

# Fluorescent Property of Gamma-irradiated Silver/ Poly(acrylic acid) Nanocomposites

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Fluorescent silver nanoparticles/nanoclusters were produced from AgNO<sub>3</sub> precursor upon its encapsulation with Poly(Acrylic Acid) and stabilization via free radical reduction using gamma-irradiation. Fluorescence spectra showed highest emission peaks around 730 nm and 775 nm when fixed wavelengths of 300 nm and 350 nm, respectively, were used to excite the solutions. Low molecular weight polymer (Ag450) showed higher intensity of emission than high molecular weight (Ag1250). Dynamic Light Scattering analysis also revealed that low molecular weight polymer could result to a less dispersed particle distribution as compared to the high molecular weight polymer. Images of the fluorescent nanocomposites were captured using Fluorescence Microscopy. Samples also showed significant changes in color after each excitation indicating occurrence of photochemical reaction.

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**Keywords:** *silver/poly (acrylic acid); gamma irradiation; fluorescence microscopy; cluster formation*

## INTRODUCTION

Metal clusters are composed of few atoms and have attracted scientists for its wide application on fluorescence (Xu, et al. 2010). This ability to fluoresce is due to the inter-band electronic transition from the submerged and quasi-continuum 5d band to the lowest unoccupied conduction band of the clusters (Dai, et al., 2007) (Zhen, et al., 2007). Literature also shows that fluorescence was attributed to the coupling of photo-activated clusters and silver oxides (Mali, et al., 2003). Dynamic light-dependent equilibrium underlying the creation and destruction of emissive few-atom Ag<sub>n</sub> clusters was also been reported to be the caused in the rise in static

intensity level when silver oxide films were excited (Peyser, et al., 2001). Metal clusters like that of silver have absorption characteristic bands at 275nm (Ag<sub>4</sub>), 300nm (trimer), 330nm (pentamer), and 345nm (heptamer) (Henglein, 1989). These are, however, short lived and form aggregation after several hours (Ershov, et al., 1998). These can then be stabilized by polymers via multi-coordination resulting to chemical adsorption of the polymer unto the metal surface as inferred from IR (Hirai, et al., 1985) and XPS (Wang, et al., 1989) spectra. Polymers such as Poly (methyl methacrylate) and Poly (acrylic acid) have strong affinity for silver ions using its carboxylic acid pendant

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group (Shang, et al., 2008) (Toshima, et al., 2001) and since these are FDA-approved water-soluble polymers they will have wide applications for bio-imaging. Radiolytic method is advantageous for the synthesis of this fluorescent material because the method is reproducible, minimum impurities is introduced and reactions proceed homogeneously (Treguer, et al., 2005).

In the present work, we study the fluorescent property of gamma-irradiated Silver/Polyacrylic acid solution. We then characterized the solution using fluorescence measurements (spectroscopy and microscopy) and finally assessed the size of water-soluble silver clusters using atomic force microscopy.

## EXPERIMENTAL

**Materials.** Polyacrylic acid (PAA) with average molecular weights of 450 kDa and 1,250 kDa were purchased from Sigma-Aldrich. Silver nitrate ( $\text{AgNO}_3$ ) with 99.99% purity was bought from Merck. Sodium hydroxide and Sulfuric acid used to adjust the pH were bought from Mallinckrodt Chemicals. All the reagents were used as received.

**Preparation Ag/PAA Solution.** Solution of 5 mL of 1.0 mg/ml PAA (MW = 450,000 Da) maintained at pH 8.2 was added dropwise with 17.5 mL of  $2.0 \times 10^{-3}$  M  $\text{AgNO}_3$ . The resulting solution was sonicated at 44-48 kHz for a total of 10 minutes to mediate particle collision. Another set of solution was also prepared using 1,250,000 Da PAA. The solutions were deoxygenated using nitrogen gas for 15 minutes and were sealed with a rubber septum before subjecting to gamma irradiation. The samples were irradiated using a Cobalt-60 Gamma cell 220, with a rate of 3.3 kGy/hr and a total dose of  $\sim 15$  kGy housed at the Philippine Nuclear Research Institute (PNRI).

**Characterization of Irradiated Ag/PAA Solution.** *Fluorescence Spectroscopy.* Spectral scans of the nanocomposites were done using a Double-beam UV-vis Spectrophotometer

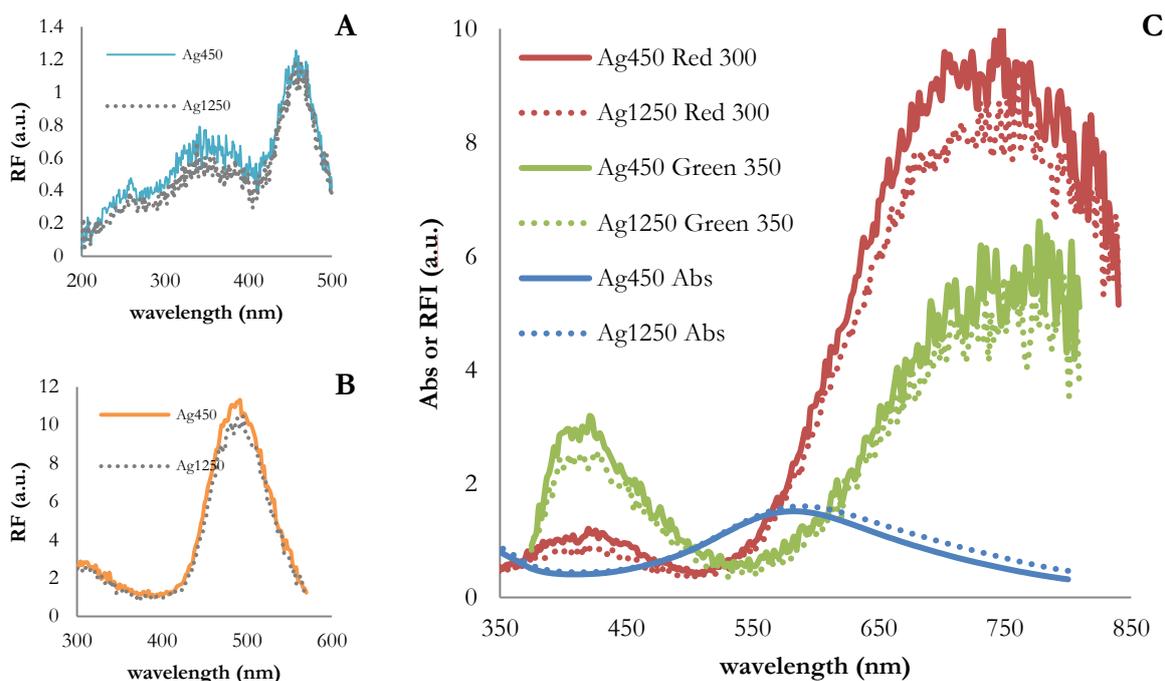
(UV-1700 Series, Shimadzu) while the fluorescence were monitored using a Multimode Spectrofluorometer (Varioskan<sup>TM</sup> Flash Multimode Reader, ThermoScientific). Microplates (Greiner Bio-One) containing 100-uL of the irradiated solution were scanned to obtain the excitation spectra (fixed wavelength scans at 520 nm and 590 nm) and was followed by another scan to obtain the emission spectra (fixed wavelength scans at 300 nm and 350 nm).

*Fluorescence Microscopy.* Images of the nanocomposites were captured using an Olympus BX-43 Fluorescence Microscope powered by a 100 W Hg-lamp. Two sets of samples were analyzed using this technique; 1. Freshly dropped on a clean glass slide 2. Dried overnight inside a dessicator. Fluorescent images were viewed using two types of filter; 1. Wide band blue excitation filter BP 460-490 nm (U-MWB2) (E1) and 2. Wide band interference green excitation filter BP 510-550 nm (U-MWG2) (E2). These were performed in dark room to avoid errors brought by stray lights.

*Particle Distribution.* The particle distribution of the nanocomposite solution were obtained using Dynamic Light Scattering Analysis (Zeta Sizer Nano ZS90, Malvern). Zetasizer performs size measurements using Dynamic light scattering (DLS) (also known as PCS – Photon Correlation Spectroscopy) that measures Brownian motion and relates this to the size of the particles. A size distribution graph shows size classes on the x-axis, while y-axis shows the relative intensity of the scattered light.

## RESULTS AND DISCUSSION

**Absorption Spectrum.** Clear solution of Ag/PAA changed to dark blue color after gamma irradiation as shown in Figure 1 producing an absorption broad band at 590 nm (blue band). The solution did not exhibit the known silver surface plasmon band around 400 nm, which is also called the Mie resonance. This then indicates that silver nanoparticles were not formed. Instead, the absorbance at 590 nm corresponds to the



**Figure 1.** Excitation spectra obtained from the fixed wavelength scans at (A) 520 nm and (B) 590 nm. Excitation wavelengths obtained are 350 nm and 300 nm respectively. (B) Comparison of absorption (blue) and emission spectra obtained from the fixed wavelength scans of irradiated Ag/PAA solution of MW 450 kDa (shown as lines) and 1250 kDa (round dots) using the excitation wavelengths obtained from A (green) and B (red).

formation of silver ion clusters. It is known that the high energy deposition from gamma rays throughout the solution leads to ionization, excitation and eventually to fragmentation of highly excited molecules forming free radical species such as solvated electrons ( $e^-_{aq}$ ) which reduce metal cation to metal atom. However, reduction of  $Ag^+_{(aq)}$  to  $Ag_{(s)}$  was incomplete due to the formation of hydroxyl radicals from the cleavage of water molecules upon irradiation. This is subsequently followed by slow aggregation of isolated atoms ( $Ag_n$ ). Coalescence of neighboring water-soluble clusters is inhibited at early stages by polymers with high affinity to Ag or  $Ag^+$  through electrostatic repulsion or steric hindrance.

**Analysis of the Fluorescence Spectra (Excitation and Emission).** Excitation spectra of irradiated solution were obtained using two different fixed wavelengths  $\lambda = 520$  nm (A) and for  $\lambda = 590$  nm (B). However, two peaks were obtained from the excitation spectrum for both wavelengths. Peaks at 350 nm and 450 nm were visible from the  $\lambda = 520$

and peaks at 300 and 500 were obtained from the  $\lambda = 590$  nm. The maxima at 350 nm (for  $\lambda = 520$ ) and 300 nm (for  $\lambda = 590$ ) were used to obtain the emission spectra of the solutions since these wavelengths were similar to the absorption of charged oligomeric silver cluster (Treguer, et al., 2005). Using 300 nm (indicated as red) and 350 nm (green), the emission spectra were then obtained (Figure 1C). Using 300 nm fixed wavelength (red lines), two fluorescence bands were observed: (1) small band around 410 nm and (2) large fluorescence band around 730 nm. Almost comparable fluorescence bands were also observed using 350 nm (green lines): (1) small bands around 415 nm and (2) large band around 775 nm. However, despite showing similar wavelengths, the two emission spectra had different intensities. In the 410 nm region, red lines showed lower intensity than green lines while in 775 nm region it is the opposite, where the red lines were more intense. In a study conducted by the group of Treguer (Treguer, et al., 2005), emission peaks at 550 nm and 700 nm for the excitation at 300 nm

could be produced due to the incomplete reduction of  $\text{Ag}^+$  and the formation of  $\text{Ag}_7^{3+}$ , which is accompanied with visible formation of blue-colored solutions. These findings indicate the presence of two distinct emission centers or one center with complex excited state structure. This is a consequence of the incomplete reduction of the silver ions due to the absence of the radical scavenger. This is similar to the results of this study, which showed emissions at the 410 nm and 700 nm regions. In addition, since emission in the visible range is also observed for excitation in the plasmon resonance frequencies of the silver sol, a strong coupling between the  $\text{Ag}_n$  colloidal particles and the emissive center(s) is expected (Treguer, et al., 2005).

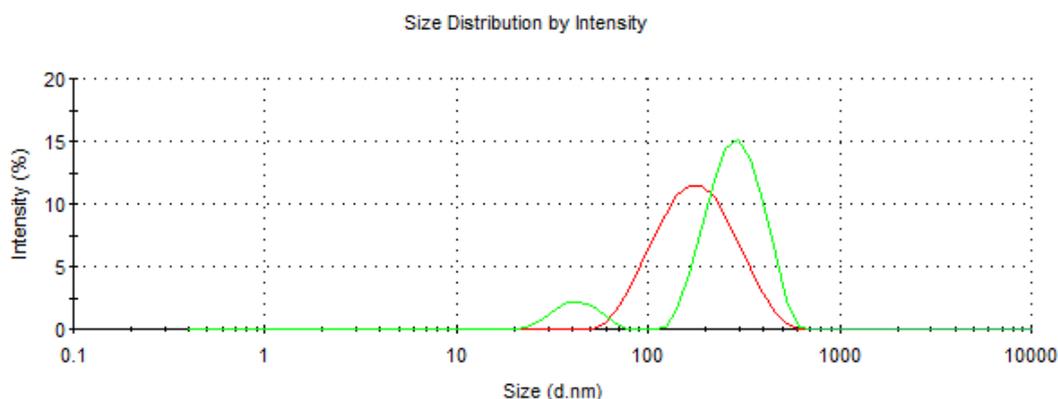
Fluorescence of sample with 450 kDa molecular weight showed lower intensity than 1,250 kDa. Low molecular weight polymer would exhibit higher fluorescence than high molecular weight using PMMA as reported by the group of Xu (Xu, et al., 2010). Lower molecular weight polymer solution is therefore a better choice to stabilize the formation of silver cluster through gamma radiation with the same rate for low molecular weight polymer. This assumption was then confirmed using Dynamic Light Scattering Analysis (DLS) via Zeta-Sizer Nano.

**Dynamic Light Scattering.** Poly(acrylic acid) with a smaller polymer molecular weight also showed better encapsulation of the nanoparticles. This was supported by the

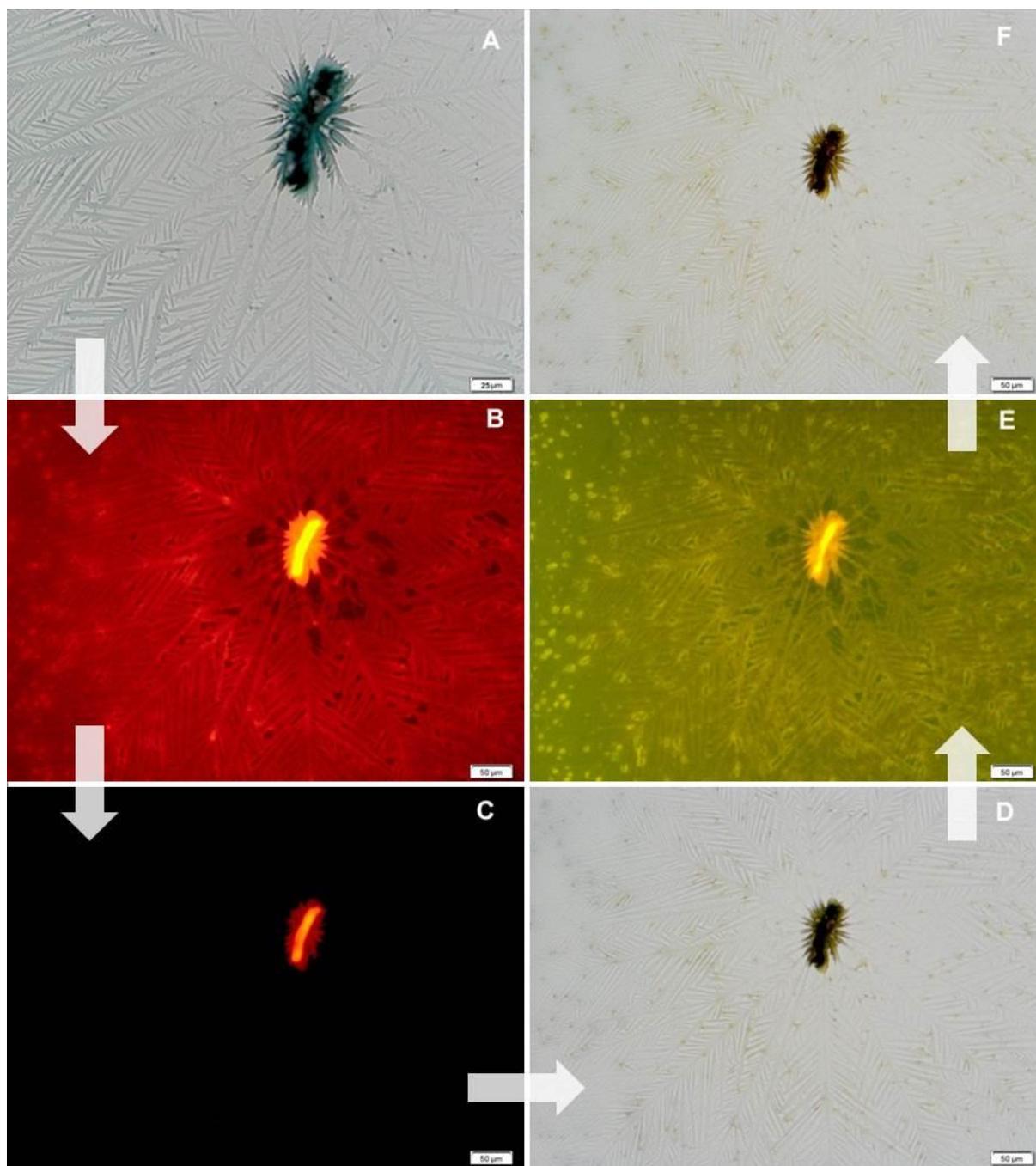
Dynamic Light Scattering (DLS) results shown in Figure 2, wherein the smaller polymer, gave rise to a better particle distribution as compared to the higher molecular weight polymer.

Average particle diameter for the two samples were 160.6 nm for Ag/PAA 450 kDa and 287.6 nm for Ag/PAA 1250 kDa. The DLS histogram also showed a single-peaked particle distribution ranging from 50 nm to 531 nm for the AgPAA 450 kDa while two peaks were visible for the AgPAA 1250 kDa, ranging from 21 nm to 78 nm and 122 nm to 615 nm respectively. The lower molecular weight polymer also showed a smaller PolyDispersity Index (PdI) with a value of 0.314 compared to the 0.538 for the higher molecular weight polymer. This means that the particles were more uniform when stabilized by the smaller polymer. The reason for these is because low MW polymer collapses into globular structures, which could perfectly encapsulate the nanoparticles while high MW polymer could form necklace structures which serve as attachments for other polymer chains, resulting to train like structures (dela Santa, 2005). Aggregation due to the interconnection of polymer chains was suspected to lower the fluorescence intensity emitted by the silver nanocluster core.

**Fluorescence Microscopy.** Fluorescence property of the nanocomposites were viewed using Fluorescence Microscopy. Excitation of the samples was done by exposing them to



**Figure 2.** DLS Histograms of the AgPAA 450 kDa (red) and AgPAA 1250 kDa (green) nanocomposite solutions.

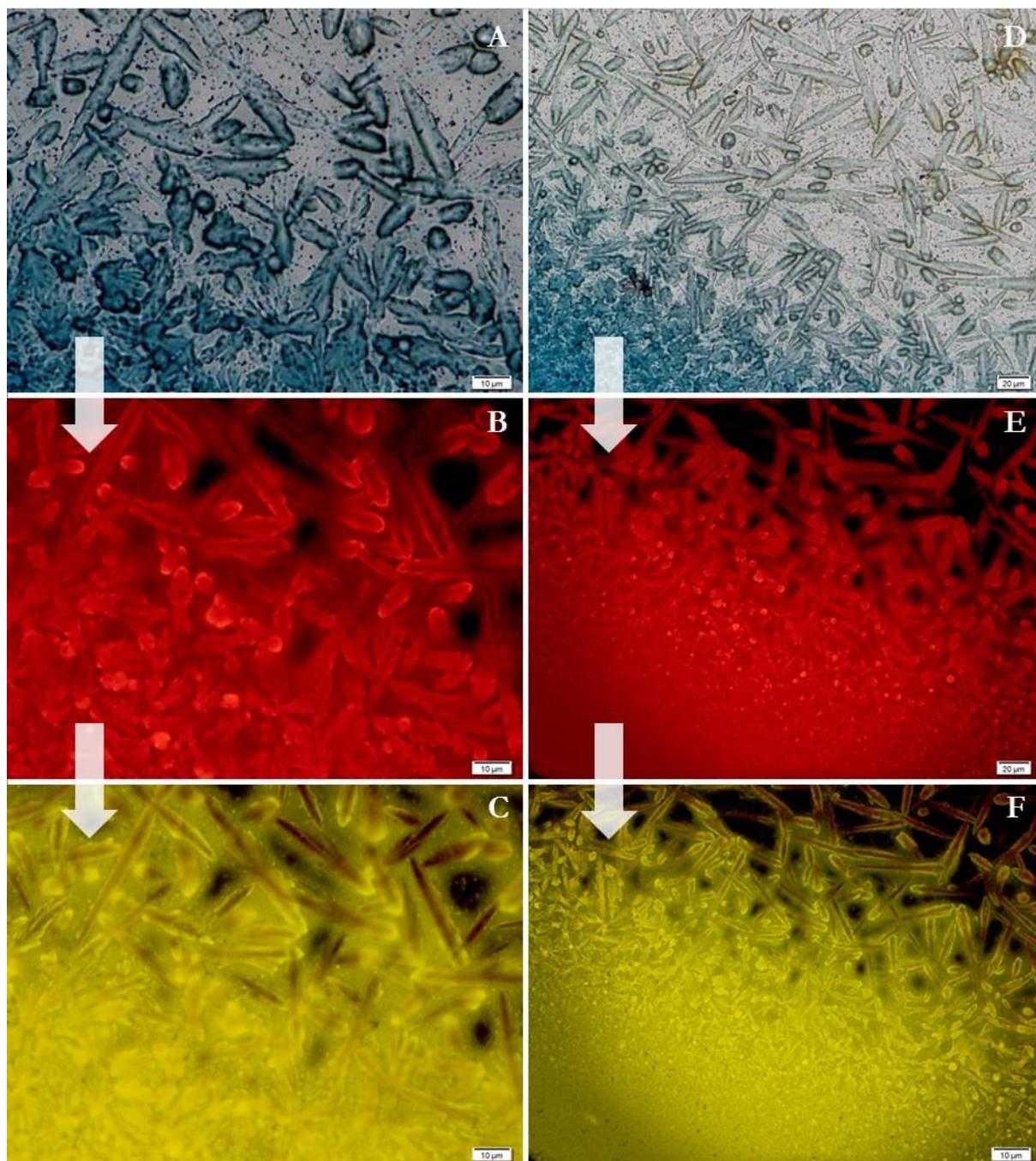


**Figure 3.** Images at the same location of dried AgPAA (MW of PAA = 450 kDa) sample. Before excitation of sample at various wavelengths, sample was imaged using light microscope (A). Fluorescence (B, C and E). Images after each fluorescence (D, F).

UV radiation while the emission of the samples was viewed by using a Fluorescence Microscope equipped with a wide band interference filter. Exposure of dried and freshly dropped nanocomposites showed photoactivity upon exposure to UV light as exhibited in Figures 3, 4, 5 and 6.

Fluorescence image of Ag/PAA (using 450 kDa) in Figure 3 (B, C and E) showed fractal-

shaped crystal formation of the nanocomposites upon drying the sample. Excitation of the samples using the two interference filter E1 and E2 (Figures 3B and 3E) showed stable fluorescence images, wherein the intensity of the emitted light from the samples lasted for more than 1 second. exposure to UV with wide band interference blue excitation filter BP 460-490 nm (U-MWB2) (E1) showed red colored images of



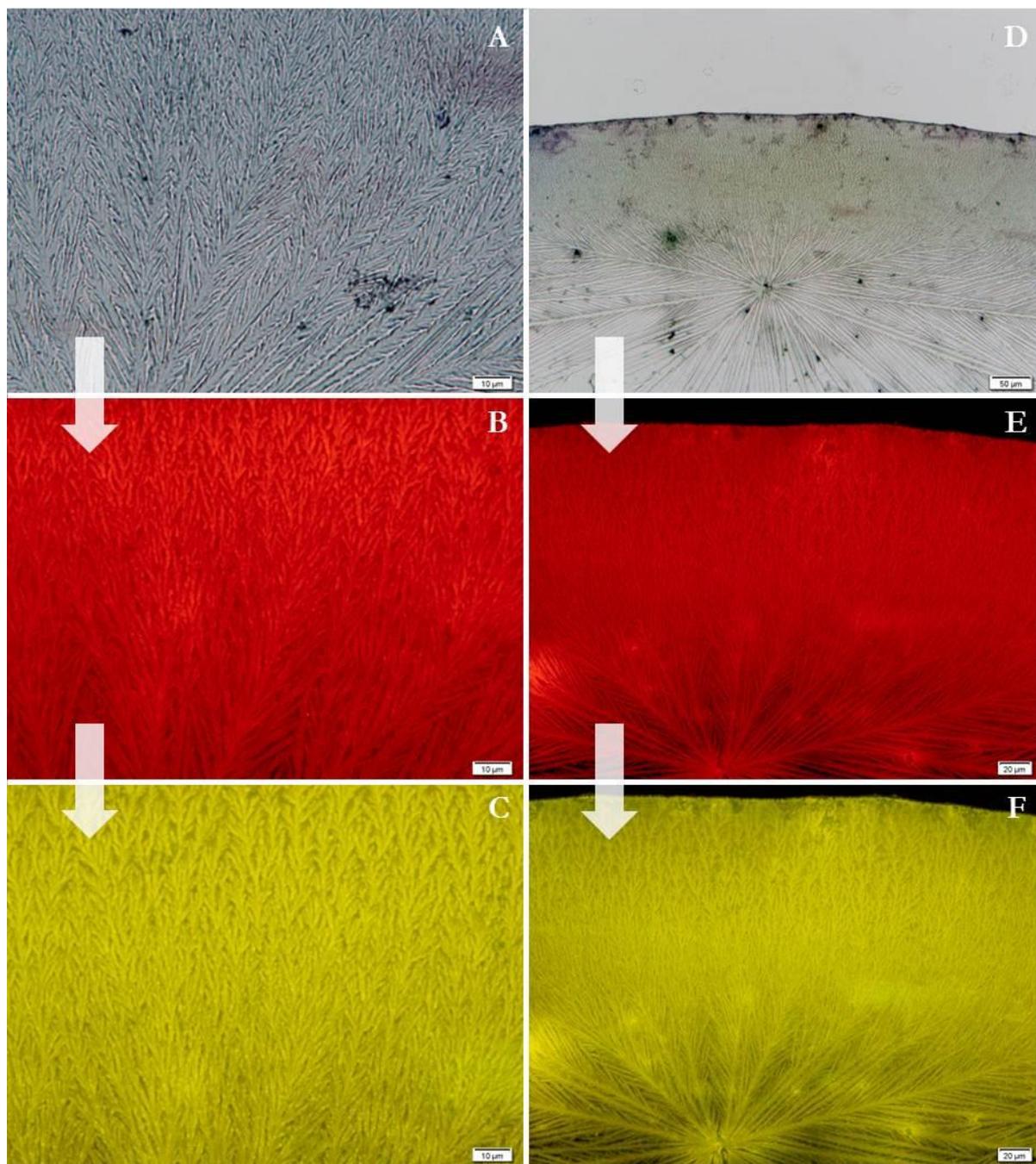
**Figure 4.** Additional fluorescence images of dried AgPAA (MW of PAA = 450 kDa) sample. Imaged using light microscope (A, D). Fluorescence using the two different excitation filters (B, C, E and F).

the nanocomposites, while exposure to the wide band interference green excitation filter BP 510-550 nm (U-MWG2) (E2) resulted to the emission of yellow colored light from the samples (Figures 3B and 3E). The massive aggregation of clusters at the center, however, exhibited bright yellow colored fluorescence for both the excitation filters.

Blue areas seen in Figure 3A image changed to dark olive green in Figure 3D and 3F which

could be the result of destruction of small clusters thereby producing photochemical reaction in the surface (bleaching of samples).

Additional images from another set of nanocomposite samples with fluorescent property presented in Figure 4B, 4C, 4E and 4F showed images of large rice-shaped nanocomposite clusters formed at the edge of the sample.



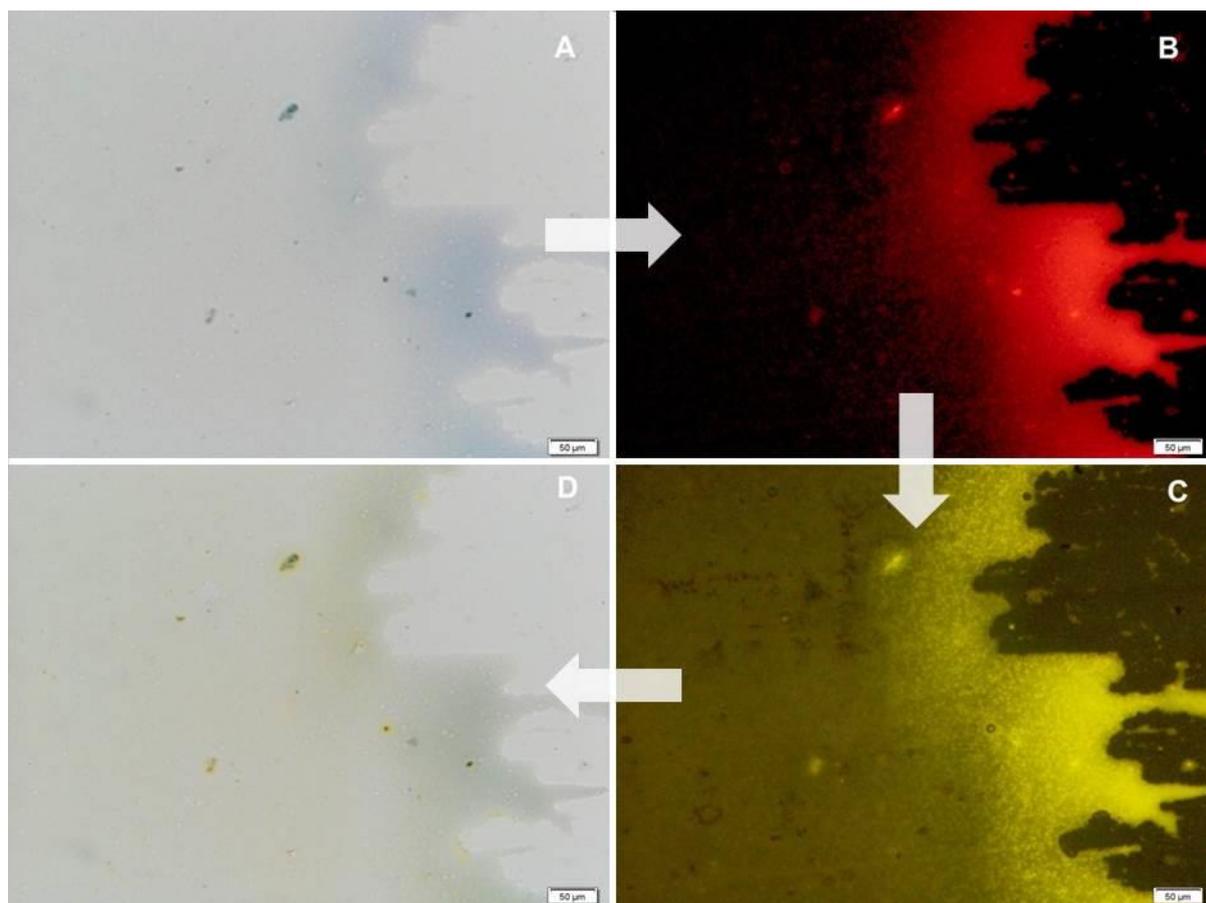
**Figure 5.** Fluorescence images of dried AgPAA (MW of PAA = 1250 kDa) sample. Imaged using light microscope (A, D). Fluorescence using the two different excitation filters (B, C, E and F).

Likewise, images of the fluorescent samples were also taken from the excitation of the AgPAA 1250 kDa nanocomposites as shown in Figure 5.

Images of freshly dropped AgPAA samples with 450 kDa (Figure 6) and 1250 kDa (Figure 7) molecular weights were also examined to show the appearance of the

clusters while exhibiting fluorescence in the solution.

The blue areas in Figure 6A and 7A changed to yellow in appearances after excitation (Figure 6D and 7E) providing more evidences of photochemical reactions on the surface.



**Figure 6.** Images at the same location of fresh dropped of AgPAA (MW of PAA = 450 kDa) sample. Before excitation of sample at various wavelength, sample was imaged first by using light microscope (A). Fluorescence (B, C). Image of after fluorescence (D).

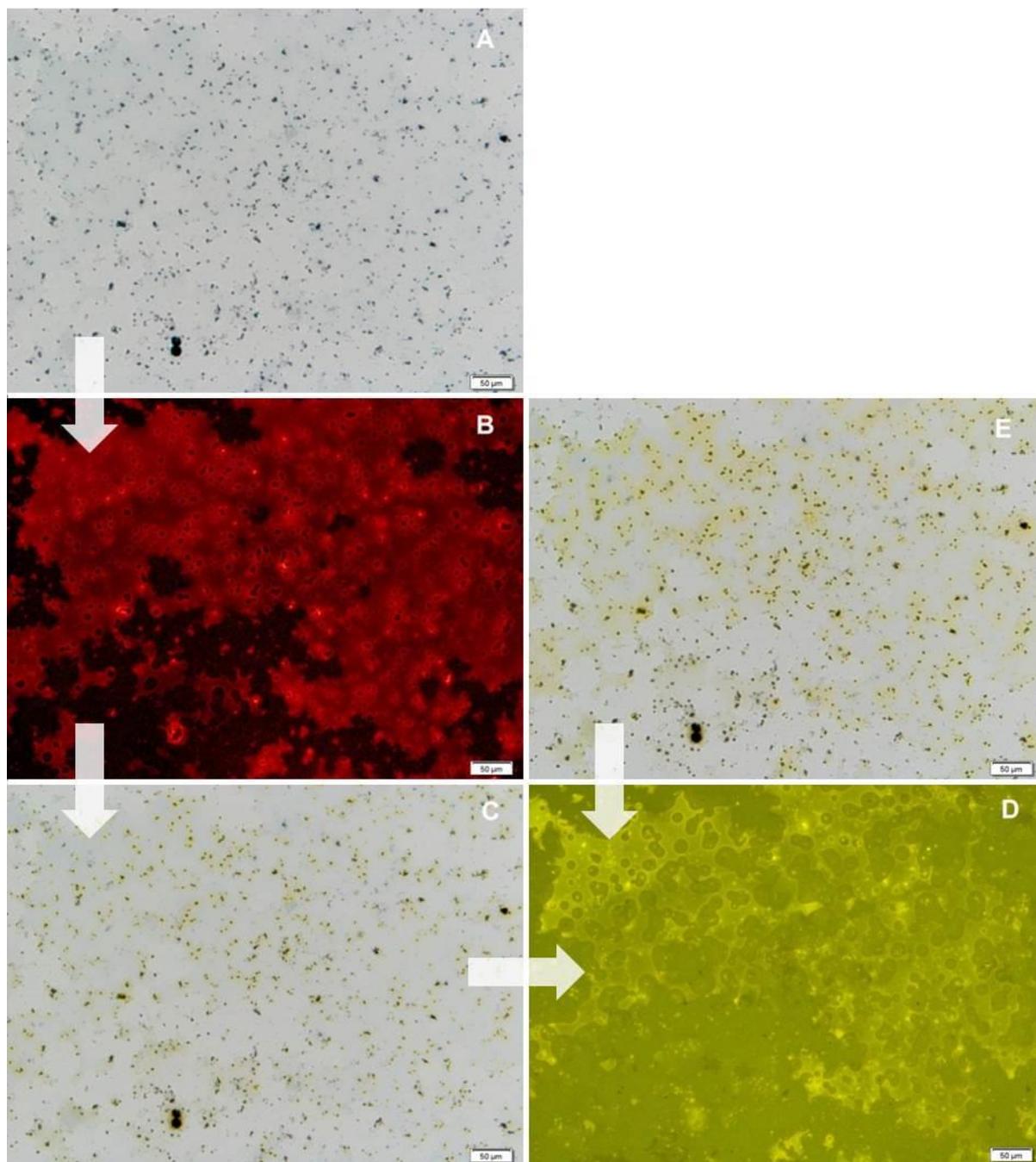
Both molecular weights also exhibited stable fluorescence using E1 and E2 unlike the dried sample in Figure 3. On one hand, very fine dots were significantly present in Figure 6C using 450 kDa polymer. These green species were attributed to the smaller Ag clusters dissolved in the solution which are also activated upon exposure to UV with E1 and E2. On the other hand, films of 1250 kDa polymer were also able to produce Ag clusters exhibiting fluorescence as imaged in Figure 7, however, sizes were in 6-7µm range. Although high molecular weight polymer contains more carboxylate group than its counterpart, silver clusters are less stabilized by this polymer in terms of rate thereby aggregation occurs as previously investigated using the Dynamic Light Scattering analysis.

## CONCLUSION

Solution of gamma-irradiated poly (acrylic acid)-stabilized silver nanoclusters gave high intensity fluorescence around the 750 nm region which resulted to the formation of water-soluble silver clusters. Samples also showed signs of photochemical reactivity after doing the fluorescence microscopy. Clusters were reactive to excitation producing changes in appearances. It is interesting to see if we could incorporate biochemical samples into these clusters and explore its interaction for a certain target molecule.

## ACKNOWLEDGEMENT

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**Figure 7.** Images at the same location of fresh dropped of AgPAA (MW of PAA = 1 250 kDa) sample. Before excitation of sample at various wavelength, sample was imaged using light microscope (A). Fluorescence (B, D). Images after each fluorescence (C, E).

## REFERENCES

Dai Y, Hu X, Wang C, Chen D, Jiang X, Zhu C, et al. Fluorescent Ag nanoclusters in glass induced by an infrared femtosecond laser. *Chem Phys Lett.* 2007; 439:81-84.

dela Santa A. Novel Polymer Spheres and Nanocomposites from the Collapse of

Polyacrylic Acid [thesis]. [Canada]: University of Toronto; 2002.

Ershov BG, Henglein A. Reduction of Ag<sup>+</sup> on polyacrylate chains in aqueous solution. *J Phys Chem B.* 1998; 102:10663-10667.

Henglein A. Nonmetallic silver clusters in aqueous solution: stabilization

- and *chemical* reactions. Chem Phys Lett. 1989; 154:473.
- Hirai H, Chawanya H, Toshima N. Colloidal palladium protected with poly(N-vinyl-2-pyrrolidone) for selective hydrogenation of cyclopentadiene. Reactive Polym. 1985; 3:127-141.
- Maali A, Cardinal T, Treguer-Delapierre M. Low-dimensional Systems and Nanostructures. Physics E. 2003; 17:559-560.
- Peysers L, Vinson A, Bartko A, Dickson R. Photoactivated Fluorescence from Individual Silver Nanoclusters. Science. 2001; 291:103-106.
- Shang L, Dong S. Facile preparation of water-soluble fluorescent silver nanoclusters using a polyelectrolyte template. Chem Commun. 2008; 9:1088-1090.
- Toshima N, Shiraishi Y, Teranishi T. Effect of additional metal ions on catalyses of polymer-stabilized metal nanoclusters. J Mol Catalysis A. 2001; 177:139-147.
- Treguer M, Rocco F, Lelong G, Le Nestour A, Cardinal T, Maali A, et al. Fluorescent silver oligomeric clusters and colloidal particles. Solid State Sci. 2005; 7:812-818.
- Wang Y, Liu H, Jiang Y. A New Method for Immobilization of Polymer-Positive Colloidal Platinum Metals via Co-Ordination Capture with Anchored Ligands. Chem Commun. 1989; 1878-1879.
- Xu H, Suslick KS. Water-soluble fluorescent silver nanoclusters. Adv Mater. 2010; 22(10): 1078-1082.
- Zheng J, Nicovich PR, Dickson RM. Highly Fluorescent Noble Metal Quantum Dots. Annu Rev Phys Chem. 2007; 58:409-431.
- Zhou HS, Honma I, Komiyama H. Controlled synthesis and quantum-size effect in gold-coated nanoparticles. Phys Rev B. 1994; 50:12052-12056.