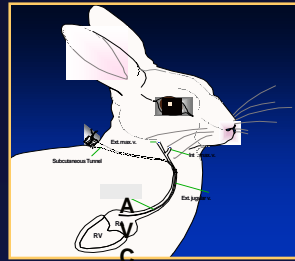


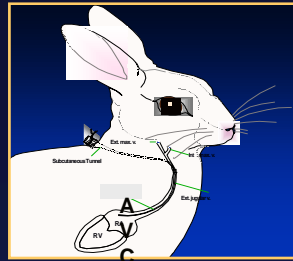
Characterization, Identification, and Comparative Proteomic
Kinetics of Innate Host Defense Proteins in Experimental IPA
and *Pseudomonas* Pneumonia

Thomas Walsh, MD
NIH/NCI

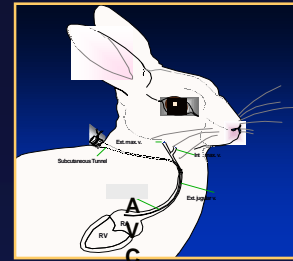
Pulmonary Aspergillosis & Pseudomonas Pneumonia



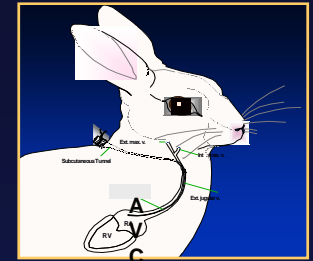
Normal



Neutropenic



Pseudomonas



Aspergillus

++

+

+++

Cytarabine



Methylprednisolone



Antibiotics



Intratracheal *A. fumigatus*



Blood Sampling

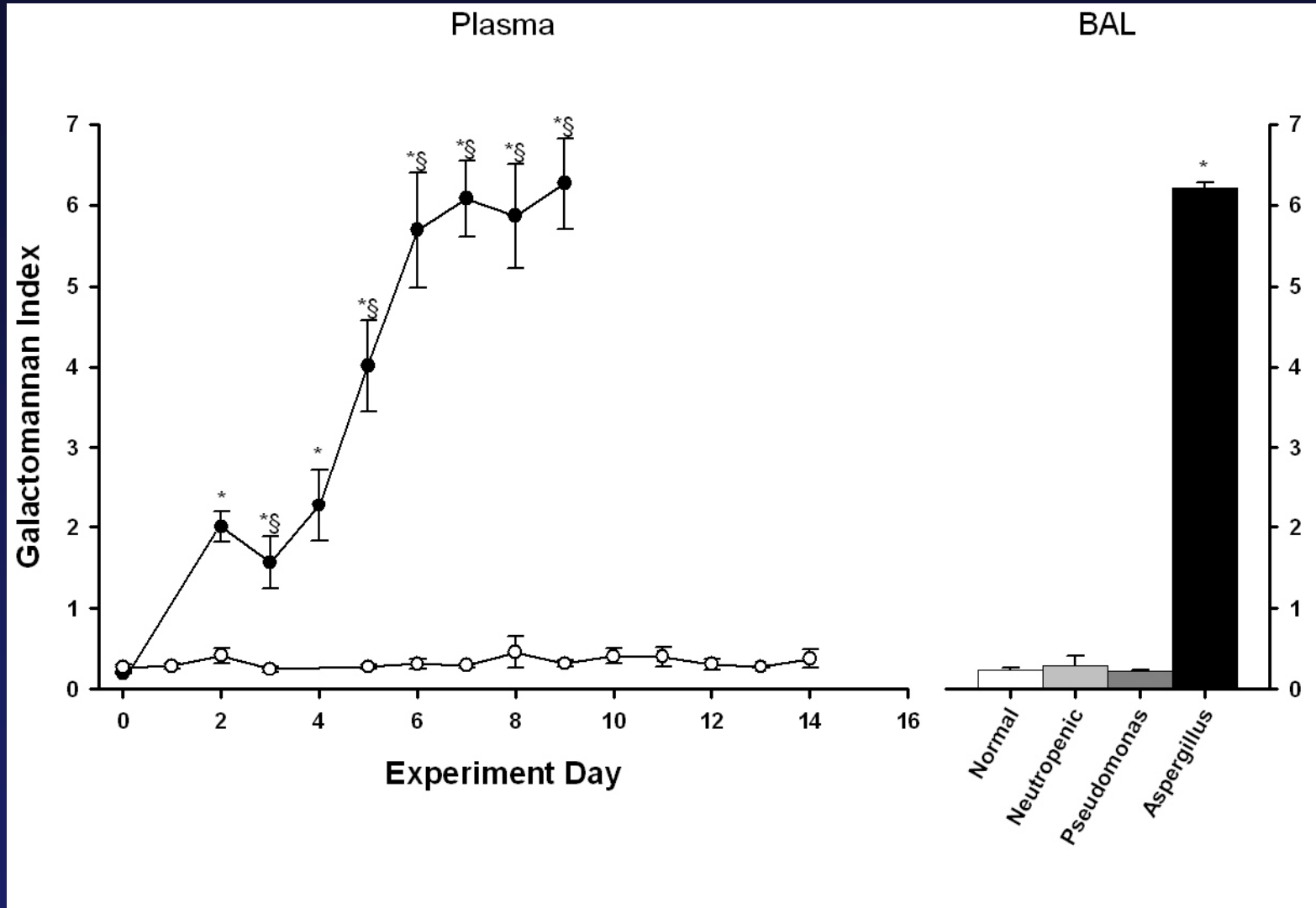


BAL Sampling

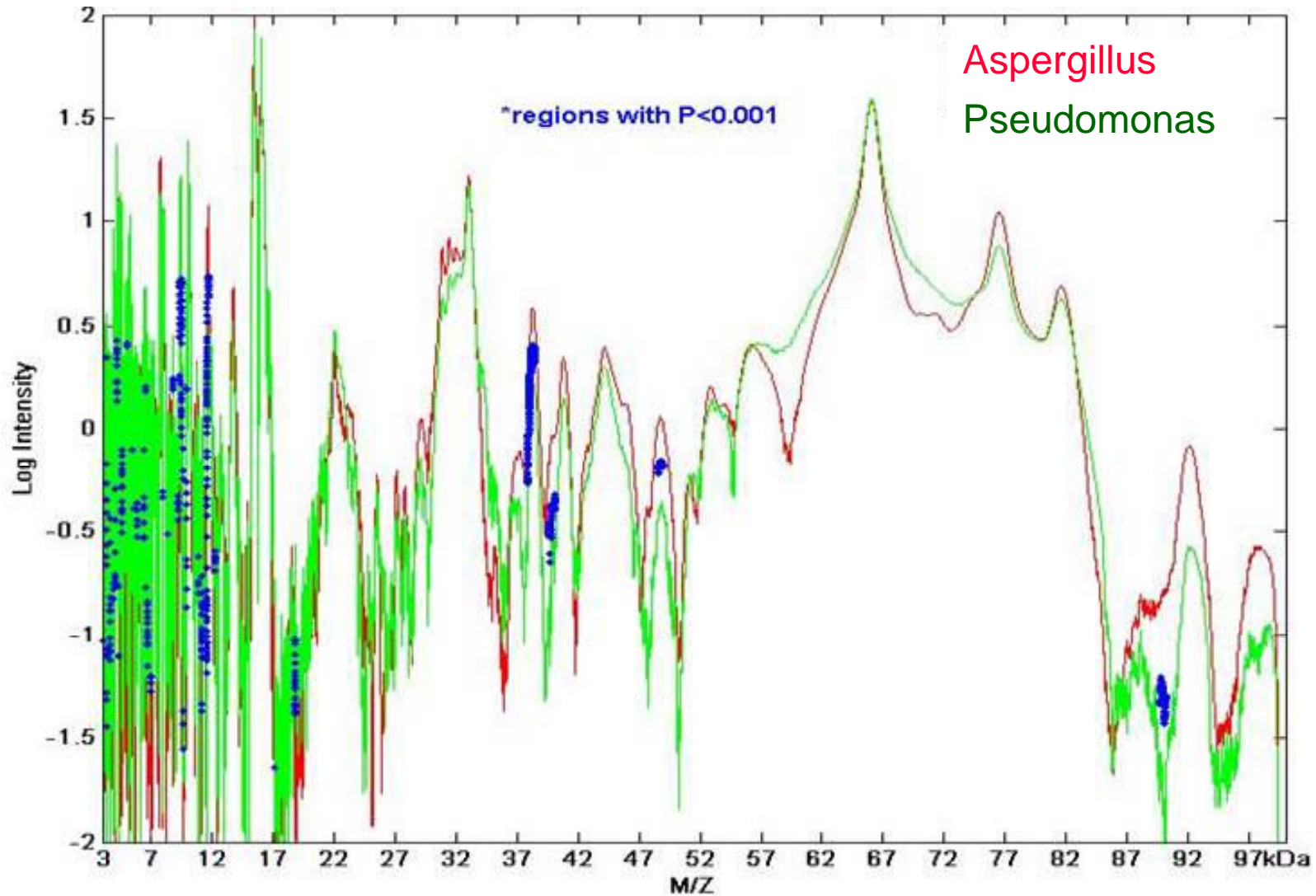


Day 0 1 2 3 4 5 6 7 8 9 1014

Expression of Serum and BAL Galactomannan



Comparison of Spectral Regions of Plasma in Aspergillus and Pseudomonas Pneumonia



Hierarchical Clustering of Spectra

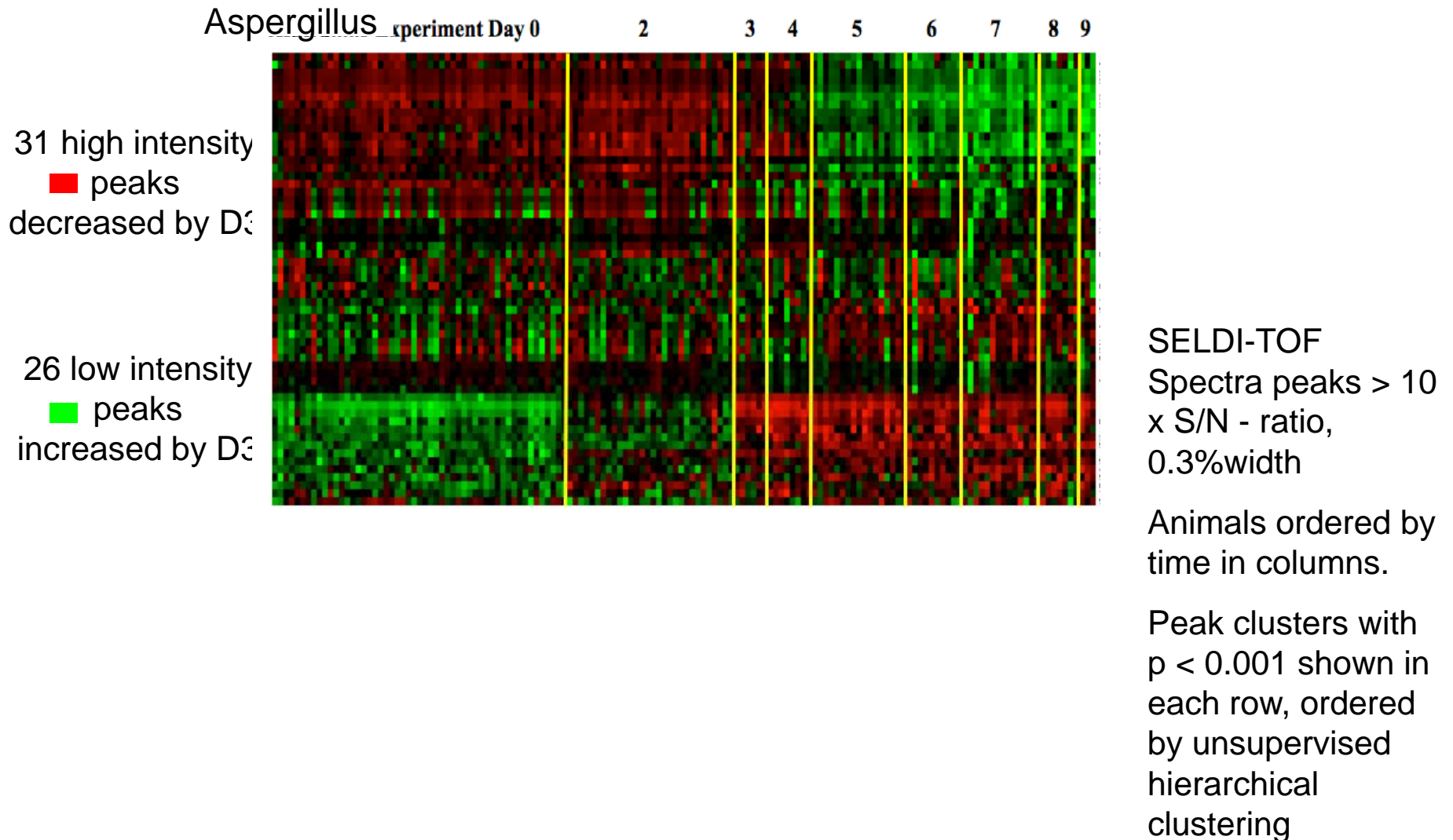
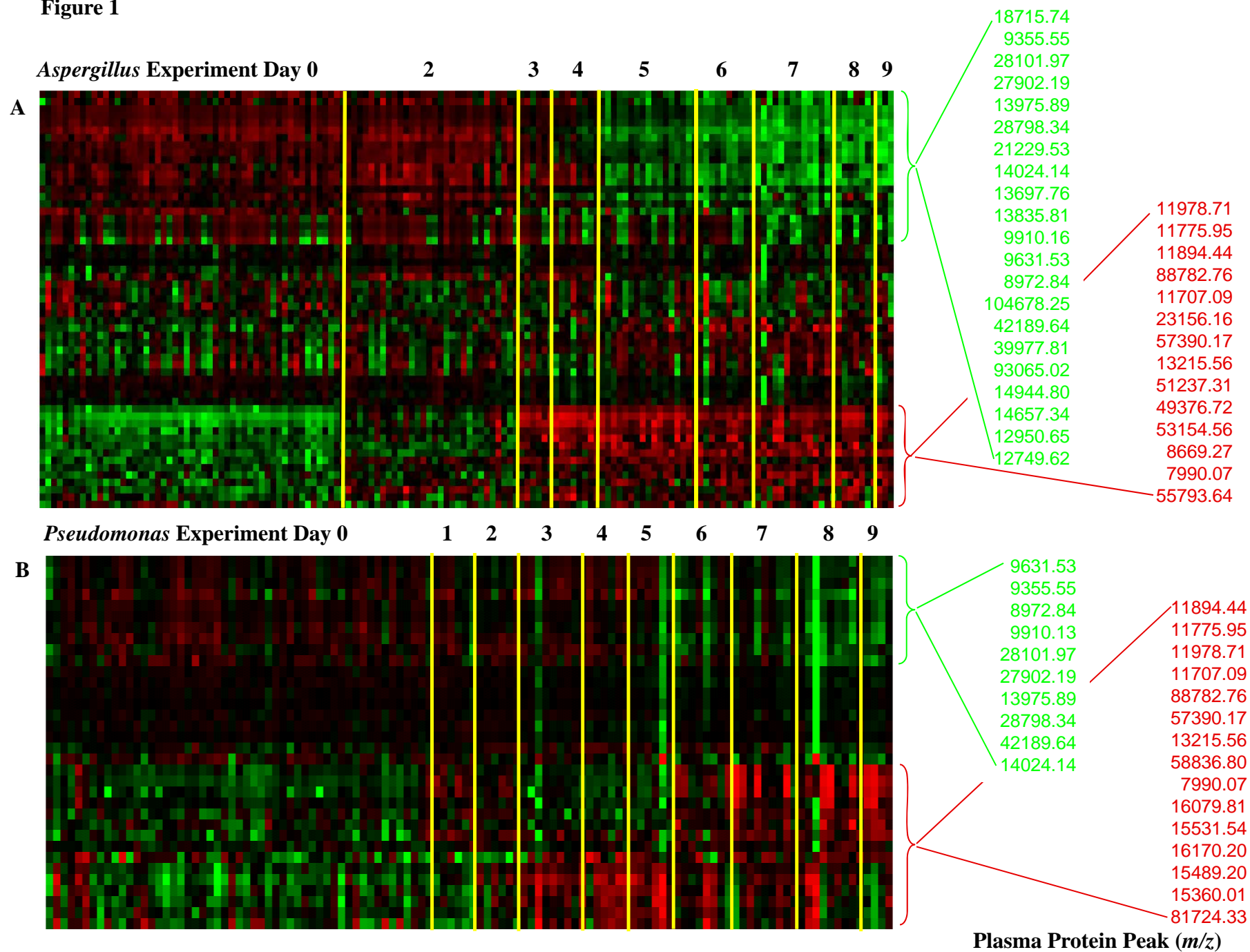
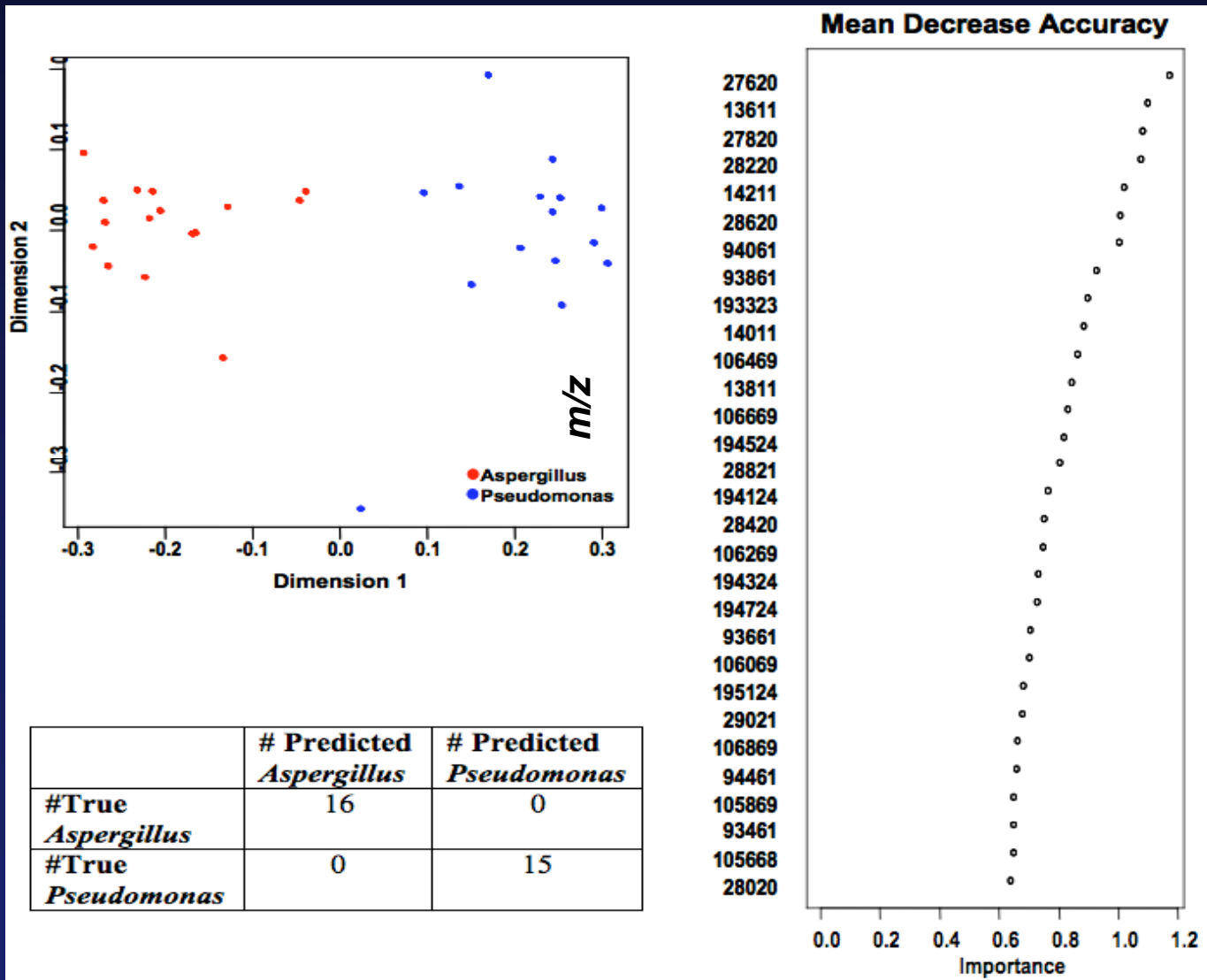
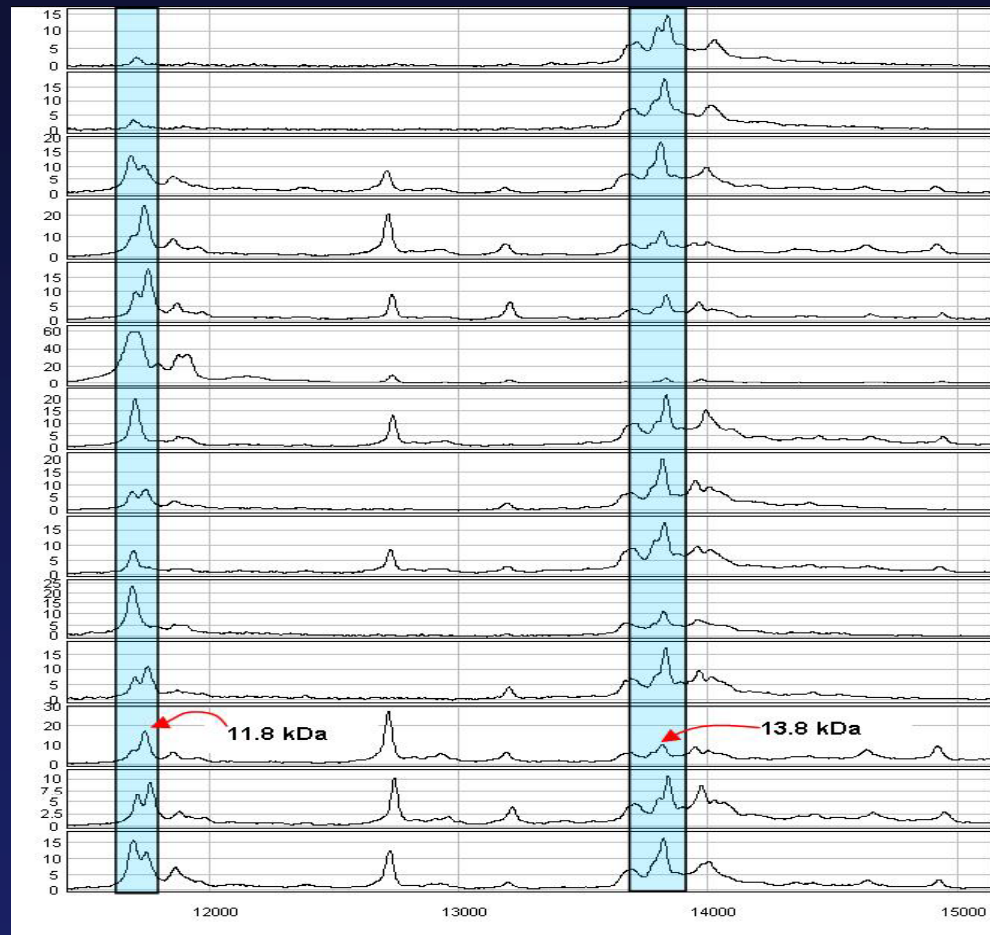
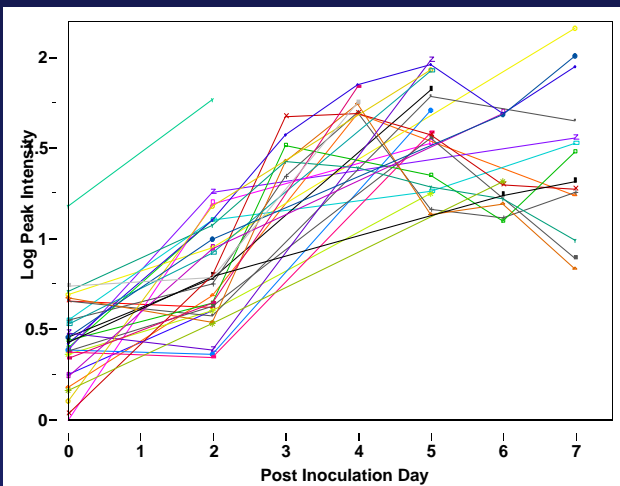
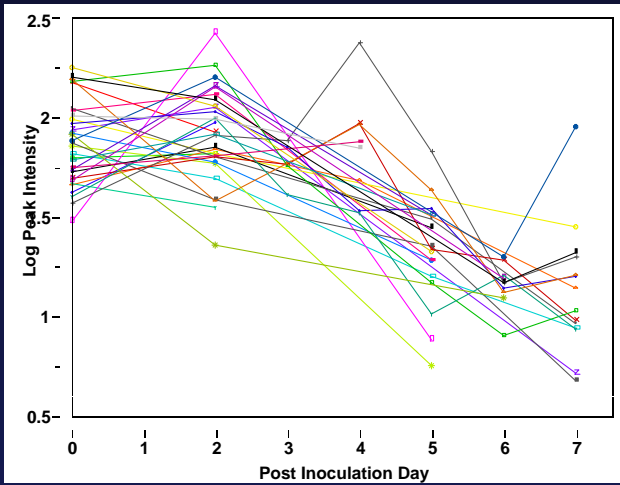


Figure 1



Random Forest Analysis of Top 30 Spectral Points Distinguish Between *Aspergillus* and *Pseudomonas* Pneumonia





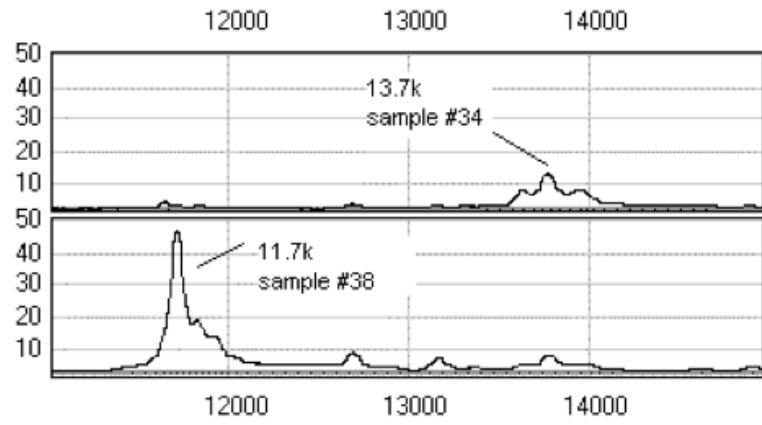
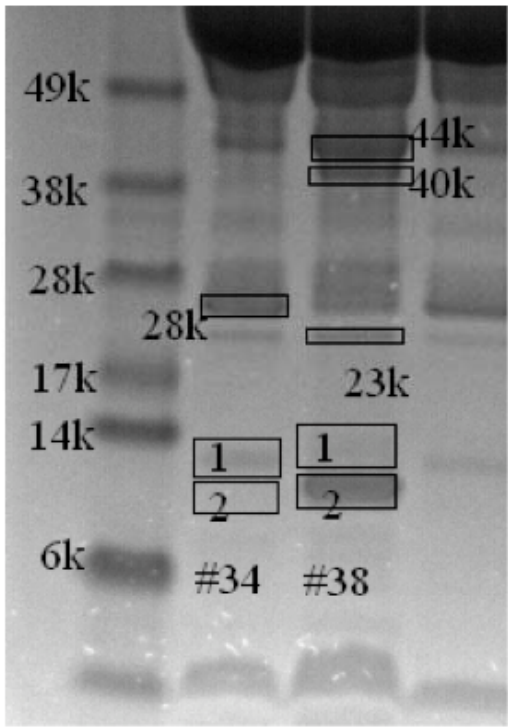


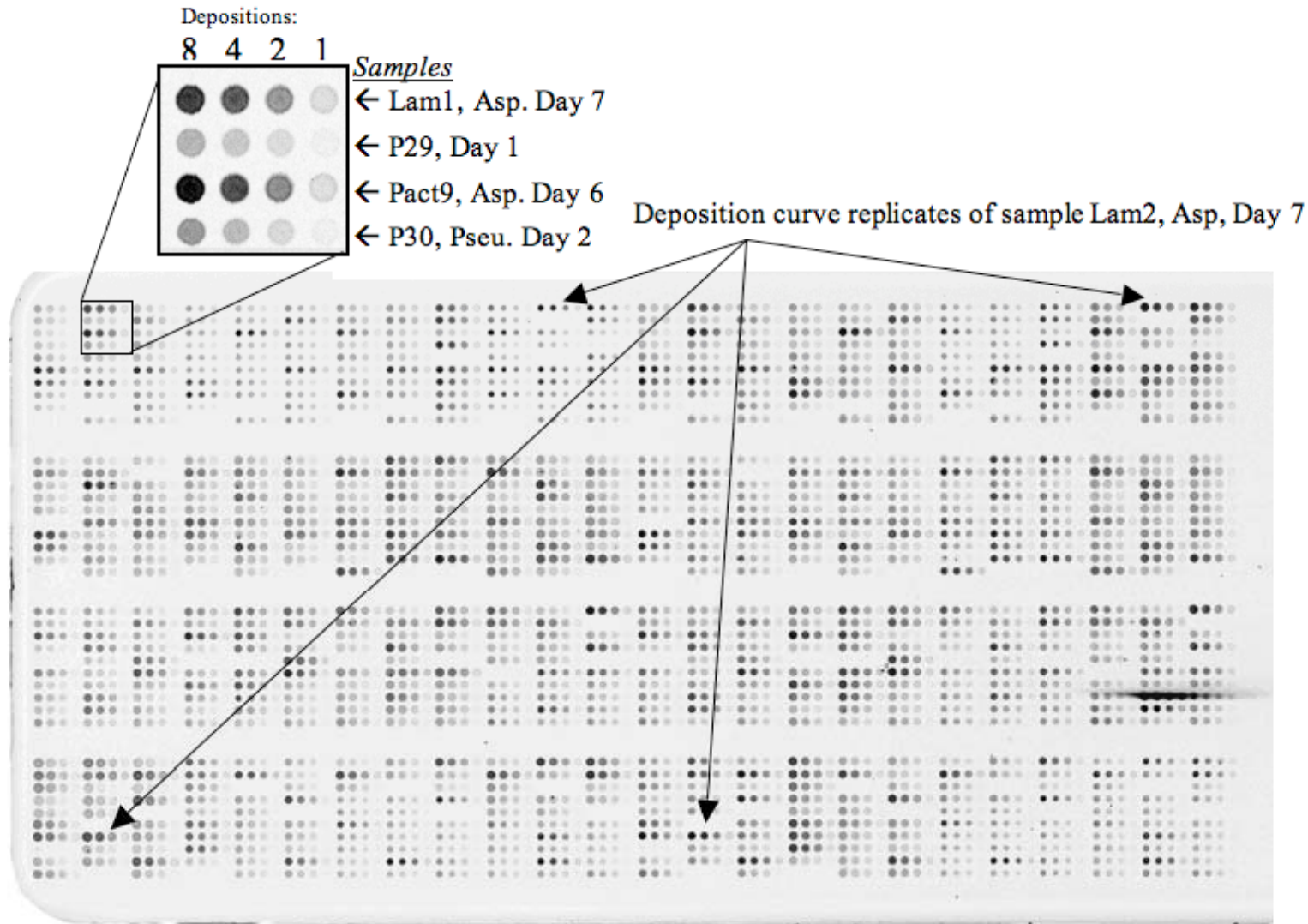
Figure A: SELDI-TOF shows the peaks of 11.7kDa and 13.7kDa in sample #34 and #38.



- 44K: HPT (1736), CO3 (432), PON1 (246), FIBA (103), ALBU (103), CLUS (39)
- 40K: HPT (737), FIBA (214), ALBU
- 28K: APOA1 (166)
- 23K: ALBU (174), HPT (147), KAC4 (113), CRP (73)
- #34_1: TTHY (129)
- #34_2: No ID
- #38_1: HBB (265), TTHY (167), HBE (101), HBA, HBG, CA2D1
- #38_2: HPT (399), TTHY (185), SAA2 (177) (or SAA), LOX12 (41), B2MG (41)

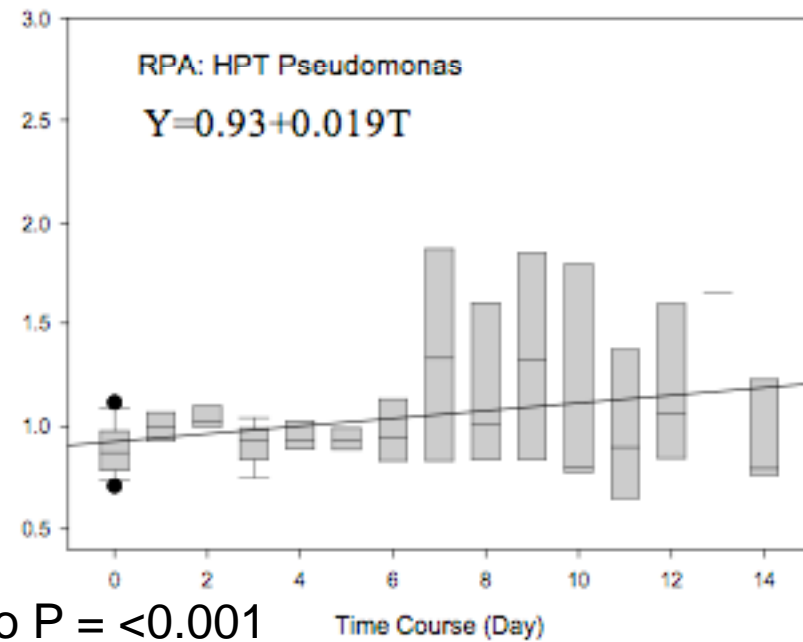
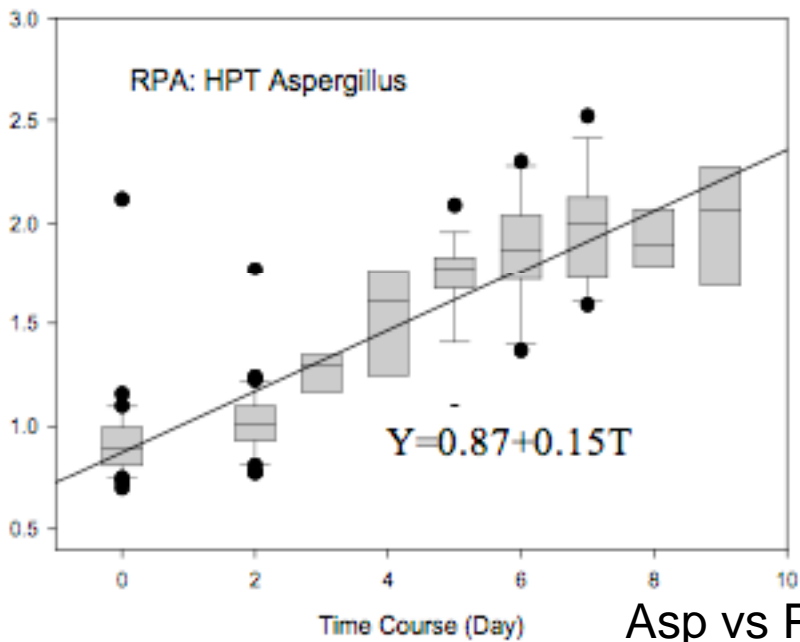
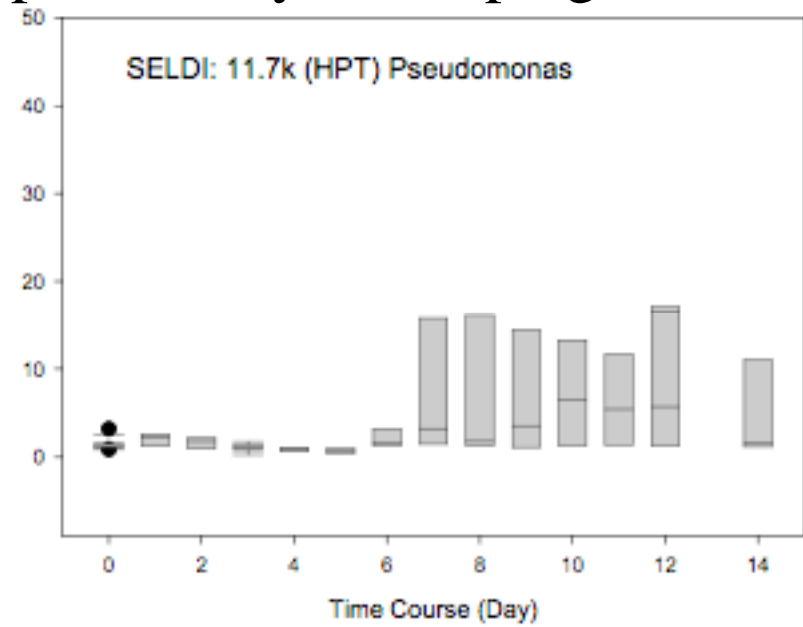
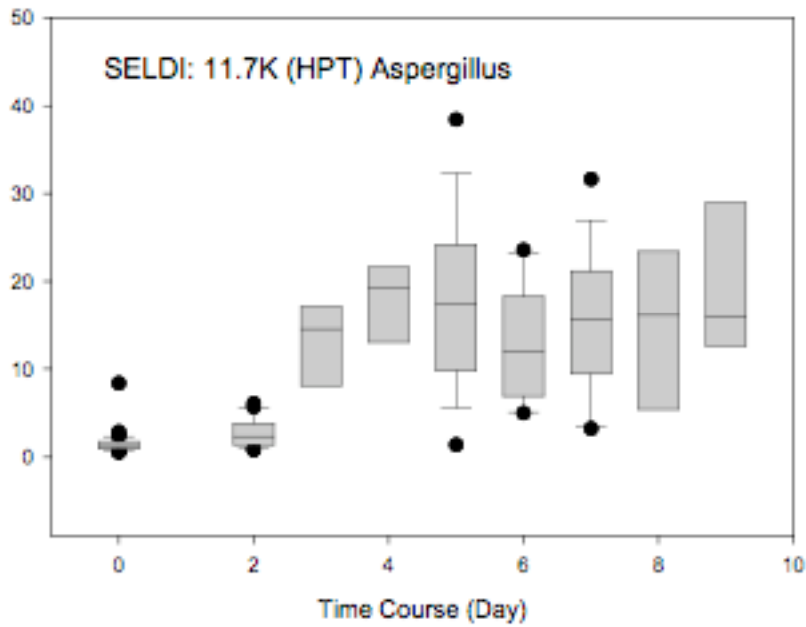
The image of 1D SDS-Page gel with coomassie blue stain. Line 1 is molecular markers; Line 2 is sample #34 (pseudomonas) and Line 3 is sample #38 (aspergillus). On the right image, 23k, 28k, 40k and 44k are just band labels. They do not necessarily represent their actual molecular weight. The protein IDs with significant hit ($p < 0.05$) are listed on the left. The numeric value in the parenthesis () is Mowse score.

Reverse antibody array



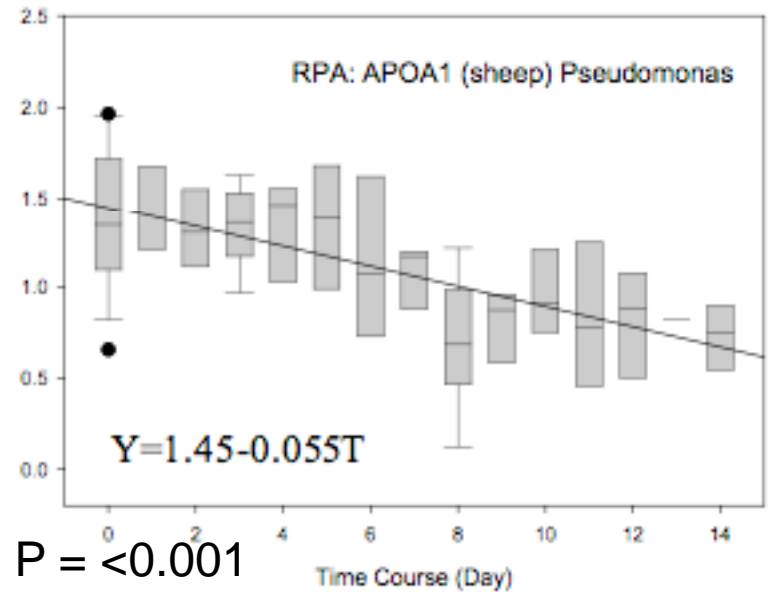
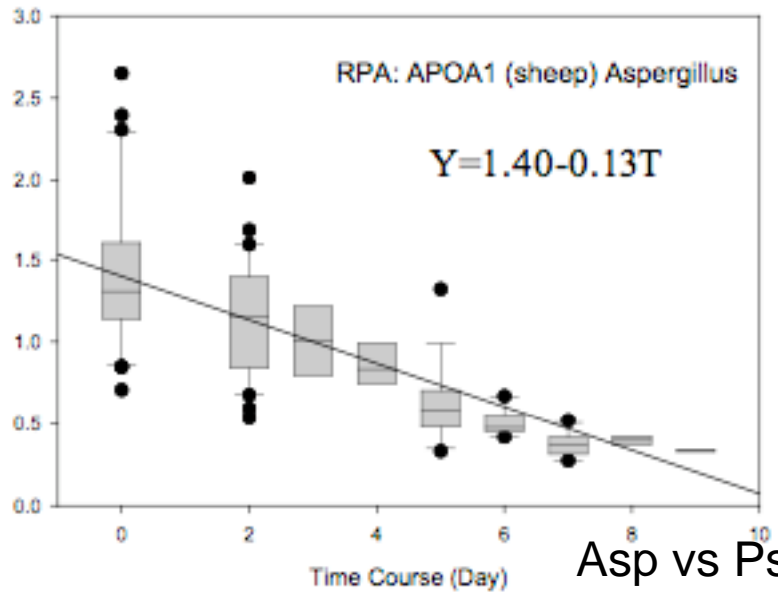
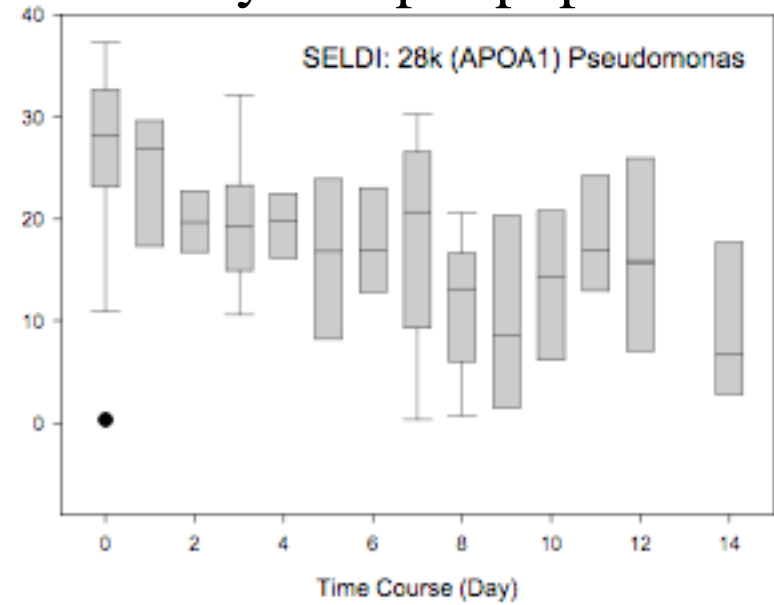
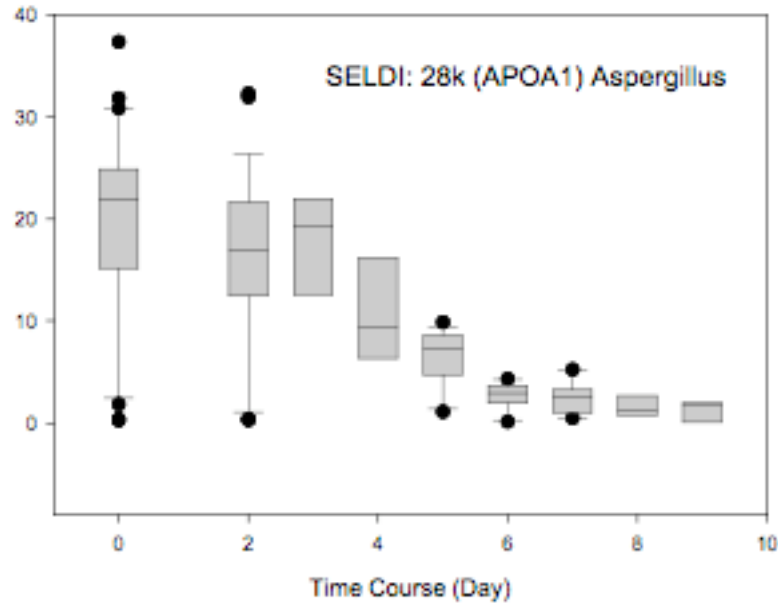
This RPA image (contrast adjusted) shows an array immuno-stained with anti-rabbit HPT antibody and indicates the layout of samples on the slide. Each unique sample has 16 spots composed of 8, 4, 2 and 1 deposition curves printed in quadruplicate.

Protein validation with reversed phase array for Haptoglobin



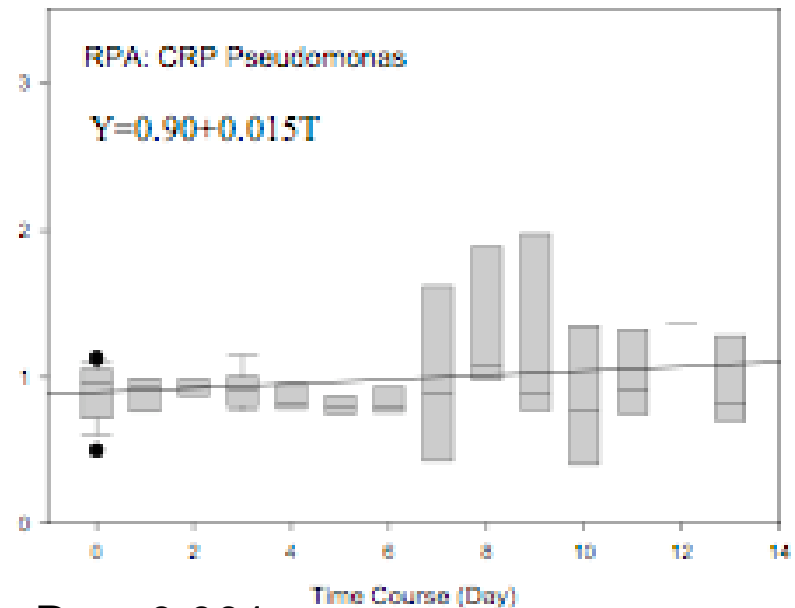
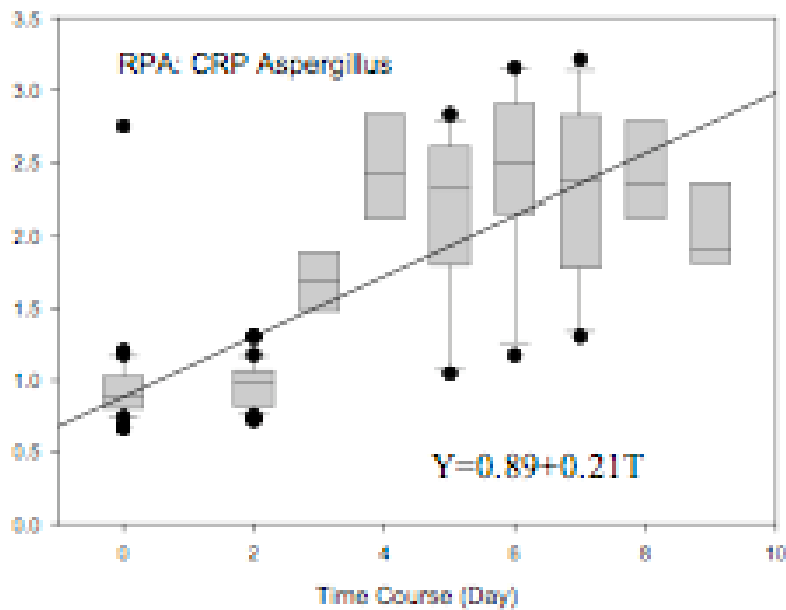
Asp vs Pseudo P = <0.001

Protein validation with reversed phase array of ApolipoproteinA1



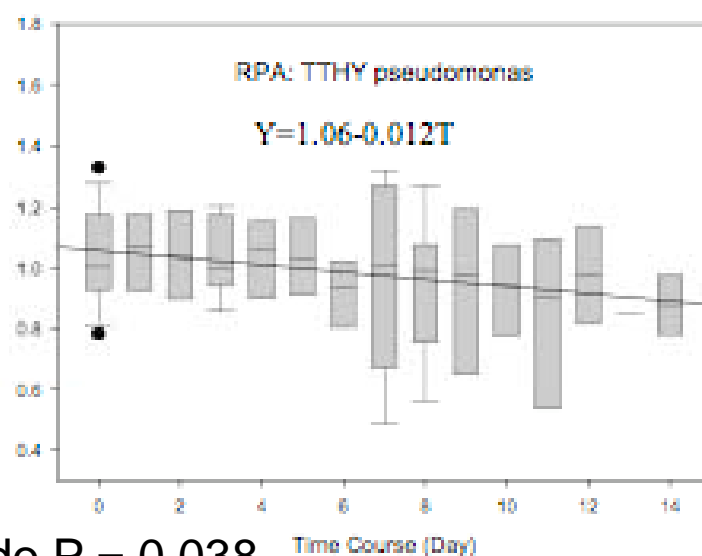
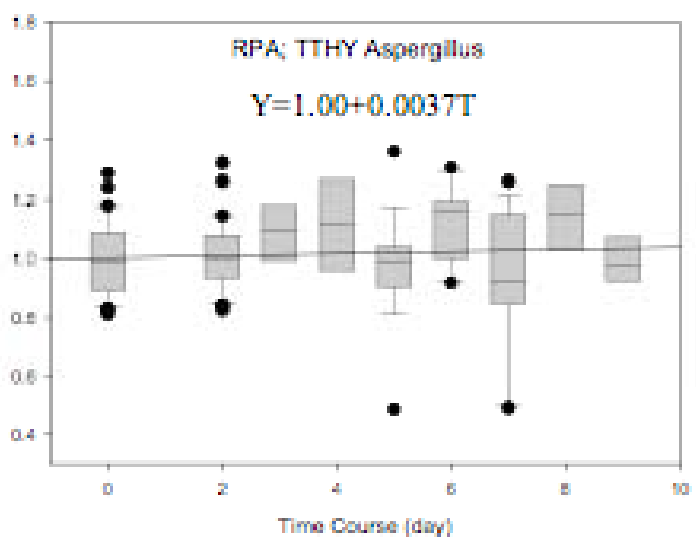
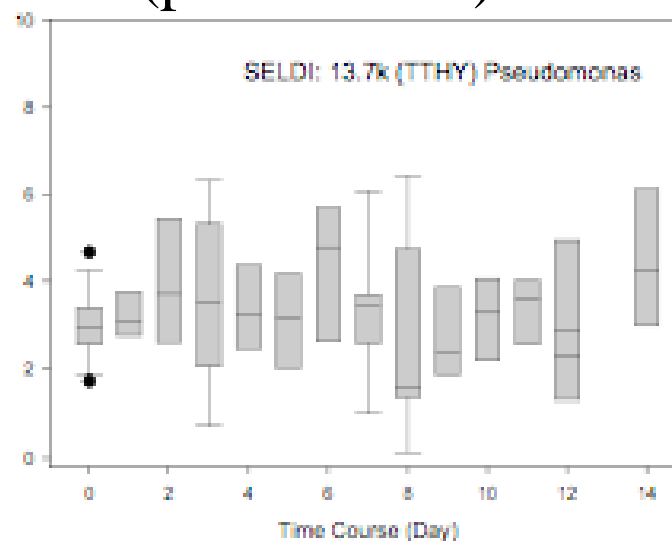
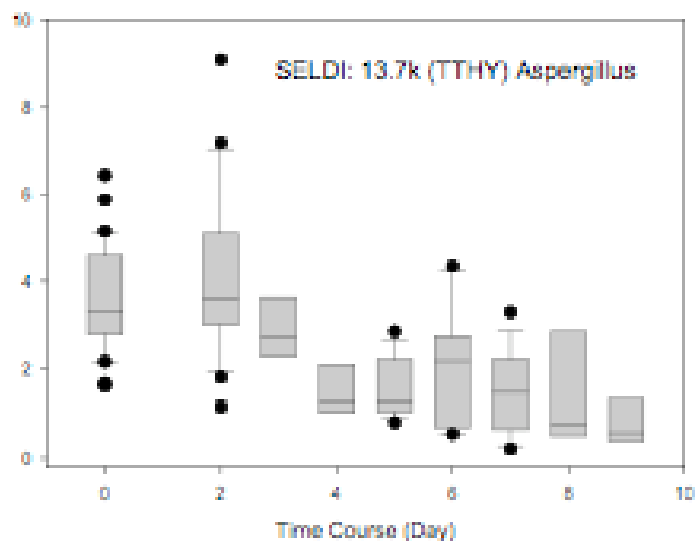
Asp vs Pseudo P = <0.001

The change of relative CRP level with time course in Aspergillus and Pseudomonas animals by reverse phase array



Asp vs Pseudo P = <0.001

Protein validation with reverse phase array for transthyretin (pre-albumin)



Asp vs Pseudo P = 0.038

Conclusions

- Proteomic analysis of serum and BAL proteins in experimental *Aspergillus* and *Pseudomonas* pneumonia demonstrate spectral regions that are shared in early infection and distinguish between the two etiologies at later stages of infection
- These data support the concept that a pattern of proteins are expressed with different microbial infections
- Haptoglobin, apolipoprotein A1, and C-reactive protein display distinct kinetic profiles and with probable roles in the pathogenesis of invasive pulmonary aspergillosis
- This approach may enhance our understanding of host-pathogen interactions, detection and prognosis

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