CHAPTER I

INTRODUCTION

1.1 Polydiacetylene

Polydiacetylene (PDA) is an ene-yne conjugated polymer consisting of double and triple bonds in its backbone. It is normally prepared via a topological 1,4-addition polymerization of diacetylene monomer by heat, UV-light or γ rays.[1] PDA possess several interesting optical properties including solvatochromism, thermochromism and mechanochromism.

Topological polymerization is a polymerization which requires precise prelignment of the monomer that is usually found in solid state crystal.[2-4] The distance between diacetylene monomer which the polymerization occur is ~5 Å and the orientation angle (θ) is ~45° relative to the translation axis (Figure 1.1).[5-6]

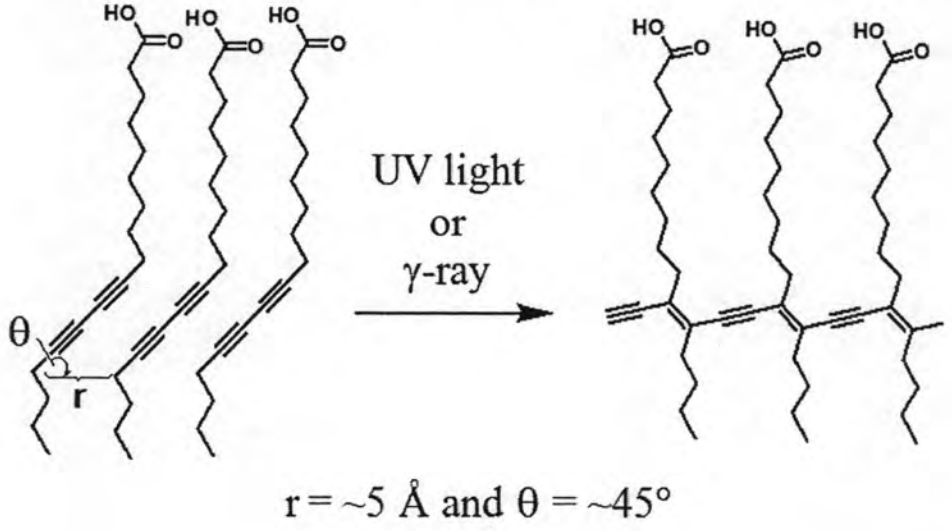

Figure 1.1 Packing parameters: r = ~5 Å and θ = ~45° required for the topological polymerization of a diacetylene monomer.

1.2 Polydiacetylene vesicles

There are numerous forms which PDA can be structured: bulk single crystal, Langmuir monolayer film, multilayer film, nano tube and vesicle. The vesicle is one of the most widely used forms of polydiacetylene for sensor applications . It is a water filled spherical assembly of lipid bilayer. The formation of vesicle is thermodynamically driven so that the hydrophobic surface of the lipid molecules is not exposed to water. Only the hydrophilic head group of the lipid is exposed to water inside and outside of the bilayer membrane. A number of lipids containing diyne unit can self assemble into vesicle that have the right packing parameters for topological polymerization to form PDA vesicles (Figure 1.2). In the form of vesicle, PDA can be homogeneously dispersed in an aqueous media to form a sol type colloid which is convenient for further characterization, fabrication and applications. The most studied diacetylene lipids are 10,12-pentacosadiynoic acid (PCDA). The interest in these lipid vesicles is mainly related to the development into bio-[7] and chemosensors[8].

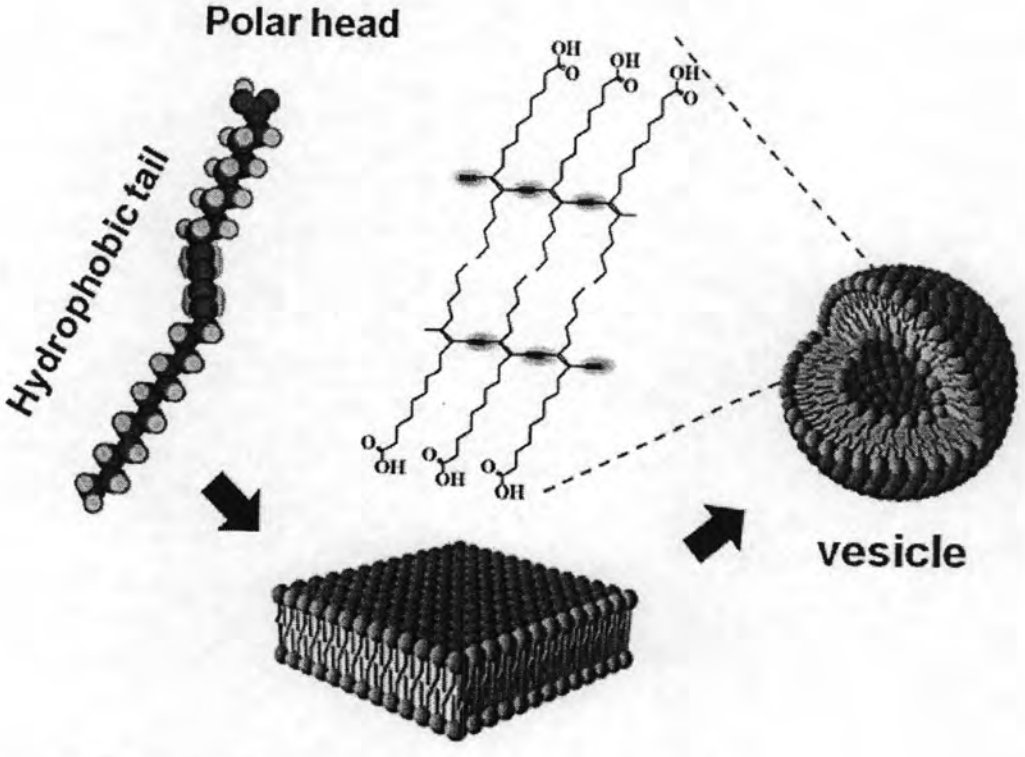

Figure 1.2 Structure and formation of a PDA lipid vesicle.

1.3 Polydiacetylene films

It is widely accepted that the sensing process of PDA of diacetylene lipids is initiated at the lipid head group. Intuitively, the most desirable form of PDA for sensing applications is thin film as it can maximize sensing area and it is easy to be developed into simple to use device. Several techniques have been utilized for making different types of thin films on the surface of substrates.

1.3.1 Langmuir-Blodgett film

Langmuir-Blodgett film can generate an ideal highly ordered monolayer assembly of lipid molecules at water/air interface. The monolayer can be transferred onto solid substrate by horizontal or vertical deposition (Figure 1.3). The monolayer Langmuir-Blodgett film is however often fragile and difficult to prepare in large area. The number of layers can be increased by repeating the transferring process with a newly generated monolayer. Hence, it can be quite bothersome to prepare a multilayer film as the preparation of each new monolayer requires a slow surface compression and the transferring step needs adroit attention. This type of PDA film is one of the most interesting form of PDA in research but the development of the technique for practical mass production remains elusive.[9]

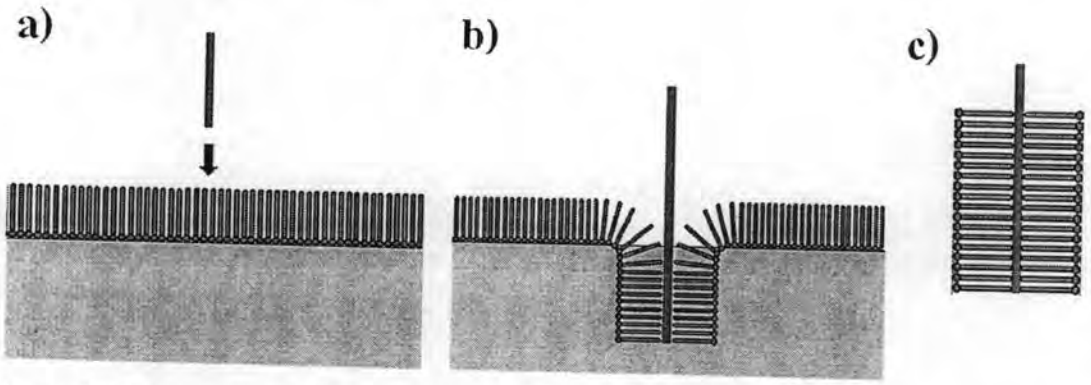

Figure 1.3 Preparation of Langmuir Blodgett film a) floating of condensed monolayer film on the water subphase, b) film deposition, c) monolayer films on the substrate.

1.3.2 Covalently linked polydiacetylene vesicles on the substrate surface

Immobilization of polydiacetylene vesicle is one of the most fashionable techniques to prepare monolayer of vesicle film. In 2001, immobilization of polymerized diacetylene vesicles prepared from a mixed lipid of diacetylene phospholipid and disulfide-modified diacetylene lipid onto gold film was reported.[10] The surface roughness of a gold film after immobilization of the vesicles without

disulfide-modified functional group was 2.2 nm while that of the gold film immobilized with vesicle with disulfide group was 83 nm (Figure 1.4). However it is not mentioned in the report about the color phase (blue or red) of the PDA vesicles deposited on the surface.

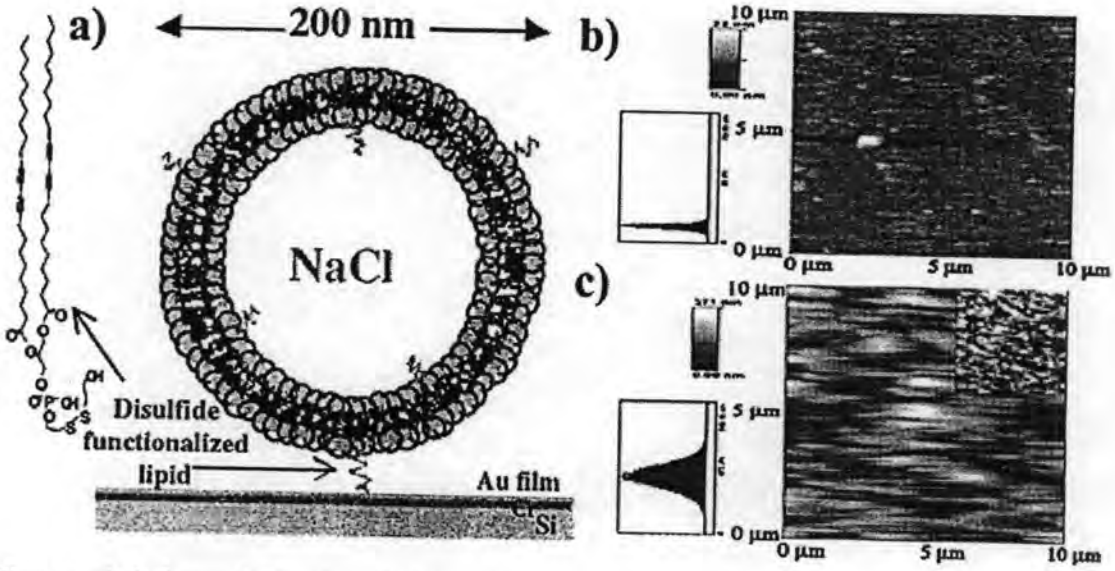

Figure 1.4 Immobilized polydiacetylene vesicles on gold surface by disulfide-modified diacetylene. a) depiction of vesicles attached to Au layer by disulfide linkage, AFM image of a gold film after immobilization of vesicles b) without disulfide modification and c) with disulfide modification.[10]

Since one of the most important properties of PDA is the blue-to-red color transition, for sensing application, it is thus necessary to immobilize the vesicle in the blue phase. Kim and coworkers proposed a new route to immobilize PDA vesicles onto glass slide by using an aldehyde modified glass slide and amino terminated diacetylene (Figure 1.5a).[11] Two routes of immobilization were studied. In method A, the diacetylene vesicles were irradiated with UV-irradiation first to convert the monomeric vesicles to the corresponding PDA vesicles and then immobilized the resulting PDA vesicles on the glass substrate. In method B, the diacetylene vesicles were immobilized first then followed by the irradiation. The film prepared by method A showed higher electronic absorption at 550 nm in comparison to that prepared by method B indicating the red phase occurred in method A more than method B. The control experiment using unmodified glass slide did not show the absorption of vesicles.

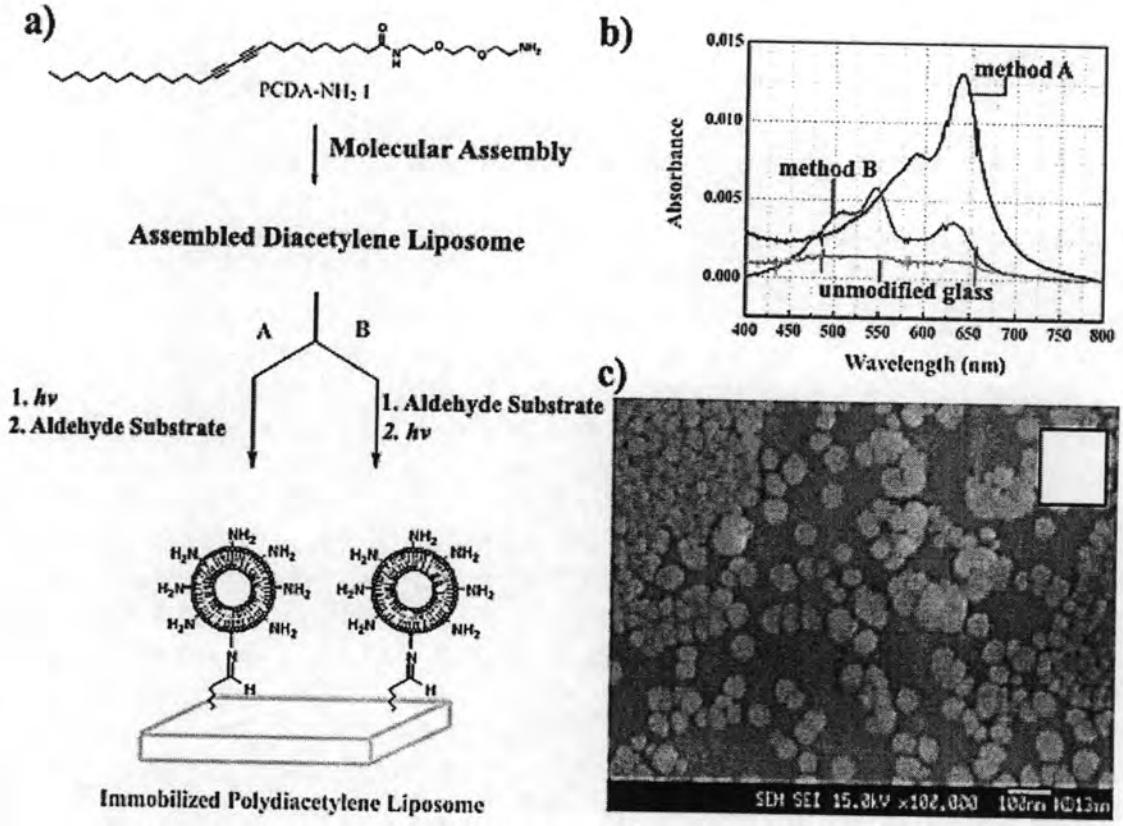

Figure 1.5 Immobilization of polydiacetylene vesicle onto aldehyde modified-glass. a) schematic immobilization by method A and B, b) absorption spectra of vesicle immobilized films, c) AFM picture of vesicles on modified glass.[11]

An alternative approach utilized an amide bond formation between *N*-hydroxy succinamide active ester of 10,12-pentacosadiynoic acid (PCDA) and amino functionalized glass substrate.[12] The active diacetylene was incorporated into vesicle before immobilization. Again, the imobilization of the monomeric vesicles prior to the polymerization (method B) successfully yielded the blue PDA vesicles attached on the glass slide but the immobilization of the prepolymerized vesicles (method A) resulted in the red vesicles (Figure 1.6).

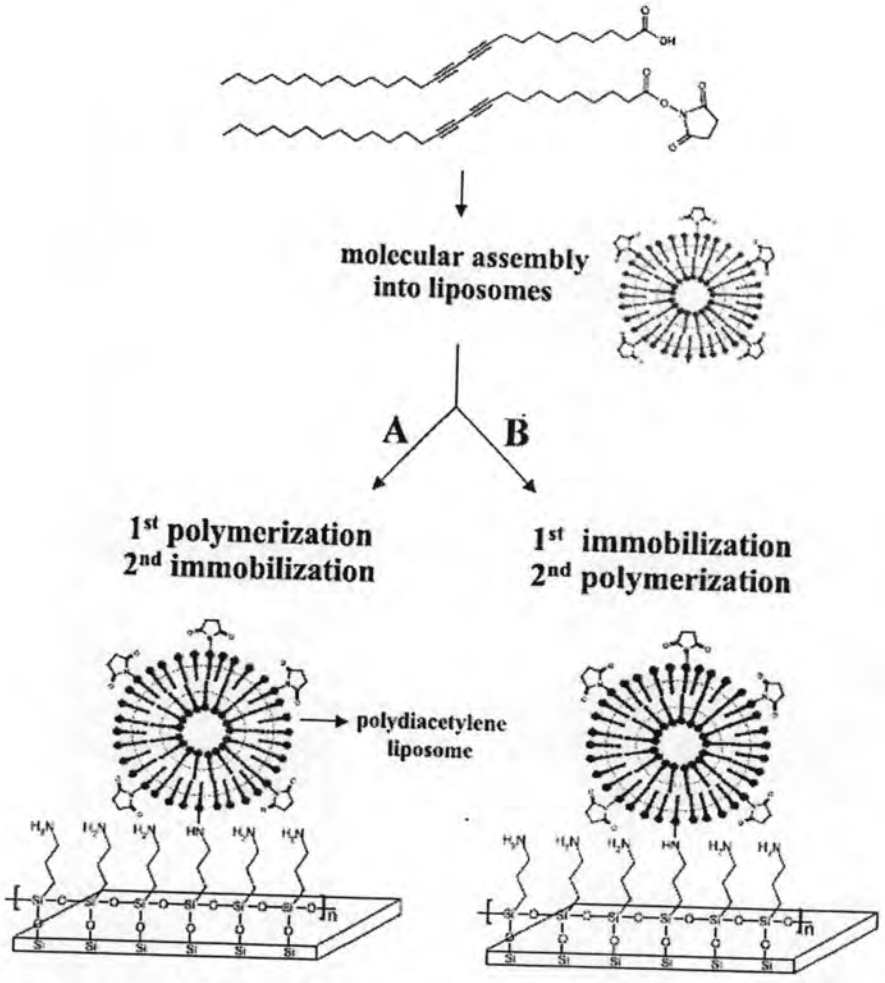

Figure 1.6 Illustration of the two different methods to immobilize polydiacetylene vesicle onto modified-glass by amide linkage.[12]

The covalent bond formation provides a very robust attachment of the vesicles to the substrate surface. An assembly of a multilayer film by this technique is however not yet to be realized. This technique is hence only suitable for the preparation of a single layer film which give very low electronic absorption and the color is not visible.

1.3.3 Polyelectrolyte multilayers films

The polyelectrolyte multilayers (PEM) films can be prepared by dipping a hydrophilic substrate into a solution of polyanion and polycation in an alternating fashion which is called layer-by-layer depostion.[13-14] The thickness of the film grows by electrostatic interaction between the positive charges of polycation and the negative charges of polyanion. The number of layers in the PEM films can be controlled by the number of dipping cycles (Figure 1.7). Comparing to the other

techniques mentioned earlier, this technique is far more convenient for preparation of multilayer films and has been extended with many materials such as proteins and colloids.

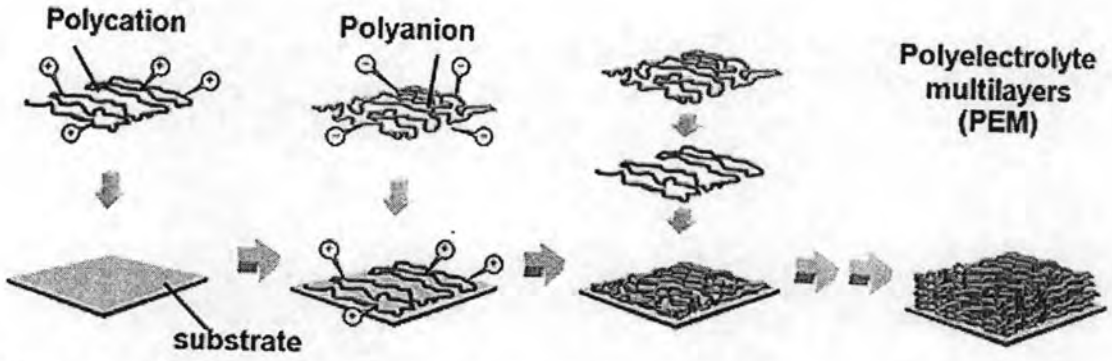

Figure 1.7 Schematic illustration of polyelectrolyte multilayer film (PEM) preparation.

In 1997, Saremi studied a layer-by-layer deposition of diacetylene bolaamphiphile on the glass substrate precoated with poly(allylamine) hydrochloride (PAH) and polystyrene sulfonate (PSS). The anionic head groups of the bolaamphiphile were either sulfonate or phosphate and the cationic head groups of the bolaamphiphile was pyridinium. The prepared multilayer films of the anionic and cationic diacetylene monomers can be polymerized by UV irradiation. The polydiacetylene films obtained was red with the maximum absorption at 500 – 550 nm (Figure 1.8).[15]

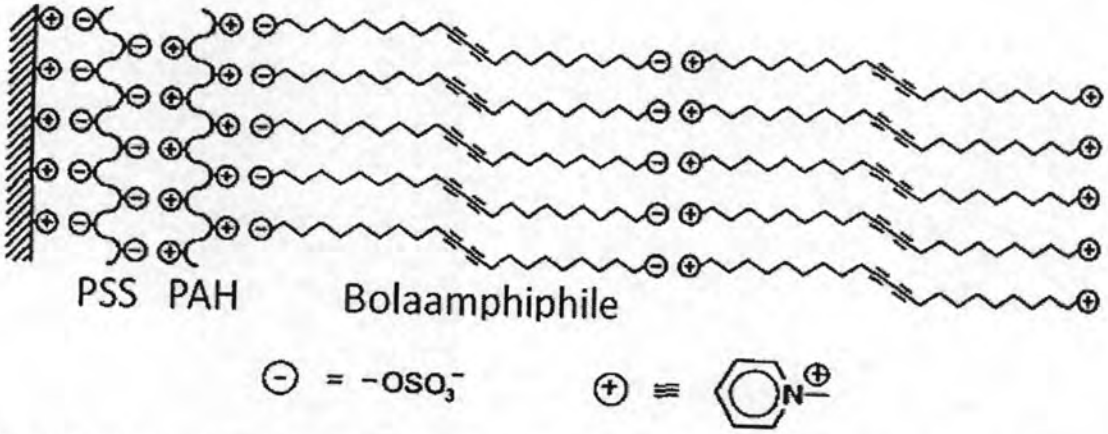

Figure 1.8 Deposition of a bolaamphiphile diacetylene monomer on PSS/PAH coated glass.[15]

In 2005, Su studied a layer-by-layer deposition of vesicles of diacetylene lipids on a quartz slide by alternate dipping of 10,12-pentacosadiynoic acid (PCDA)

and aminoethyl-10,12-pentacosadiynamide ($PCDANH_2$) vesicles. The multilayer films of vesicle assembly could be polymerized to give blue polydiacetyene films.[16] Using of polymerized polydiacetylene vesicles to prepare PEM film did not report in this work (Figure 1.9).

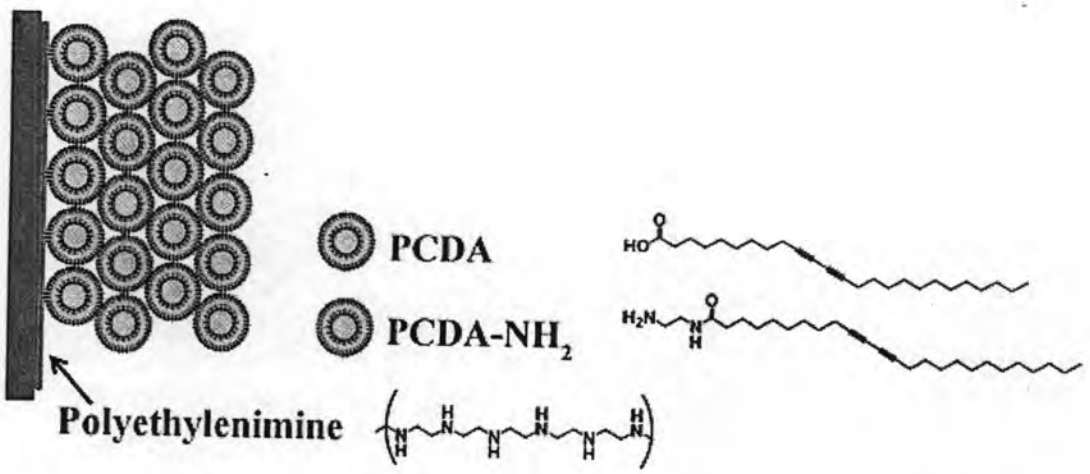

Figure 1.9 Depiction of layer-by-layer deposition of unpolymerized diacetylene vesicle between PCDA and PCDA-NH_2.[16]

In 2006, Kim studied the PEM films between polystyrene sulfonate and vesicles of poly (DADMDPA-bis-PCDA) which is a disubstitution derivative of 3,3'-diamino-*N*-methyldipropylamine with two PCDA units (Figure 1.10).[17] The disubstituted polydiacetylene vesicle was highly stable in various pH condition due to there is no protonated group. The PEM films can exhibit only thermochromic properties. These method can produce a highly stable PEM film of polydiacetylene vesicle but cannot be used for chemosensor application.

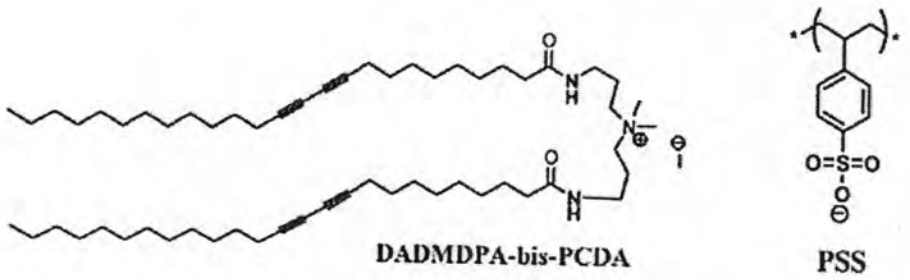

Figure 1.10 Structure of DADMDPA-bis-PCDA and PSS.[17]

In this dissertation, the novel polyelectrolyte multilayer films of polymerized polydiacetylene vesicle which can exhibit chromic properties under influences of pH, organic solvent and heat was investigated.

1.4 Optical and electronic properties

Most diacetylene monomers are white or have no color but after polymerization the ene-yne conjugation backbone of PDA produces a color which depends on the substituent of the diyne monomer and the condition used for polymerization.[18] For example, Fujita and coworkers studied a relative of the number of methylene sidechain in monomer had effect on the color of polymer (Figure 1.11).[19] The author prepared polymer gel in hexane which exhibited blue color when the number of methylene side chain was even while exhibited red color when the number of methylene side chain was odd. The author proposed the different bending angle of polymer chain which the "odd monomer" had more bending than the "even monomer". The higher bending of polymer chain produced a less conjugation of backbone which effected on the electronic absorption of polymer.

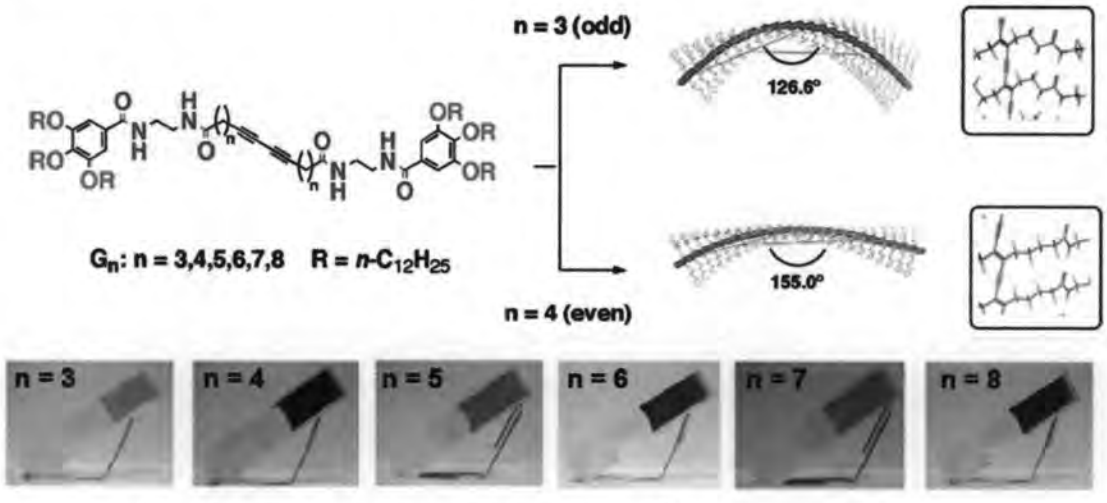

Figure 1.11 Color of polydiacetylene gel G_3 to G_8.[19]

The strong coupling between the electronic structure and the UV-visible absorption of these polymers leaded to unique optical and electronic properties. Those properties demonstrated tremendous interests in polydiacetylenes. The blue color forms are most widely used for sensing applications. Usually, a blue PDA shows the maximum of electronic absorption around 630-650 nm. Some PDA are obtained in red color in which case the absorption maximum appeared at lower wavelength around 530-550 nm.[20] The electronic absorption associates with the excitation

energy of electron in a HOMO ground state to a LUMO excited state. It is thus possible to modify the electronic absorption spectrum of PDA by varying the degree of polymerization and steric factors of the substituents in the diyne monomer.[21-22]

1.4.1 Chromism properties

One of the most intriguing properties of PDA is its ability to change color upon its exposure to an external stimulant such as heat, chemicals and mechanical stress. The color change induced by heat is called thermochromism which always causes the electronic absortion of PDA shifted to shorter wavelength resulting in the color change from blue to red or from red to yellow.[23] The color change caused by chemical exposure maybe categorized as chemochromism[8] and the color change caused by mechanical stress is called mechanochromism.[24] The terms solvatochromism, alkalinochromism and biochromism have also been used to call the color transition process induced by solvent, base and biological agents, respectively.[25]

Colorimetric response (%CR)

A quantitative value for the extent of blue-to-red color transition is given by the colorimetric response (%CR) which is defined as $\%CR = (PB_0\text{-}PB)/PB_0X100$. Where $PB = A_{blue}/(A_{blue}+A_{red})$, A_{blue} and A_{red} are the absorbance of the blue and the red phases at 630 and 540 nm, respectively. PB_0 is the initial percent blue of the vesicle solution.

Thermochromism

Thermochromism is probably the earliest chromism property found for PDA.[25-28] The most notable thermochromism involves the color change from blue to red that have been developed for various colorimetric and fluorescence sensing applications. The thermochromism can be either reversible and irreversible depending on the interaction between the side chain substituents and side chain head groups. There are many dedicated effort research to elucidate the mechanisms of color change in PDA materials which is still not fully understood. It is liked more than one mechanism causing chromic change, with the specific mechanism which depended on the nature of polymer. The report has shown that both side chain order, head group also hydrogen bonding affect the chromic state. For example Kim and coworkers have

reported the relation between intramolecular hydrogen bonding of 10,12 pentacosadiynamide of *m*-aminobenzoic acid (PCDA-mBZA) which showed the thermochromic reversibility (Figure 1.12).[29-30]

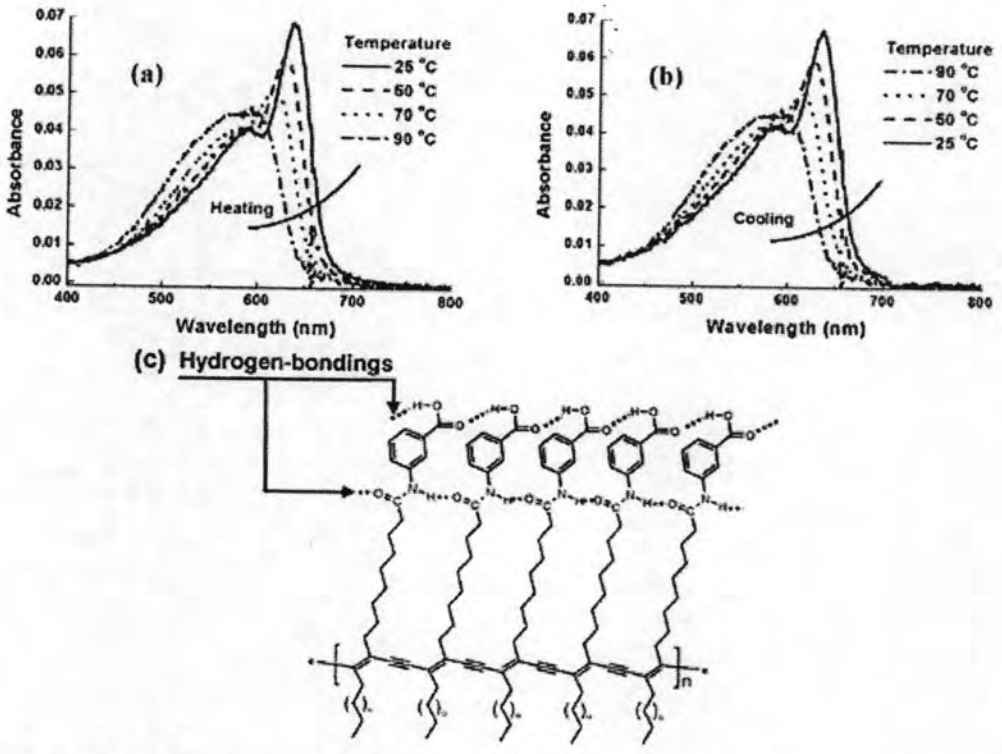

Figure 1.12 Absorption spectrum of PCDA-mBzA which show thermochromic reversibility. a) heating, b) cooling, c) intramolecular hydrogen bonding of PCDA-mBzA.[30]

Dautel and coworkers reported the thermochromic reversibility of polydiacetylene nanofiber which had color reversible between purple to red (Figure 1.13). The interaction between urea groups produced a thermochromic reversibility.[22]

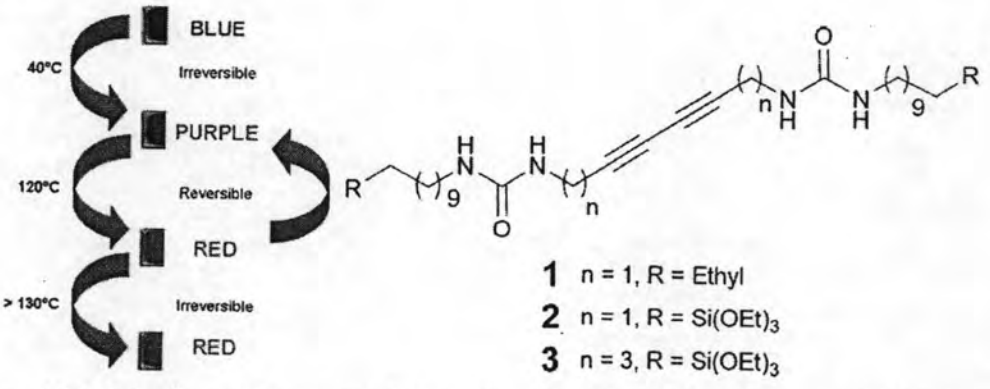

Figure 1.13 Polydiacetylene nanofiber gel with thermochromic reversibility between purple to red.[22]

In case of polydiacetylene nanotube, the nanolipid assembly of diacetylene ammonium salt produce a tubule structure. The thermochromism of nanotube demonstrated thermochromic reversibility from blue to red in between 20° to 50°C (Figure 1.14).[31]

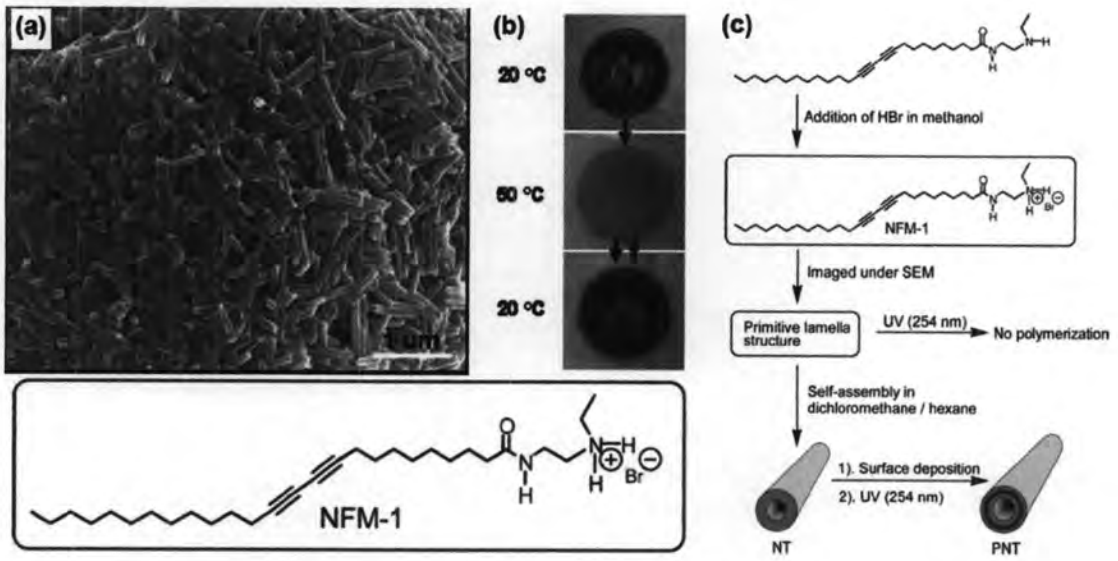

Figure 1.14 Thermochromism of polydiacetylene nanotube. a) polydiacetylene nanotube, b) thermochromic reversibility, c) preparation of lipid assembly nanotube.[31]

In 2008, Gu and coworkers investigated the blending product of nanocrystal of 10,12 pentacosadiynoic acid (PCDA) and poly(vinylpyrolidone). The blending product had intermolecular hydrogen bonding between carboxylic group of PCDA with pyrolidone that showed thermochromic reversibility (Figure 1.15).[32]

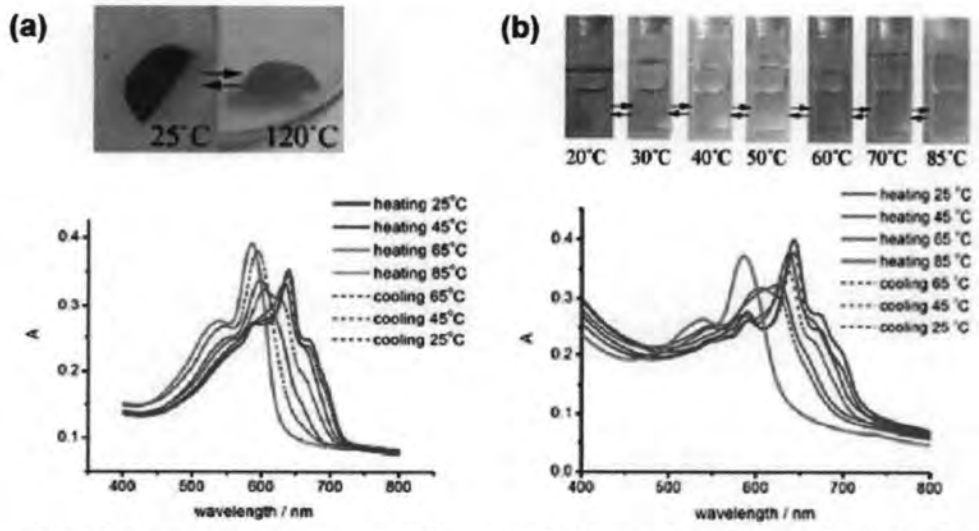

Figure 1.15 Thermochromic reversibility of polydiacetylene blended with poly(vinyl pyrolidone) a) polymer blend film, b) its aqueous suspension.[32]

An anothor example for the thermochromism of polydiacetylene from red to yellow was reported by Dei S. and coworkers. The poly(benzyl-10,12-pentacosadiynoate) was polymerized by gamma radiation in crystalline form. The polymer had reversible thermochromism from 40°C to 130°C (Figure 1.16).[23]

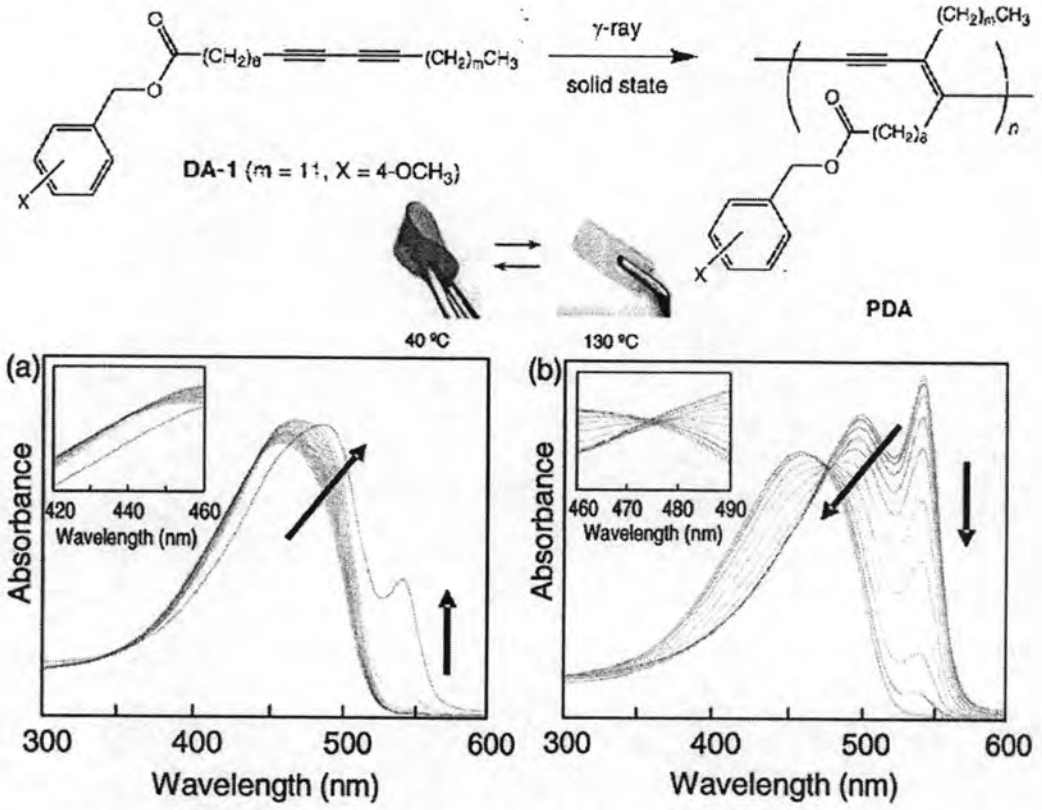

Figure 1.16 Thermochromic reversibility of poly(benzyl-10,12-pentacosadiynoate). a) cooling, b) heating.[23]

Mechanochromism

Mechanochromism is a property where a material change its color in response to an applied strain. The blue-to-red transition of polydiacetylene can be activated by some mechanical force. For example, Muller and Eckhardt observed an irreversible transition in a PDA single crystal induced by compressive stress, which resulted in coexisting blue and red phases.[33] Nallicheri and Rubner observed reversible mechanochromism for conjugated PDA chains embedded in a host elastomer that was subjected to tensile strain.[24]

Solvatochromism

Solvatochromism is one of the chromic properties of polydiacetylene. It is believed that solvation of polymer side chain by organic solvent was caused the side chain disorder which affected on the conjugation of polydiacetylene backbone. The color of the polymer changed from blue to red or yellow. For example, Lim and coworkers synthesized four substituted calixaline (PC4BU) to study solvatoshromism of them. The PC4BU showed solvatochromic properties by changing color from blue to red in DMSO,DMF, toluene and THF while changing color from blue to yellow in $CHCl_3$ (Figure 1.17).[1]

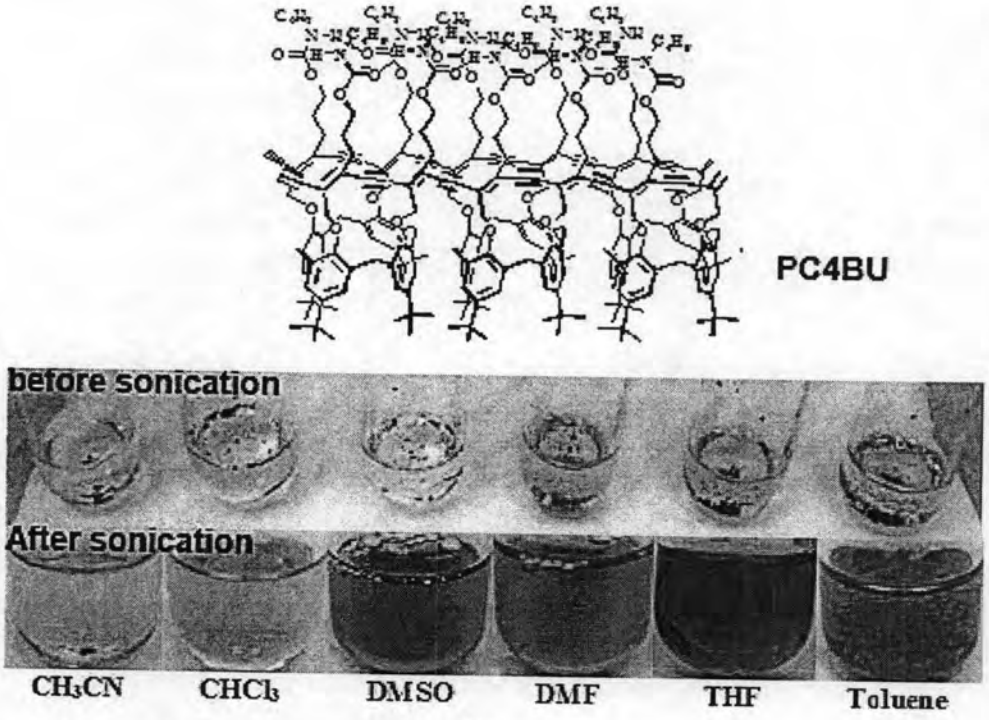

Figure 1.17 Solvatochromism of PC4BU.[1]

Kim and coworkers invented polydiacetylene embedded electrospun fiber mat to make a colorimetric sensor array for volatile organic compounds. The electrospun mat that produce from four different types of diacetylene monomer showed unique color response pattern in each organic volatile (Figure 1.18).[8]

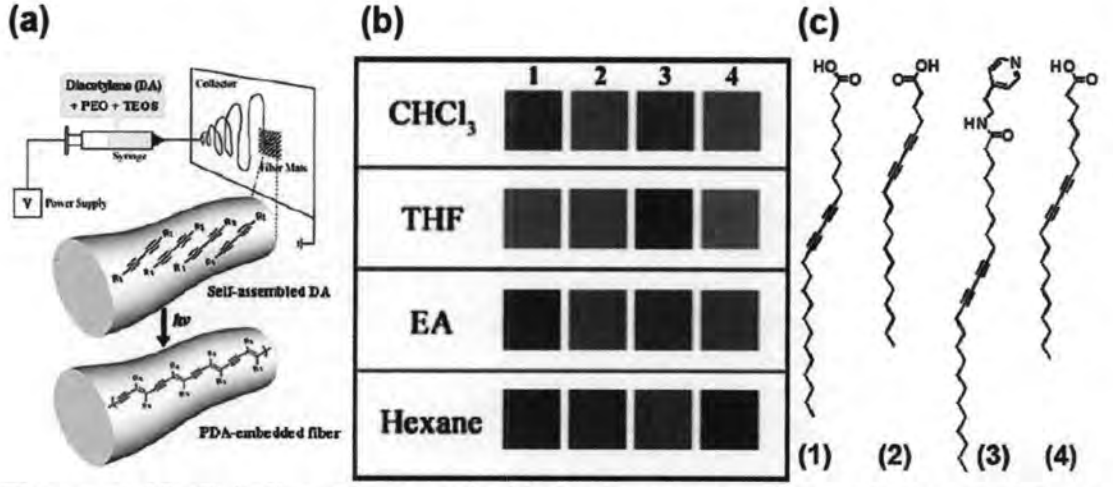

Figure 1.18 Polydiacetylene embedded electrospun fiber. a) preparation of electrospun fiber, b) polydiacetylene embedded electrospun mat sensor array after exposure to volatile organic compounds, c) four types of diacetylene monomer.[8]

Alkalinochromism and Acidochromism

Alkalinochromism and acidochromism were chromic properties that material can change color by deprotonation in case of alkalinochromism and protonation in case of acidochromism. There was reported by Kew that the poly10,12 tricosadiynoic acid (poly(TCDA)) exhibited the alkalinochromic by changing its color from blue to red during the addition of basic solution. The titration of poly(TCDA) vesicle by NaOH solution produced a red vesicle. The pKa of polydiacetylene was in the range of 9.5-9.9 depending on a kind of metal hydroxide (Figure 1.19).[34] The author proposed the color transition mechanism which started by 1) deprotonation of carboxylic proton, 2) metal ion binding with the carboxylate anion and 3) the alkyl chain changed conformation which caused a color change of poly(TCDA).

In the alkalinochromism and acidochromism, the role of head group of diacetylene monomer had directly impact on the colorimetric transition as reported by Cheng and coworkers. A series of amino acid-derivatized 10, 12-pentacosadiynoic acid had been synthesized and studied the colorimetric response in various pH solutions. The result showed a different colorimetric response based on type of head groups. For example, Glu-PDA which has dicarboxylic head group changed the color from blue to red at pH 6 by deprotonation of the carboxylic group. The protonation of the tertiary amine head group of DMAP-PDA changed its color at pH 5. His-PDA which has both of carboxylic and imidazole changed the color at pH below 0 and pH 8-9 (Figure 1.20).[35]

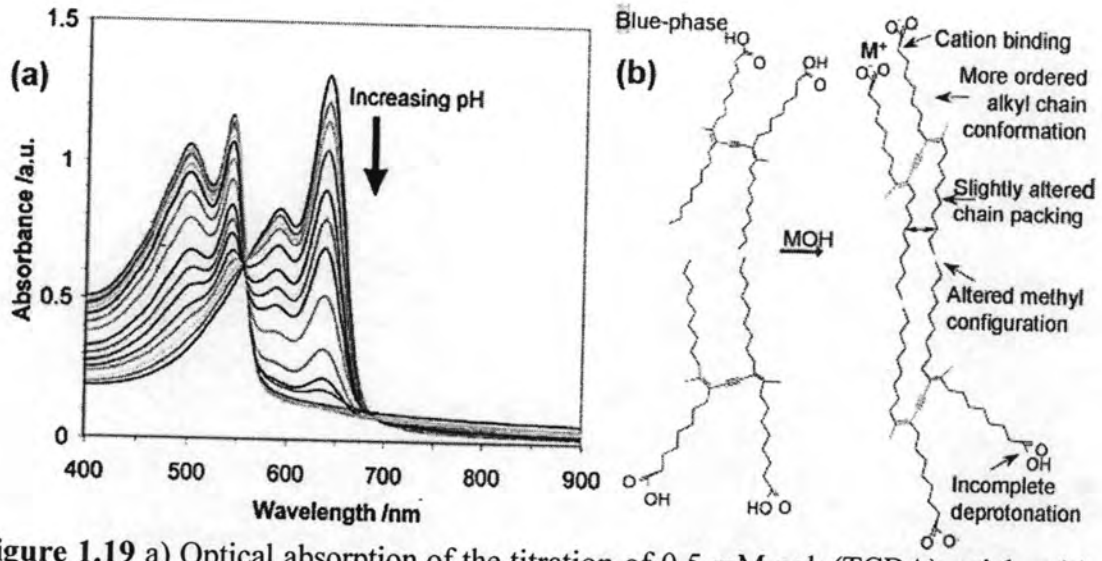

Figure 1.19 a) Optical absorption of the titration of 0.5 mM poly(TCDA)vesicle with 0.1 N NaOH solution, b) propose of color transition mechanism by alkalinochromism.[34]

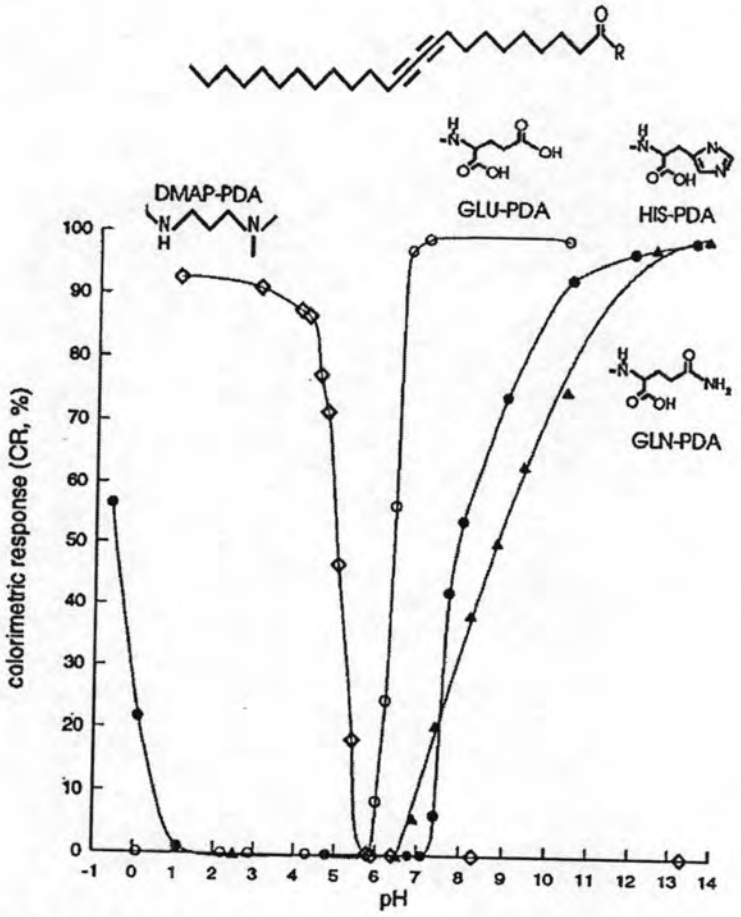

Figure 1.20 Colorimetric response (%CR) of amino acid terminate polydiacetylene vesicle as a function of solution pH.[35]

Reversible alkalinochromism and acidochromism was reported by Charych and coworkers. The hydrazine functionalized polydiacetylene vesicle exhibited a

reversible color change when the pH of the surrounding aqueous medium is cycled between acidic and basic conditions (Figure 1.21).[36] An ab initio computer model observed a protonated hydrazide had stronger intermolecular interaction than normal hydrazide derivative (Figure 1.22).

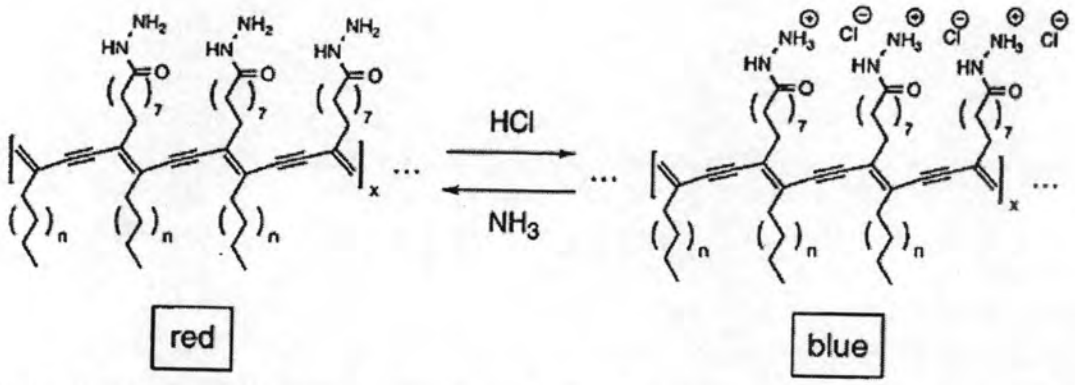

Figure 1.21 Reversible of alkalinochromic and acidochromic of hydrazine modified polydiacetylene.[36]

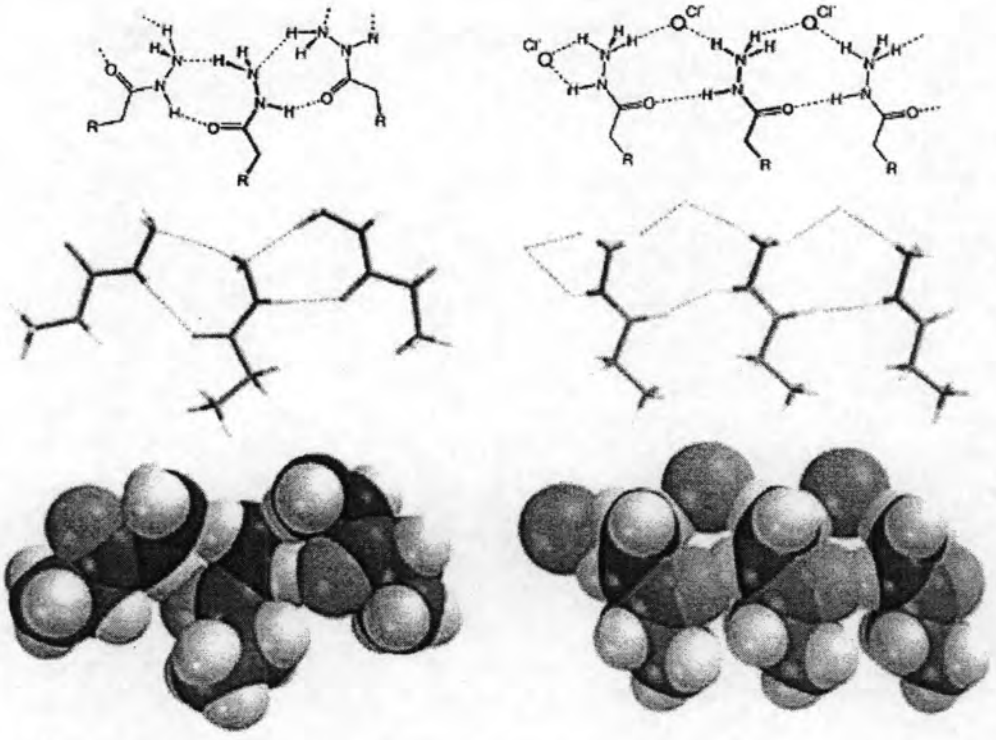

Figure 1.22 Computer models of geometry-optimized trimers for (left) neutral propanohydrazide, (right) the HCl salt of propanohydrazide.[36]

1.5 Application of PDA vesicles as sensing materials

There are two approaches to use polydiacetylene as sensing material which are 1) mimicking of the lipid membrane structure by modification receptor site on diacetylene monomer which produce an interaction of biological molecule and 2) incorporating phopholipid into the membrane structure and investigated membrane permeation. This polymer has previously been shown to undergo chromatic transitions in response to virus or bacteria. [37-39] Charych and coworkers investigated an influenza virus sensor by incorporation the virus receptor ligand into polydiacetylene Langmuir film. The influenza virus can then bind with sialic acid residues on the surface results in a blue to red color transition (Figure 1.23).[40]

By changing from film to solution, an influenza virus sensor was prepared in vesicle form. The vesicle can identify the influenza virus sample in a microliter well plate (Figure 1.24).[41]

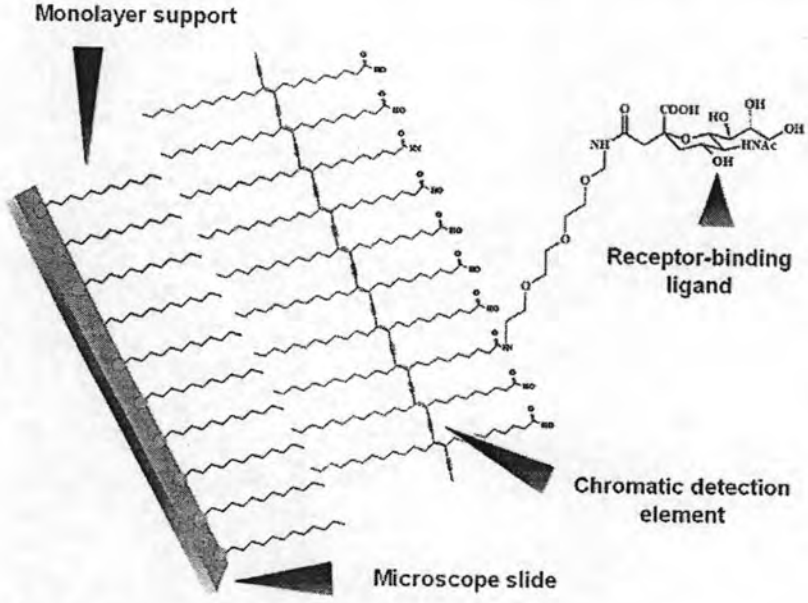

Figure 1.23 Schematic diagram of the influenza virus sensor.[40]

In the case of bacteria sensor, an incorporation of GM_1 into artificial membrane as polydiacetylene created a biosensor for cholera toxin. The Cholera toxin is a neurotoxin produced from bacteria *Escherichia Coli.* The toxin had specific binding with a carbohydrate receptor GM_1 (Figures 1.25 and 1.26).[4] The polydiacetylene changed color from blue to pink after exposure to toxin at least 40 ppm.

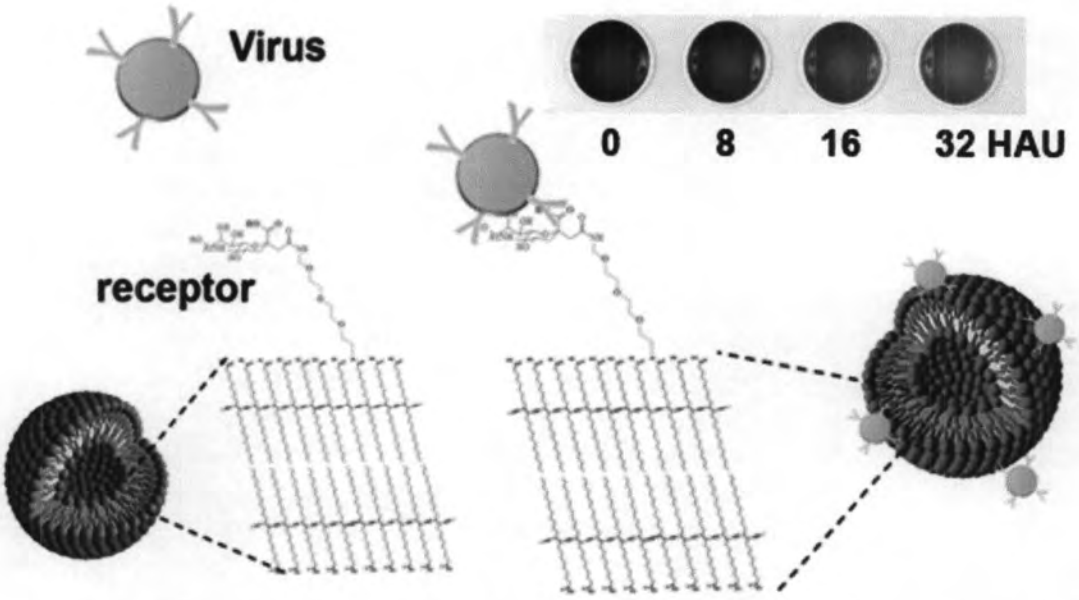

Figure 1.24 Illustrate a diagram of the influenza virus sensor based on polydiacetylene vesicle.[41]

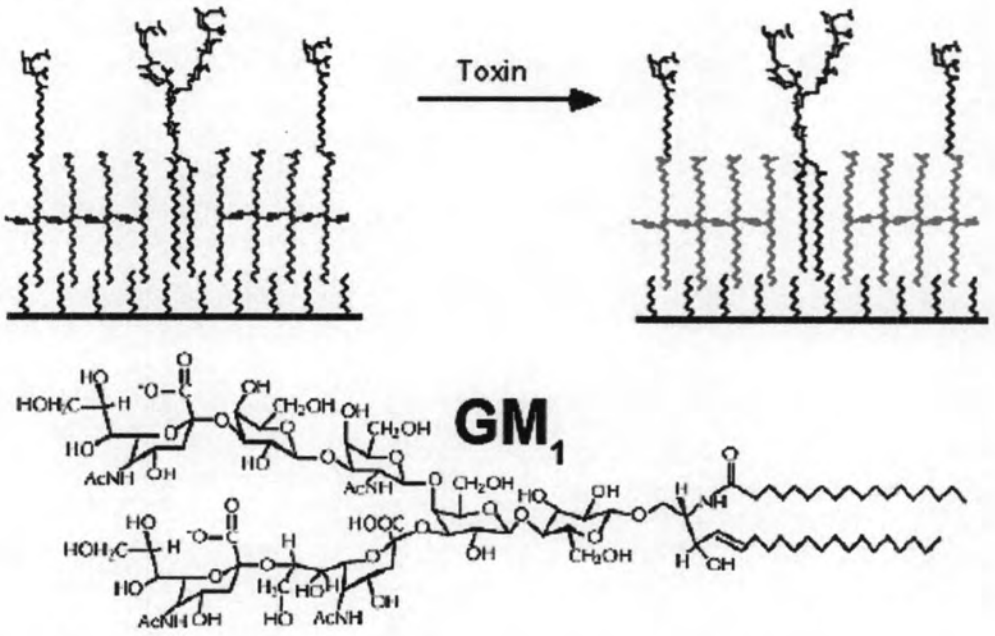

Figure 1.25 A diagram of the cholera toxin sensor based on polydiacetylene film.[41]

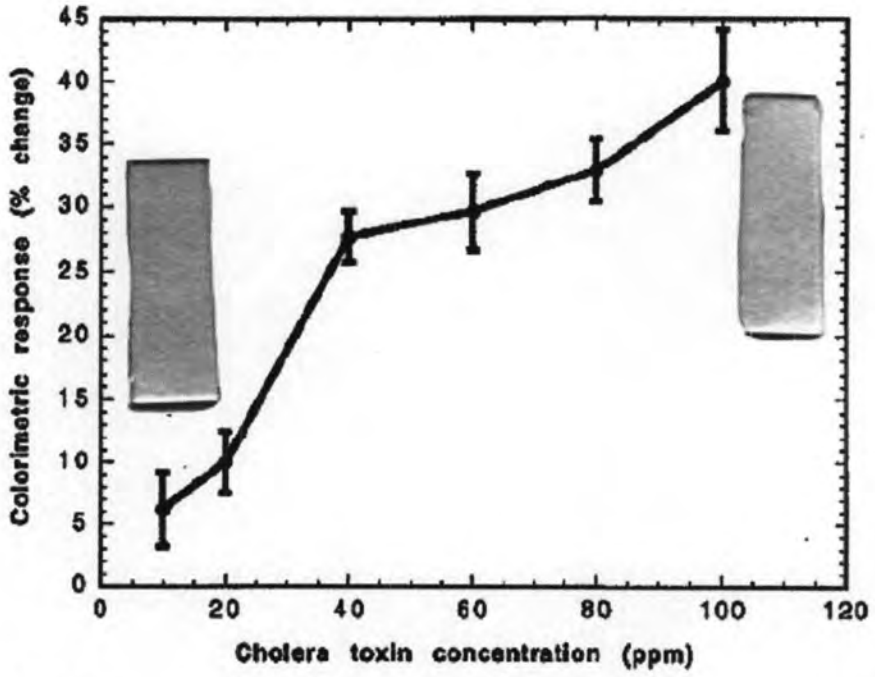

Figure 1.26 Colorimetric response of GM_1 incorporated polydiacetylene Langmuir film as a function of cholera toxin concentration.[41]

Development of artificial membrane from polydiacetylene as membrane permeable assay was investigated by Jalinek and coworkers A mixing phopholipid and polydiacetylene vesicle can use for detected an interaction of biological molecule that can bind with phospholipid or penetrate through lipid membrane (Figure 1.27).[42-43] The method can apply in drug screening, cosmetic and other permeable assay.

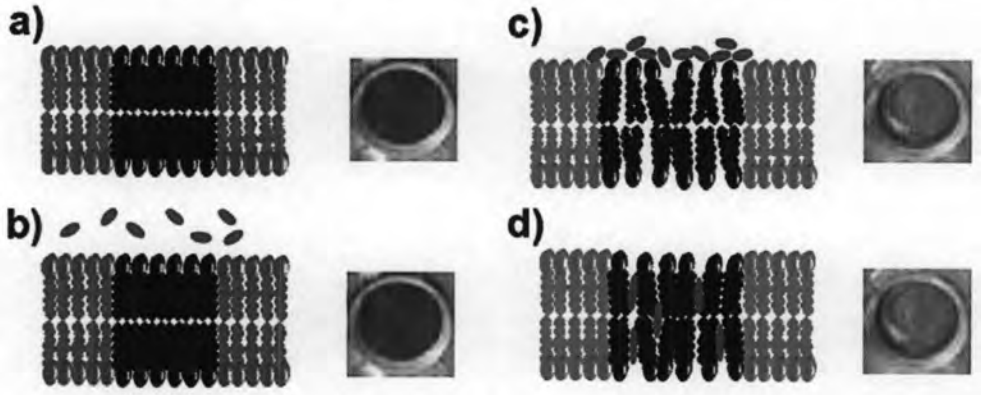

Figure 1.27 Schematic descriptions of the phopholipid/PDA vesicle and the chromatic transition of artificial membrane. a) initially prepared PDA vesicle. The phopholipid are shown in black, PDA in blue, b) The analyte molecule do not interact with the bilayer, c) blue-to-red transition induced by analyte compound bound at the bilayer surface, d) the analyte compound penetrating into the phospholipid bilayer.[42]

The chromatic response induced by penetration was used in bacteria pore forming toxin sensor.[44] Mixed vesicle composed of glycine terminated PCDA (Gly-PCDA), phosphatidyl cholinediyne (PC-DIYNE) and cholesterol (CHO) was incubated with steptrolysin (SLO) which are pore forming toxin. After incubated mixed vesicle with 500 HU/mL of SLO, the color of vesicle change to purple. The TEM picture showed the formation of pore which cause by toxin on the vesicle surface (Figure 1.28).

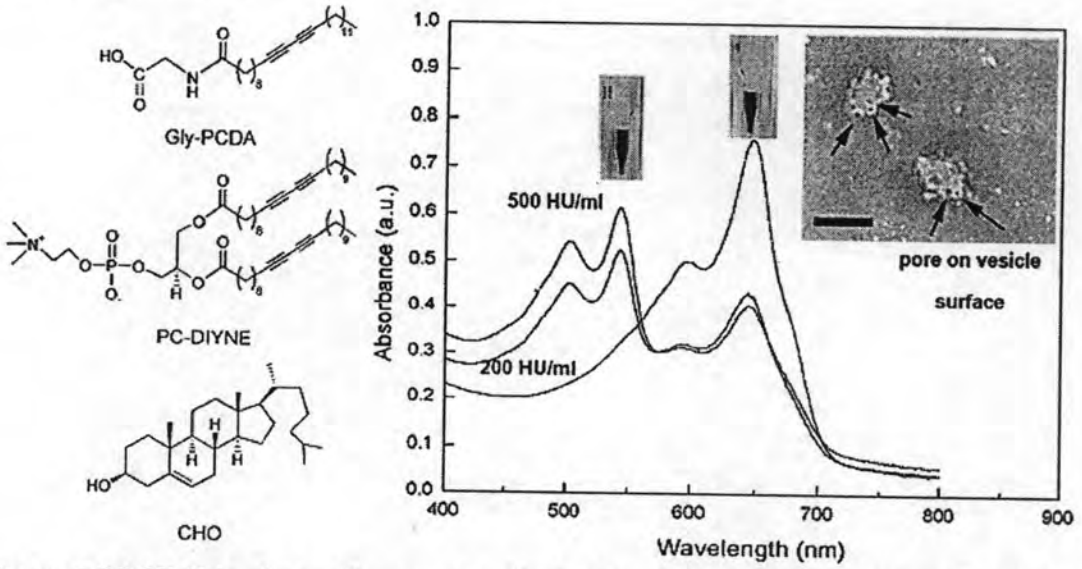

Figure 1.28 Visible absorption spectra of mixed vesicle after incubated with SLO at 200 and 500 HU/mL. The TEM picture shows the pore formation on the vesicle surface.[44]

For protein detection, three different types of calixarine were embedded into mixed phosphatidylcholine (PC) and PDA vesicle. The charge on the vesicle surface was modified by charge of calixarene used. The electrostatic interaction between vesicle and protein analyte had different colorimetric response depend on the isoelectric point of protein. The fingerprinting of colorimetric response from four types of vesicle was used to recognize a kind of protein (Figure 1.29 and 1.30).[45]

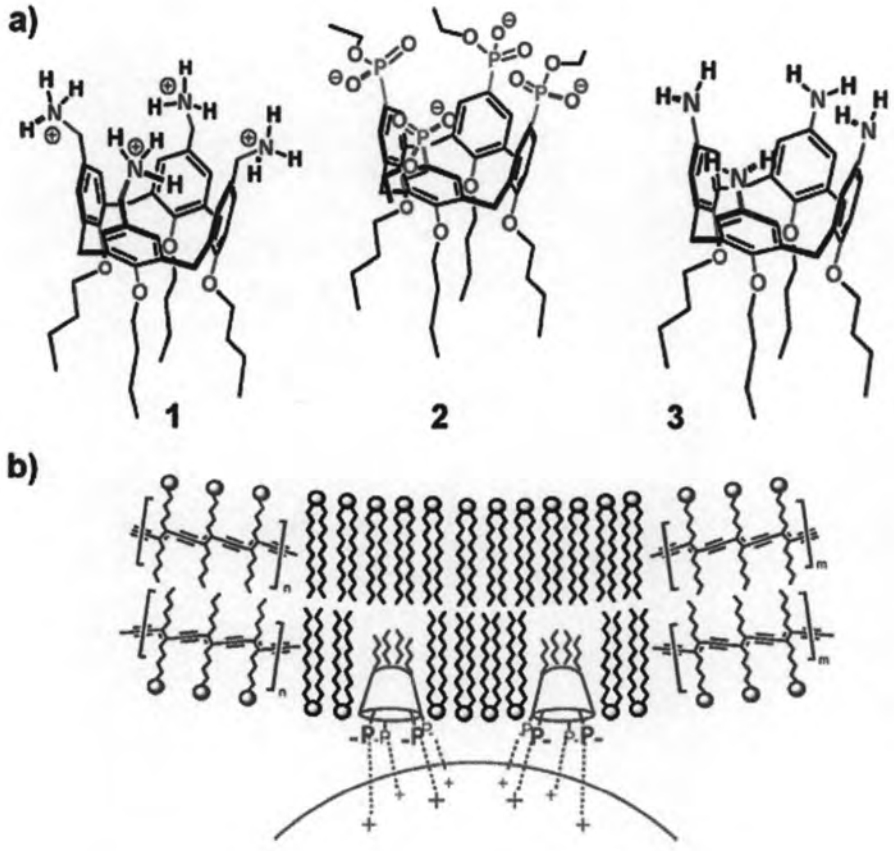

Figure 1.29 a) structure of three calixarenes use in protein sensor, b) schematic structure of the receptor/phospholipid/PDA assemblies.[45]

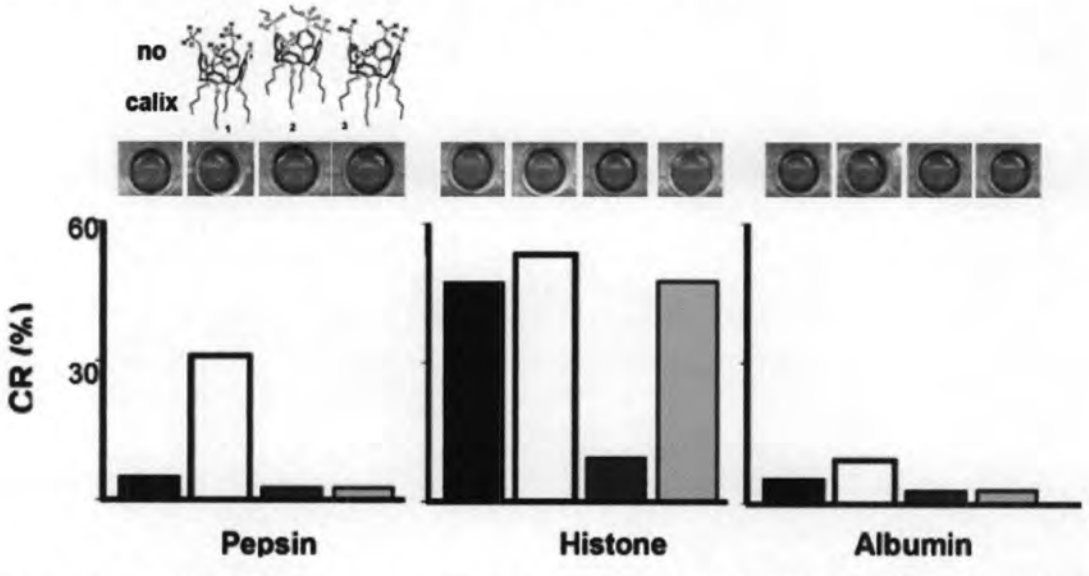

Figure 1.30 Colorimetric response of the receptor/PC/PDA system to protein addition. From left to right, (black column) PC/PCDA vesicle without calix receptor, (white column) embedded of calix 1 with PC/PCDA vesicle, (black column) embedded of calix 2 with PC/PCDA vesicle, (gray column) embedded of calix 3 with PC/PCDA vesicle.[45]

In the case of chemosensor, poly 10,12-pentacosadiynoic acid vesicle poly(PCDA) has a unique effect with the α-cyclodextrin (α-CD). Kim and coworkers investigated the specific inclusion complex of α-CD with a linear alkyl chain of aliphatic side chain. The polymer changed from blue to red after inclusion. The β-and γ-cyclodextrins did not form a complex with poly(PCDA) vesicle (Figure 1.31).[46]

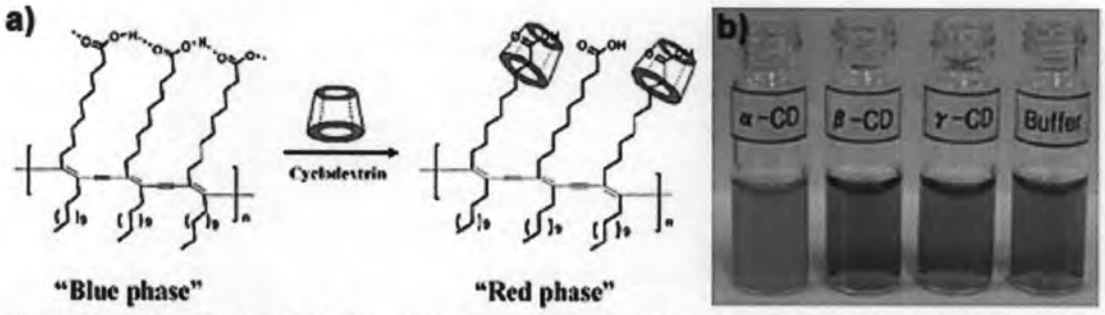

Figure 1.31 a) schematic diagram of an inclusion complex of polydiacetylene vesicle and α-CD, b) selectivity of inclusion complex with α-CD.[46]

For the ion selective color sensor, Jelinek and coworkers investigated a supramolecular assemblies of vesicles composed of ionophore and phospholipid (PC) embedded in polymerized diacetylene. Ionophore is a protein that can bind and transport a specific cation pass through lipid membrane. Mixing of ionophore embedded polydiacetylene vesicle are shown to undergo color change from blue to red after additional of specific cation. The color transition of the vesicle is directly related to the binding of cations with the ionophore. For example, a bacteria ionophore namely valinomycin has a specific binding with $Rb^+ > K^+ > Cs^+$. After additional of Rb^+ in a valinomycin embedded vesicle, the color of vesicle change from blue to red while additional of Li^+ and Na^+ ion did not change (Figure 1.32).[47] Titration of the cation selective sensor showed a detection limit less than 0.5 mM (Figure 1.33).

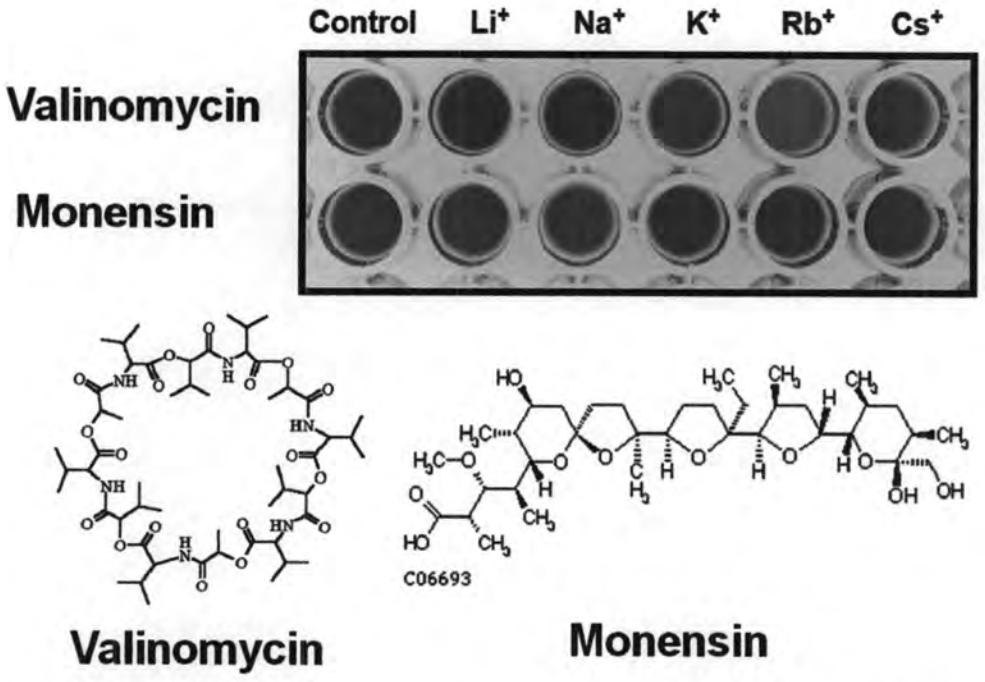

Figure 1.32 Picture of a well plate containing: valinomycin/PC/PDA and Monensin/PC/PDA solutions after addition of ions.[47]

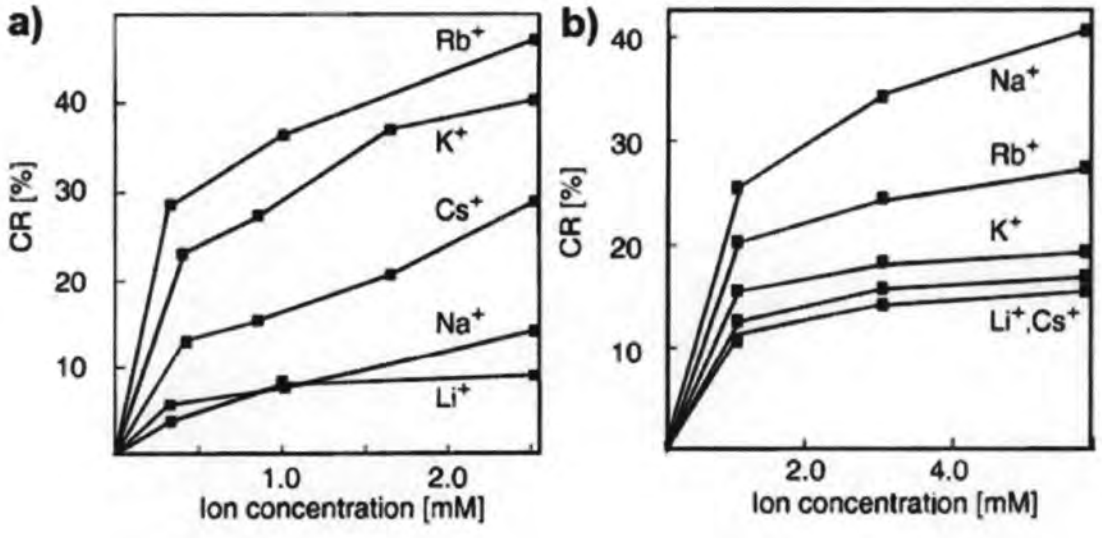

Figure 1.33 Titration of the ion selective cation sensor with alkaline ion, a) valinomycin/PC/PCDA, b) monensin/PC/PDA.[47]

1.6 Interaction of metal ion with functionalized lipid membrane

Chemical recognition events on lipid membranes are the initiating steps toward cellular signaling. Particularly interesting are the recognition phenomena that occur with the cell membrane which pathogenic agents must be rapidly and precisely distinguished from innocuous. Although some lipids have complex functionality for

recognition of specific ligands, even the simplest structures also selectively bind metal ions and ligand. Changed in the lipid's phase transition temperature and aggregational state in the membrane can lead to marked changes in the membrane structure. The metal ion interaction with membrane can be exploited as metal ion sensor material.

In 2002, Sasaki and coworker studied an interaction of crown six ether modified lipid in phopholipid membrane with some heavy metal ion such as Zn^{2+}, Hg^{2+}, Pb^{2+}. The fluorophore "pyrene" was connected with crown ether lipid to use as a fluorescence probe(Figure 1.34 a). The results from fluorescence emission spectroscopy and AFM image indicated an aggregation of crown ether lipid on the phospholipid membrane. Recognition of lead ion by the crown ether cause the lipid's headgroup to become cationic resulting in electrostatic repulsion between Pb^{2+} bound lipid (Figure 1.34).[48]

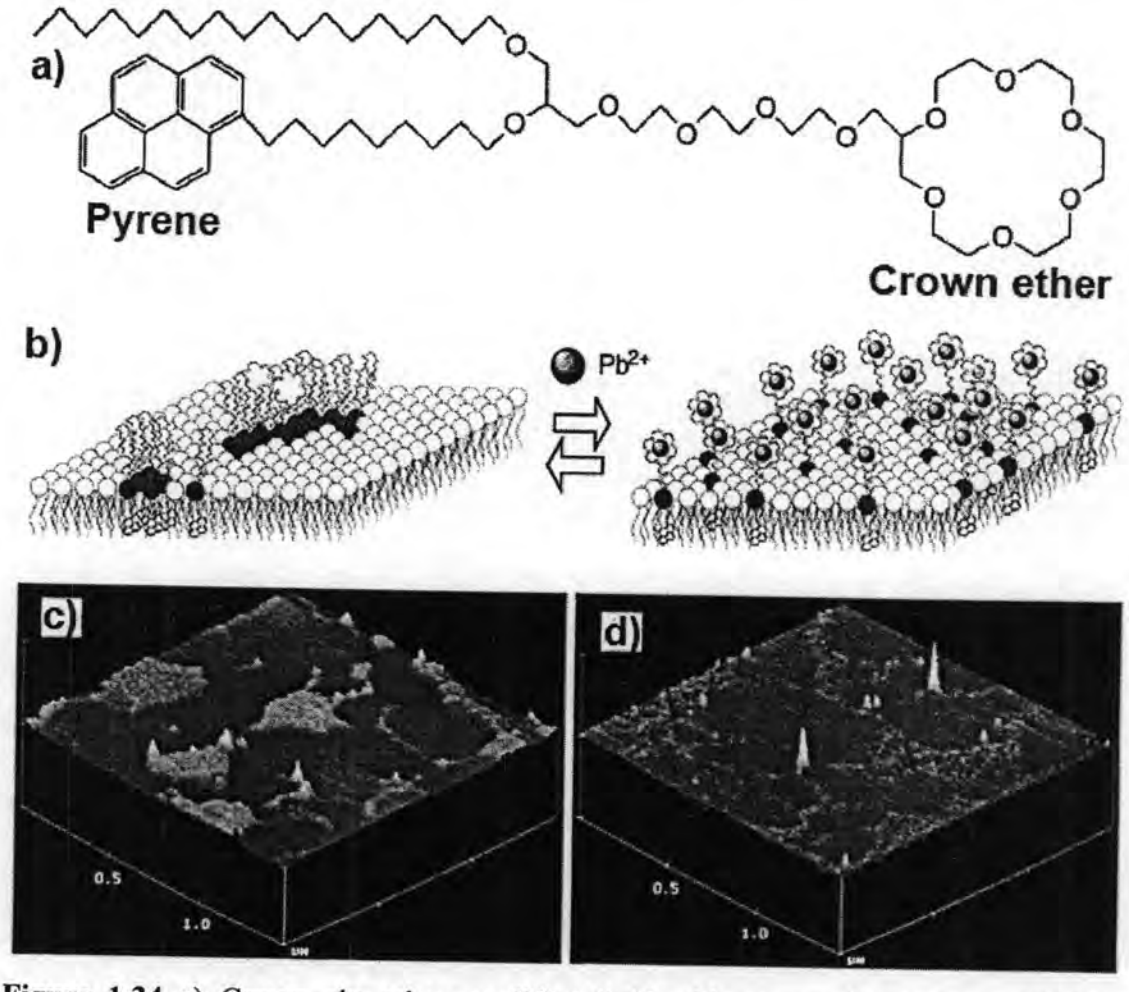

Figure 1.34 a) Crown six ether modified lipid with pyrene fluorophore, b) The response of crown ether modified lipid in phopholipid membrane with Pb^{2+}, c) an AFM image of bilayer before addition of Pb^{2+}, d) After addition of Pb^{2+} the aggregation was disappear.[48]

Changing the receptor from crown ether to another cation receptor ligand gave similar results which the receptor lipid molecule changed from aggregation to dispersion by recognition the matal ion (Figure 1.35).[49] The bilayer response to select metal ions occurs as a change in ratio of the pyrene excimer to monomer fluorescence intensities (E/M). The eximer occurred by aggregation of pyrenes which shifted the emission of pyrenes to 470 nm whereas emisision of pyrene monomer was 376 nm. Selectivity of metal ion recognition depended on the receptor groups for example, PSOH, PSMA and PSIDA have selectivity with Fe^{3+} higher than other metals salt. PSBiPy has selectivity with Cu^{2+} than Fe^{3+}.

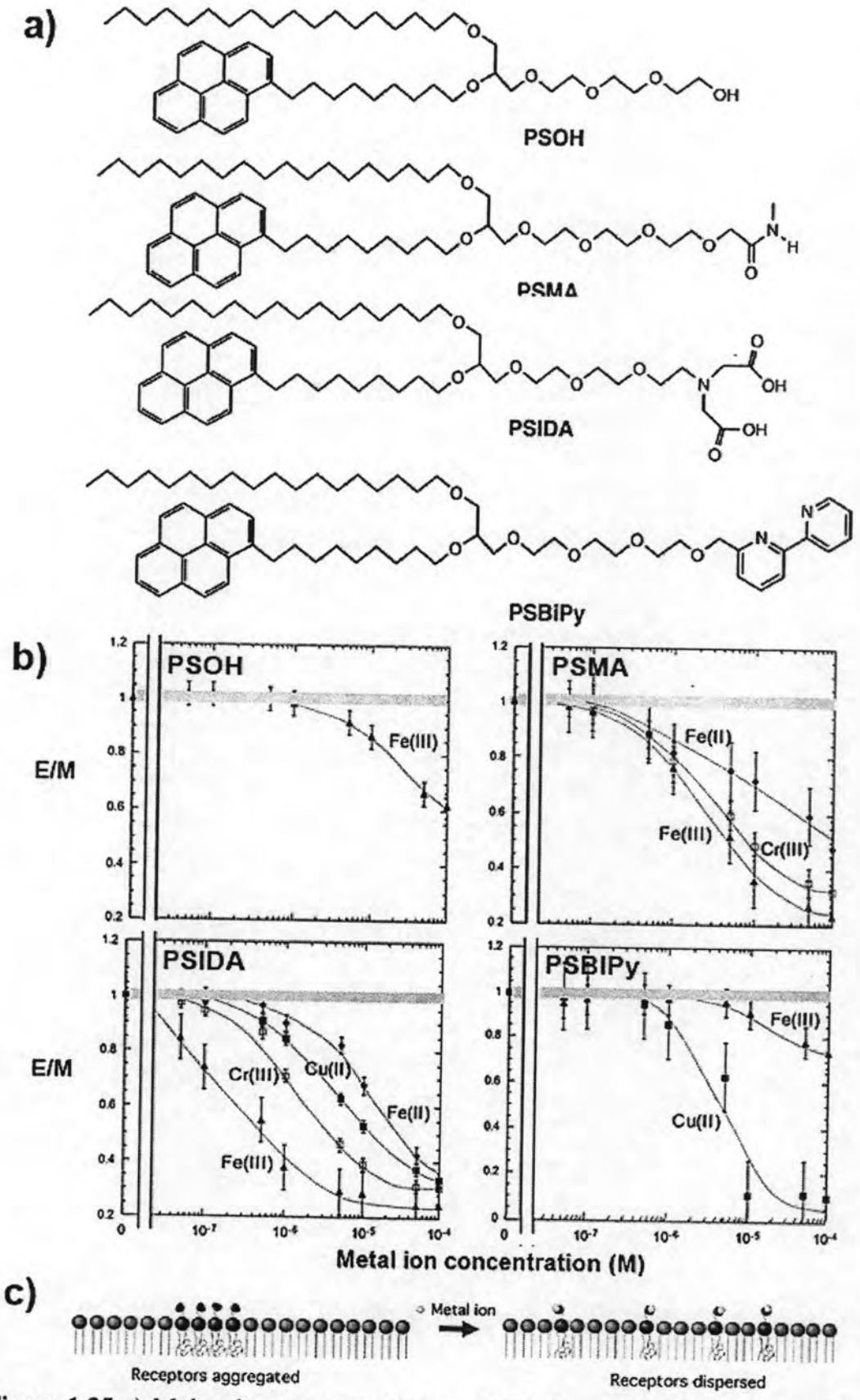

Figure 1.35 a) Molecular structure of pyrene-labeled receptor lipid, b) Fluorescence response (E=excimer, M= monomer,) vs metal ion concentration of 5% receptor/phopholipid bilayer, c) Illustration of the dispersion of initially aggregated receptor-lipids in a phospholipid membrane upon metal ion recognition.[49]

In the metal ion membrane recognition, there has possibility that, lipid membrane can aggregate wtih metal ion especially divalent and trivalent metal ion. In the case of PSIDA/phopholipid vesicle, the columnar lipid bilayer stack was found after additional Cu^{2+}. Coordination of Cu^{2+} complex with iminodiacetic acid produce the columnar lipid bilayer (Figure 1.36).[50]

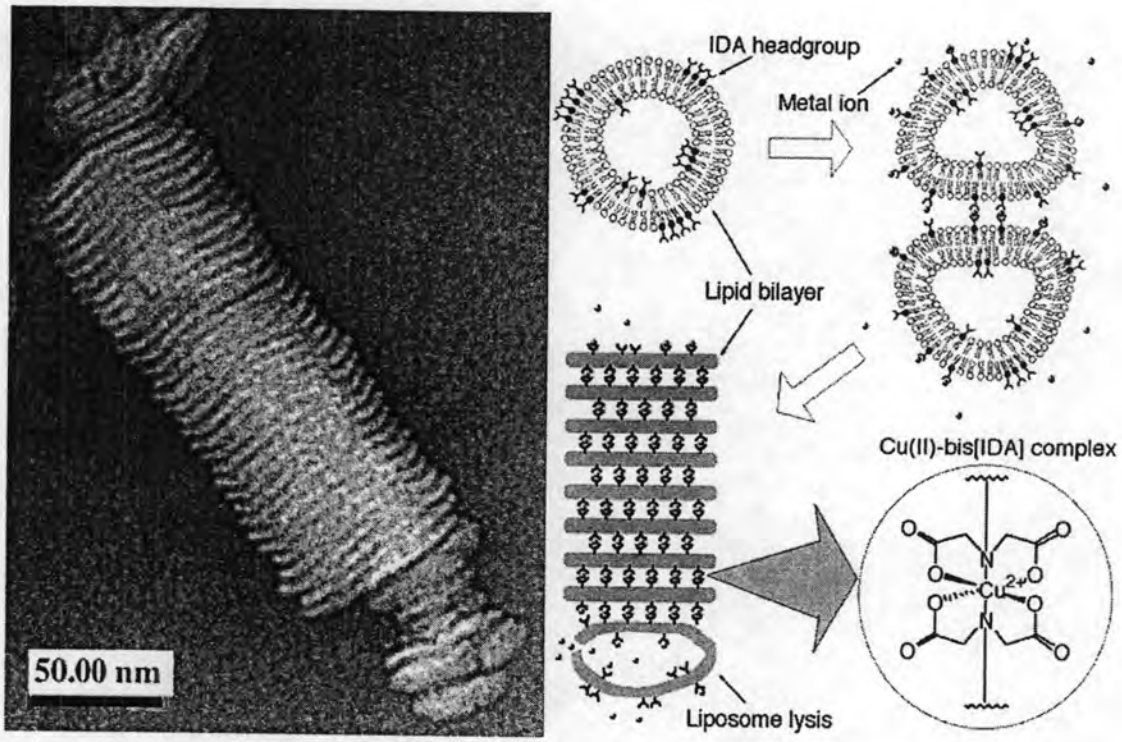

Figure 1.36 (left) TEM image of a self-assembled, columnar structured lipid bilayer stack that forms after Cu^{2+} addition to vesicle of 5%PSIDA/phospholipid, (right) proposed mechanism of lipid bilayer stack formation.[50]

1.7 Objectives and scope of thesis

The objectives of this thesis is

1) Synthesis and characterization of polydiacetylene vesicle.
2) Study of the chromic transition mechanism of solvatochromism, alkalinochromism and thermochromism.
3) Preparation of polyelectrolyte multilayers film from polydiacetylene vesicle which retained chromic properties by using layer-by-layer method.
4) Development of metal ion sensor from polydiacetylene vesicle.
5) Preparation of temperature indicating label from polydiacetylene.