FLUORESCENT SILVER NANOCLUSTERS STABILIZED BY DNA

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Intro about DNA

- Made up of sequences of nucleic bases:
  - Cytosine
  - Guanine
  - Adenine
  - Thymine

- We use
  - single stranded DNA
  - much smaller strands
  - synthetic DNA

Source: http://www.cardiff.ac.uk/
What are DNA templated silver nanoclusters?

- Different DNA sequences = variety of fluorescent colors
- $\lambda_{Em.\text{peak}}$ tunable = silver atoms rod-like shape
- Bright you can see with the naked eye
What is fluorescence?

- **Absorption**
- **Excited Singlet State**
- **Ground State**
- **Vibrational Relaxation**
- **Fluorescence**
Why study DNA stabilized silver nanoclusters (Ag\textsubscript{N}-DNA)?

- AgN-DNA are promising
  - materials research
  - biological research

- Can be used for:
  - Nano optical materials on DNA
  - Biocompatible fluorescent labels (cells)
  - Sensing metal ions and small molecules
  - Gene sensing

Cell labeling (Zhou, et al. 2013)
How to make Ag$_N$-DNA?

- Specific DNA sequences ~10 bases long
- Add AgNO$_3$
- Reduce with NaBH$_4$
- They make a fluorescent solution...maybe
This summer’s project

- Find strand sequences that yield good Ag\textsubscript{N}-DNA
  - High quantum yield
  - High chemical yield
- Optimizing synthesis
- Time and Photo-bleaching tests
- Chemical stability for isolation
The samples

- Stacy Copp tested 1058 different strands for fluorescence
- Took 11 that were brightest
- Optimized the synthesis
Optimization of synthesis

- Using a well plate method
  - Optimize…
    - DNA Concentration
    - the Ag+/DNA ratio
Data Acquisition

- Plate reader - Tecan
- Emission scan
  - Excites at UV wavelength
  - Scans for emission at visible wavelengths
  - Gives intensity in arbitrary units

![Graph showing normalized absorbance and intensity over wavelength](image)
Data Analysis

- From Tecan data \(\rightarrow\) Intensity vs. wavelength plots
  - To check for:
    - Intensity
    - Emission peak wavelength
    - One or more fluorescent species
After optimization
- fit the emission spectra to Gaussian
- determine the emission peak wavelengths
Once the sample is optimized

Is the sample...

- Time stable?
- Photo-stable?
- Purifiable?
What is time-stability?

- Fluorescence intensity changes after time
  - 1 day
  - 1 week
  - 1 month
Photo-stability

- Measuring emission intensity change after photo-bleaching

- Photo-bleaching
  - Fluorescence intensity decreases after shining light
    - Visible
    - UV

- Many experiments require detection with light

- Ideal cluster will remain stable for a reasonable amount of time

- Many are not very photo stable
Photo-stability

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- Photo-bleaching
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    - visible
    - UV
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Photo-bleaching Findings

- UV bleaching is not correlated to visible bleaching

Change in Visible vs Change in UV

Intensity/Intensity_initial (VIS)

Intensity/Intensity_initial (UV)
Visible Photo-bleaching

- LED array
  - Broad spectrum
  - Uniform
    - Time
    - Wavelength spectrum

Source: Cree Xlamp CXA3050 datasheet
Progress
Isolate fluorescent clusters

- Separate out a solution of different components
- Isolate to understand
- Number of Ag atoms
High Performance Liquid Chromatography (HPLC)

- Passes sample through a column
- Different products elute at different times
- Not all samples survive the column
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Absorbance monitors all DNA ~260nm

Excited at 270nm

Monitors emission at all wavelengths through time
Optimized synthesis

ssDNA + AgNO₃

NaBH₄

Time
Summary

Optimized synthesis

Photo-stability

Source: Cree Xlamp CXA3050 datasheet
Summary

- Optimized synthesis
- Photo-stability
- Purifiable

Diagram:
- Solvent Gradient
- Separated Ag:DNA complexes
- Column
- \( A_{260} \)
- \( \lambda \)
- \( I_{em} \)
- Mass Spec
- Collection
- 270 nm
Promising candidates

Normalized Intensity

Wavelength [nm]

400 500 600 700 800

Normalized Intensity

0.0 0.2 0.4 0.6 0.8 1.0

Em.498nm
Em.578nm
Em.643nm
Em.659nm
Em.699nm
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