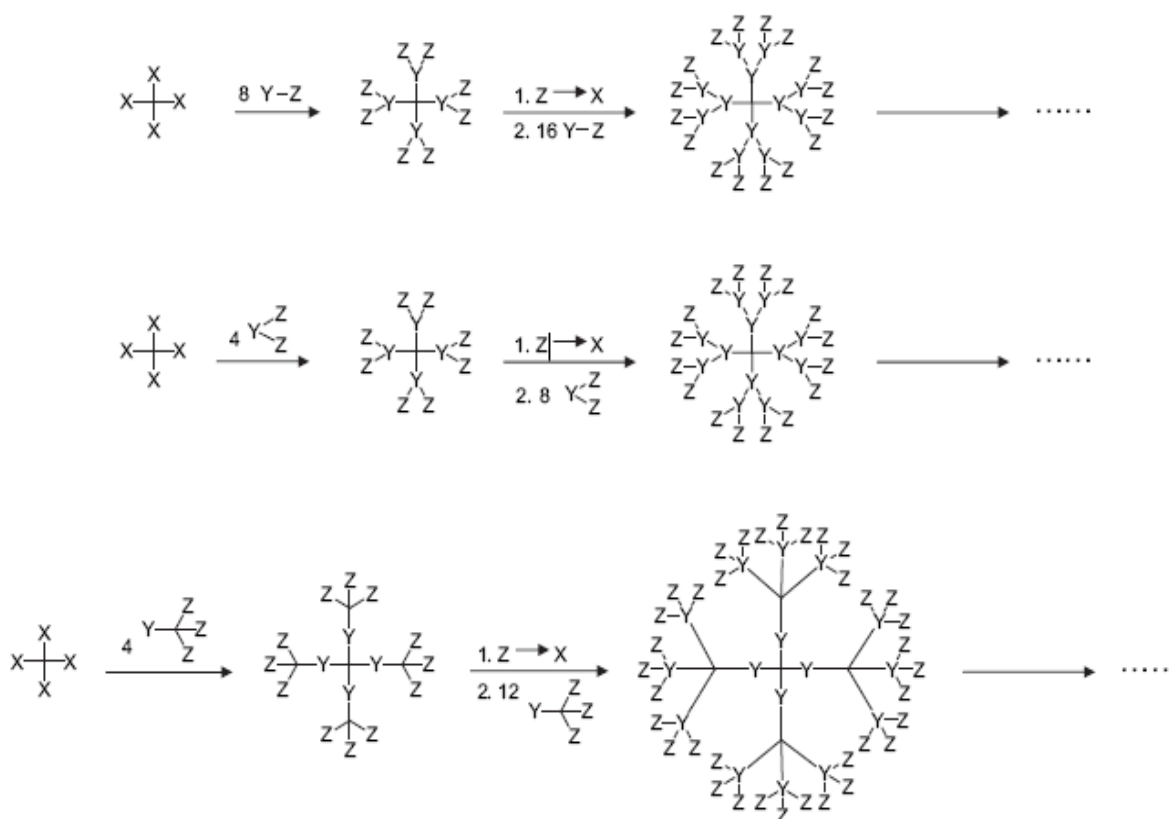


Figure 1.2: Divergent procedures for macromolecular construction.

Assuming that the monomer's functional group(s), steric hindrance, and active site accessibility do not interfere with the construction of ideal dendrimers, the divergent process permits the exponential growth of free active sites per generation. Perfect growth is only achieved when each active site is occupied by a building block (monomer) denoting the next tier; otherwise, imperfect structural assembly results leading to internal termini and variable internal void regions, which starts to resemble a hyperbranched motif normally derived by a random one-step procedure. If these imperfections or “branching defects” occur early in the generational growth, they can have serious repercussions on the overall micellar properties of the resulting dendrimer.

1.2 Problem Statement

The study of anion has been a critical part in the most recent research in the area host-guest chemistry [8]. In order to differentiate a target anion from other, the host molecule must be carefully designed, considering not only the structural complementary interaction between the ion-molecule pair, but also interaction with solvent molecules. The host molecules for anion recognition comprise at least one interaction site that improves the selective interaction for the target anion and overcomes the solvation energy of the target anion in the aqueous phase. Among this host molecules, dendrimers are of considerable interest as anion recognition material because they can provide dedicated single-point interactions with the capability to further modify the host molecular structure with a number of reactive end groups as well as possess internal cavities [2]. These characteristics, along with water solubility, are some of the features that make them attractive for environmental remediations [9].

Dendrimers that are functionalized with transition metals in the core can potentially mimic properties of enzymes, their efficient natural counterparts (e.g. cytochrome P-450), whereas the peripheral-functionalized systems is proposed to provide ideal building blocks for the development of high-capacity, selective and recyclable ligands for the recovery of anions.

One of the major problems related to the preparation of dendrimer is to modify the surface of the molecules [10]. The difficult part is to protect the active site in the dendrimer. It is known that the difference functional group at end terminal of the dendrimer gave difference characteristics. Hydrophilic functional group that is attached to end terminal will make the dendrimer soluble in water meanwhile, hydrophobic functional group given the ability to soluble in organic solvent. Recently, dendrimers have been used in medical application as a censoring device by using gold encapsulated at intermolecules cavities of the dendrimer. For example, detection of α -1-fetoprotein

(AFP) has been designed based on antibody functionalized core shell nanocomposite particles [11].

By introducing gold at the peripheral of a dendrimer molecule, sophisticated artificial receptors exhibiting specific anion recognition can be obtained. Molecular recognition moieties attached at the peripheries of dendrimers may act as *exo*-receptors for analytes. In previous studies researchers have developed metallocene as hosts for recognition of various anions [9].

The presence of $-NH$ groups within the dendritic structure was established to be important for anion recognition [12-14]. In this research, PAMAM dendrimer-gold complex was prepared by attaching gold-thiol nanoparticles to the periphery of PAMAM dendrimer by the divergent method with the aim to prevent its aggregation and improve the solubility in water. PAMAM encapsulated gold nanoparticles was prepared only as a comparison. Gold was chosen in this study because of its nanoparticles size and high sensitivity to UV-Vis detection even in a small amount and soluble in most inorganic solvents. The capability of a methanol soluble dendrimer to encapsulate and transport PAMAM-gold thiol derivatives selected both as model compounds and for their potential anion recognition properties e.g. nitrate will be investigated. The anion recognition studies will be conducted by UV-Vis spectroscopy. The recognition of anions is deemed possible as a result of electrostatic interaction between the gold third linkage of the dendrimer and the anion.

1.3 Objective of the Study

1. To synthesize and characterize Poly(amidoamine) (PAMAM) dendrimers with various generation number ($G = 0.5, 1.0, 1.5, 2.0, 2.5$ and 3.0)
2. To synthesize and characterize PAMAM dendrimer-gold complex.
3. To synthesize PAMAM encapsulated gold nanoparticles for comparison with PAMAM dendrimer-gold complex.
4. To study the ability of PAMAM dendrimer-gold complex to bind with nitrate ion using UV-Vis spectroscopy.

1.4 Scope of the Study

The scope of this study includes the synthesis of PAMAM dendrimer via Michael Addition reaction by divergent method. PAMAM dendrimer-gold complex was synthesized by addition of gold-thiol nanoparticles into the full generation of PAMAM dendrimer. PAMAM encapsulated gold nanoparticles was synthesized by reduction of tetrachloroauric acid (HAuCl_4) by using reducing agent, sodium borohydrate (NaBH_4).

The generation number of PAMAM dendrimer was determined by using several characterization techniques such as Fourier Transform Infrared (FTIR) Spectroscopy and ^1H Nuclear Magnetic Resonance (NMR) Spectroscopy. After addition of gold-thiol nanoparticles, the chemical properties of the PAMAM dendrimer gold-thiol functionalized complex was characterized using Ultra Violet-Visible (UV-vis) Spectroscopy.

1.5 Outline of the Study

This dissertation illustrates the information concerning the synthesis and characterization of PAMAM based gold nanocomposites. Chapter 1 elucidates the research background and the important strategies to respond the current issue. Chapter 2 presents the literature review regarding this project where it contains some background information about the whole research done. Chapter 3 describes the research methodology with the characterization techniques used in this research as shown in the flowchart in Figure 1.3. Chapter 4 explains the results and discussion of the PAMAM dendrimers obtained their and its characterization. Finally, chapter 5 summarizes the results obtained with recommendation for future work.

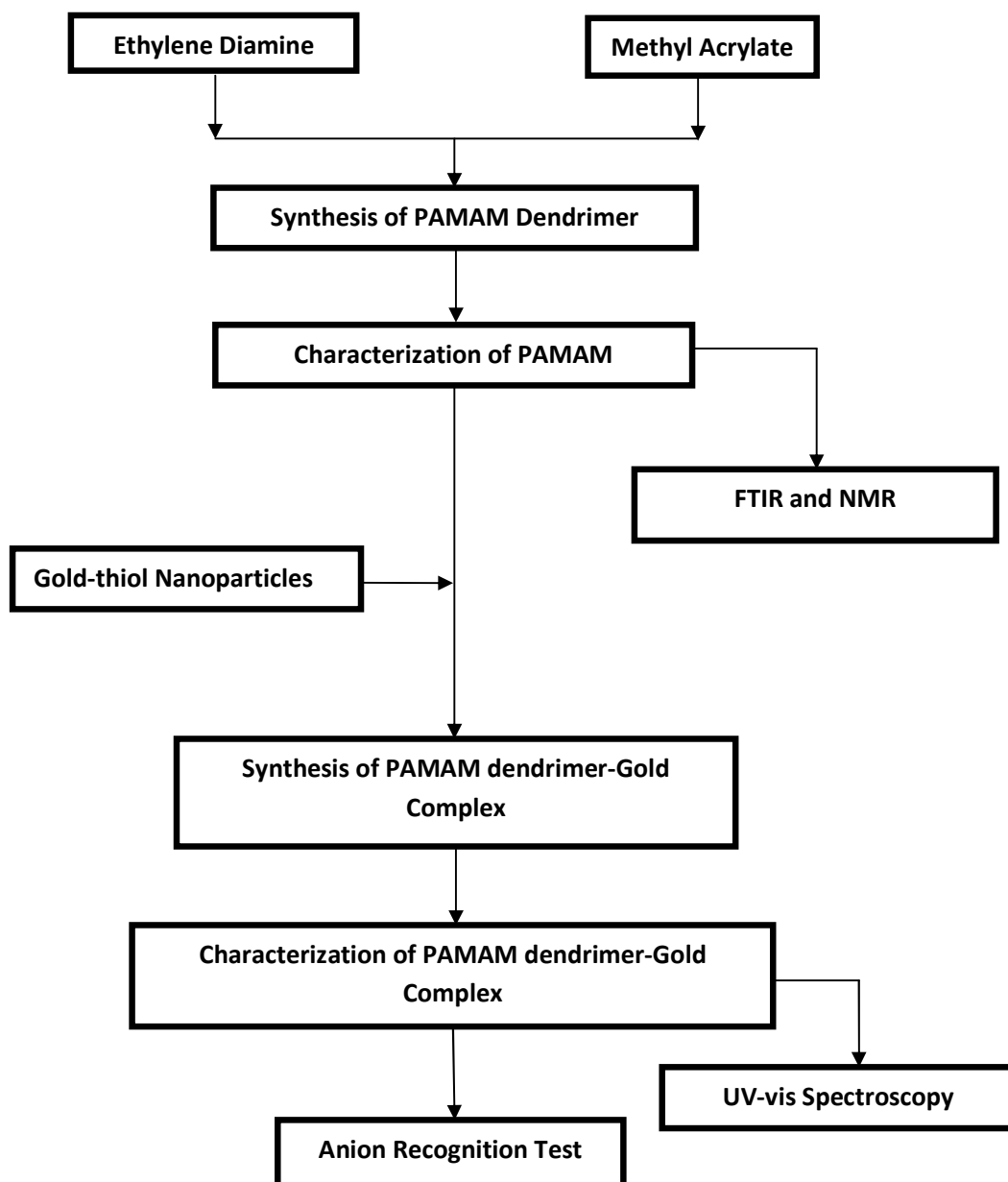


Figure 1.3: Flowchart of the Research Methodology