

Techniques for Characterization of Carbohydrate Structure of Polysaccharides

August 10-14, 2015

LABORATORY EXERCISE

Section I

Alditol acetate (AA) composition

Section II

Methyl glycosides (TMS) composition

Section III

Partially methylated alditol acetate (PMAA) linkage

Lab Instructors: Zhirui Wang, Radnaa Naran, Justyna Dobruchowska and Ian Black

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Monday, August 10, 2015

- 9:00 a.m.- 9:15 a.m. **Welcome and Introduction** - Dr. Parastoo Azadi
- 9:15 a.m.- 9:30 a.m. Hydrolysis of AA sample
- 9:30 a.m.-11:00 a.m. **Lecture: Dr. Parastoo Azadi:**
 “Structural Characterization of Carbohydrates”
- 11:00 a.m.-12:15 p.m. Remove AA sample from heat and dry down with nitrogen

 Sodium borodeuteride reduction of AA sample
- 12:15 p.m.- 1:15 p.m. **LUNCH**
- 1:15 p.m.- 2:15 p.m. Neutralization and drying of AA sample

 Acetylation of AA sample
- 2:15 p.m.- 3:45 p.m. **Lecture: Dr. Geert-Jan Boons:**
 “Different Forms of D-Glucose”
- 3:45 p.m.- 5:00 p.m. Drying, extraction and injection of AA sample

 Overnight methanolysis of methyl glycosides (TMS) sample

Tuesday, August 11, 2015

- 9:00 a.m.- 9:30 a.m. Dry down methyl glycosides (TMS) sample.

 Prepare NaOH base for permethylation (Linkage)
- 9:30 a.m.-10:25 a.m. Permethylation of the linkage sample

 N-acetylation of methyl glycosides samples (TMS)
- 10:25 a.m.-10:40 a.m. **BREAK**

- 10:40 a.m.-11:15 a.m. Remove the CH₃I, DCM Extraction and drying (Linkage)
Drying of N-acetylated methyl glycosides (TMS) sample after 30 min reaction
- 11:15 a.m.-12:15 p.m. **Lecture: Dr. Debra Mohnen**
TBA
- 12:15 p.m.- 1:15 p.m. **LUNCH**
- 1:15 p.m.- 2:15 p.m. Silylation of the methyl glycosides (TMS) sample
Laboratory demonstration (Lab 1025)
Preparation of (dimesyl) potassium dimethylsulphinyl anion
- 2:15 p.m.- 2:30 p.m. **BREAK**
- 2:30 p.m.- 4:00 p.m. Laboratory
Drying, filtration and injection of the methyl glycosides (TMS) samples
- 4:00 p.m.- 5:00 p.m. **Lecture: Dr. Artur Muszynski**
“Isolation and Characterization of Bacterial Surface Polysaccharides”

Wednesday, August 12, 2015

- 9:00 a.m.- 9:15 a.m. Hydrolysis of PMAA sample
- 9:15 a.m.- 9:45 a.m. **Demonstration: Dr. Roberto Sonon**
“Lignin, MBMS”
- 9:45 a.m.- 10:00 a.m. **BREAK**
- 10:00 a.m.-11:00 a.m.
Lecture: Dr. Parastoo Azadi
“Introduction to High Performance Anion Exchange Chromatography”
- 11:00 a.m.-12:15 p.m. Remove linkage sample from heat and dry down with nitrogen
Sodium borodeuteride reduction of linkage sample
- 12:15 p.m.- 1:15 p.m. **LUNCH**
- 1:15 p.m.- 2:30 p.m. Neutralization and drying of AA sample

Acetylation of AA sample

2:30 p.m.- 3:15 p.m. Data analysis of AA: Dr. Parastoo Azadi

3:15 p.m.- 3:30 p.m. **BREAK**

3:30 p.m.- 5:00 p.m. Drying, extraction and injection of PMAA sample

Thursday, August 13, 2015

9:00 a.m.- 9:45 a.m. Data analysis of TMS: Dr. Zhirui Wang

9:45 a.m.-10:00 a.m. Break

10:00 a.m.-11:00 a.m. **Lecture: Dr. Michael Hahn**
TBA

11:00 a.m.-12:15 p.m. **Lecture: Dr. Stephanie Archer-Hartmann**
“Labeling and Separation of Carbohydrates”

12:15 p.m.- 1:15 p.m. **LUNCH**

1:15 p.m.- 2:00 p.m. HPAEC demonstration: Oligosaccharide profiling analysis and monosaccharide composition analysis: Dr. Roberto Sonon

2:00 p.m.- 2:45 p.m. Data analysis of linkage: Dr. Parastoo Azadi

2:45 p.m.- 3:20 p.m. **BREAK**

3:20 p.m.- 3:45 p.m. **Lecture: Saeid Roushanzamir**
“FACES Scheduling”

3:45 p.m.- 5:00 pm Questions and answers for data interpretation of all the analyses

Friday, August 14, 2015

Choose one of the following:

NMR Module or Mass Spectrometry Module

NMR Module

9:00 a.m.-12:00 p.m. **Lecture: Dr. John Glushka**
“Introduction to NMR of Glycoproteins and Carbohydrates”

