

Proteome vs. Transcriptome: Comparisons between microarray data and proteomic data for the organism *Yersinia pestis*

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Overview

Label-free relative quantities of proteins produced by *Y. pestis* under growth conditions that mimic growth in a flea or mammalian host were obtained by using the accurate mass and time tag (AMT) approach.

The data were compared with microarray data grown under similar conditions. mRNA and protein expression ratios are not typically the same but in the case of the type III secretion system, there is a lot of similarity between the results of the two techniques.

Introduction

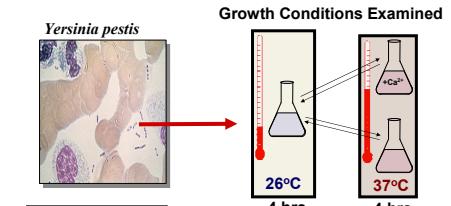
Until recently, global protein expression changes in organisms cultured or isolated under different conditions were based on relative mRNA expression levels, and this data was used to make inferences on regulation and protein expression. However, recent studies have demonstrated poor correlations of mRNA and protein levels in cellular systems.

With the more recent advent of high-throughput proteomics methodologies, we can now directly observe both the genetic output of the transcriptome and proteome. This combined knowledge provides together a deeper understanding of a cell system than one technique alone. We have applied a high-throughput characterization of the proteome of *Yersinia pestis* (agent of plague) and have compared the output to mRNA expression levels of the same sample conditions.

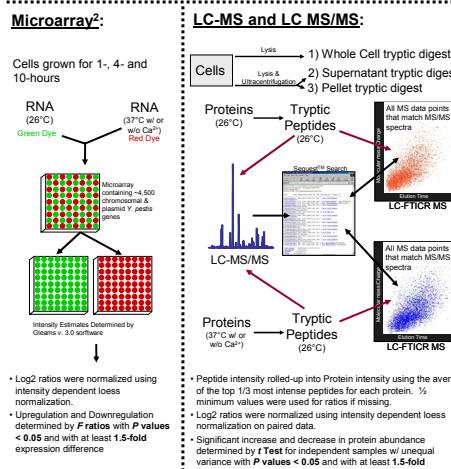
Methods

Cells from *Y. pestis* were cultured under conditions that would evoke response triggers similar to those encountered in growth in a flea vector or mammalian host. DNA microarrays encompassing the entire genome of *Yersinia pestis* were used to characterize global regulatory changes during growth occurring after shift from 26 to 37 °C in the presence and absence of Ca²⁺. With analogous *Y. pestis* cells, genome-wide protein identities and intensities were made using LC-FTICR MS, utilizing a pre-existing mass tag database established using MS/MS. The log base 2 ratios of the mRNA and protein products were compared.

Y. Pestis cells were grown with environmental triggers to evoke similar virulence associated responses to mimic growth in either a flea vector or mammalian host. Temperature and lack or presence of Ca²⁺ was used to evoke protein and mRNA change.



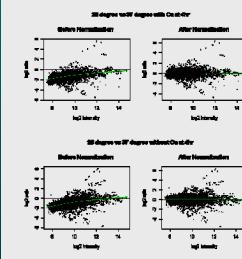
- Polar activation of Type III secretion system at bacteria/eukaryotic cell contact point
 - Artificial secretion induced by removal of Ca²⁺ from the growth medium
 - Virulent Factor Regulator – Ca²⁺ concentration
- Growth conditions mimic growth states between the flea vector and mammalian host.



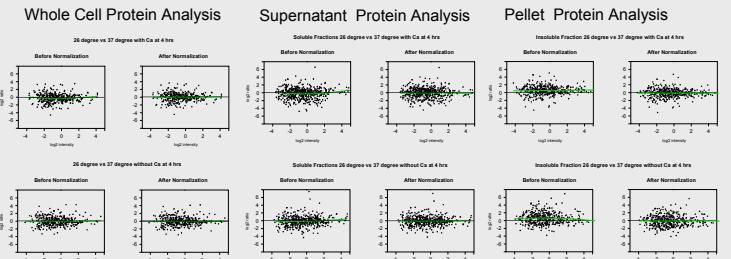
Results

Ratio vs. Intensity plots for data obtained from Microarray and Proteomics Analyses

2^{Microarray Data}

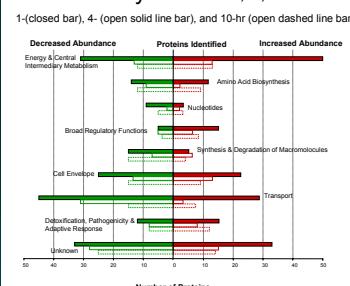


Proteomics Data

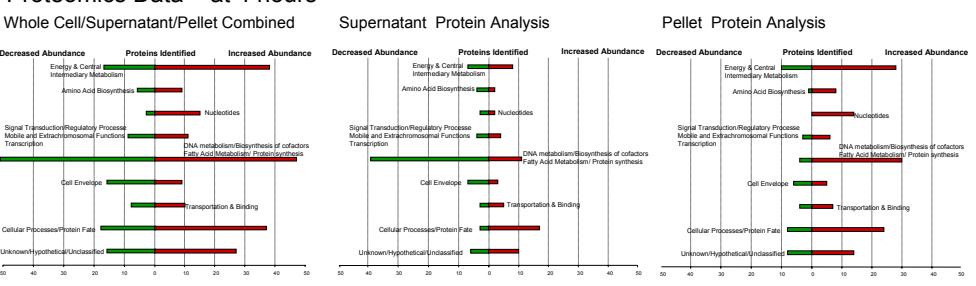


Functional classification of thermoregulated chromosomal genes and proteins of *Y. pestis*

2^{Microarray Data – 1, 4, and 10 hr}



Proteomics Data – at 4 hours



Thermoregulated virulence associated genes and proteins of *Y. pestis* – genes and proteins of plasmid CD1

Yersinia pestis uses the Type III secretion system (TTSS) to inject virulence associated proteins into a host cell. These proteins lead to decreased immune response and cell destruction. This TTSS is coded for by plasmid CD1.

Figure 2. Illustration of the Type III secretion system machinery of *Y. pestis* coded for on plasmid pCD1. The components are primarily assembled under conditions of low calcium at 37 °C which mimic conditions experienced by the bacteria when attached to a mammalian host cell. Virulent proteins "injected" into the host cell include YopM, YopH, YopE, YopP, YpkA, and YopT.

Gene or ORF	Microarray Data		Proteomics Data	
	1 hr Ca ²⁺	4 hrs Ca ²⁺	10 hrs Ca ²⁺	4 hrs Ca ²⁺
Y0077c_yopE	4.6	4.5	8.1	5.8
Y0078_yopE	3.1	9.4	8	5.7
Y0079_yopC	3.9	4.1	2.7	5.6
Y0080_yopC	3.1	6.1	1.1	6.3
Y0082_yopG	5.1	4.4	7.3	6.4
Y0083_yopG	2.5	4.7	2.0	3.3
Y0084_yopG	2.5	4.7	2.0	3.3
Y0085_yopG	1.8	1.7	2.8	4.5
Y0086_yopG	1.8	2.1	2.7	6.4
Y0087_yopG	4.8	3.4	4.6	5.2
Y0088_yopG	2.7	2.1	2.9	2.3
Y0089_yopG	2.7	2.9	3.7	4.5
Y0090_yopG	1.9	1.7	2.8	3.7
Y0091_yopG	1.9	1.7	2.8	3.7
Y0092_yopG	1.9	1.7	2.8	3.7
Y0093_yopG	1.9	1.7	2.8	3.7
Y0094_yopG	1.9	1.7	2.8	3.7
Y0095_yopG	1.9	1.7	2.8	3.7
Y0096_yopG	1.9	1.7	2.8	3.7
Y0097_yopG	1.9	1.7	2.8	3.7
Y0098_yopG	1.9	1.7	2.8	3.7
Y0099_yopG	1.9	1.7	2.8	3.7
Y0100_yopG	1.9	1.7	2.8	3.7
Y0101_yopG	1.9	1.7	2.8	3.7
Y0102_yopG	1.9	1.7	2.8	3.7
Y0103_yopG	1.9	1.7	2.8	3.7
Y0104_yopG	1.9	1.7	2.8	3.7
Y0105_yopG	1.9	1.7	2.8	3.7
Y0106_yopG	1.9	1.7	2.8	3.7
Y0107_yopG	1.9	1.7	2.8	3.7
Y0108_yopG	1.9	1.7	2.8	3.7
Y0109_yopG	1.9	1.7	2.8	3.7
Y0110_yopG	1.9	1.7	2.8	3.7
Y0111_yopG	1.9	1.7	2.8	3.7
Y0112_yopG	1.9	1.7	2.8	3.7
Y0113_yopG	1.9	1.7	2.8	3.7
Y0114_yopG	1.9	1.7	2.8	3.7
Y0115_yopG	1.9	1.7	2.8	3.7
Y0116_yopG	1.9	1.7	2.8	3.7
Y0117_yopG	1.9	1.7	2.8	3.7
Y0118_yopG	1.9	1.7	2.8	3.7
Y0119_yopG	1.9	1.7	2.8	3.7
Y0120_yopG	1.9	1.7	2.8	3.7
Y0121_yopG	1.9	1.7	2.8	3.7
Y0122_yopG	1.9	1.7	2.8	3.7
Y0123_yopG	1.9	1.7	2.8	3.7
Y0124_yopG	1.9	1.7	2.8	3.7
Y0125_yopG	1.9	1.7	2.8	3.7
Y0126_yopG	1.9	1.7	2.8	3.7
Y0127_yopG	1.9	1.7	2.8	3.7
Y0128_yopG	1.9	1.7	2.8	3.7
Y0129_yopG	1.9	1.7	2.8	3.7
Y0130_yopG	1.9	1.7	2.8	3.7
Y0131_yopG	1.9	1.7	2.8	3.7
Y0132_yopG	1.9	1.7	2.8	3.7
Y0133_yopG	1.9	1.7	2.8	3.7
Y0134_yopG	1.9	1.7	2.8	3.7
Y0135_yopG	1.9	1.7	2.8	3.7
Y0136_yopG	1.9	1.7	2.8	3.7
Y0137_yopG	1.9	1.7	2.8	3.7
Y0138_yopG	1.9	1.7	2.8	3.7
Y0139_yopG	1.9	1.7	2.8	3.7
Y0140_yopG	1.9	1.7	2.8	3.7
Y0141_yopG	1.9	1.7	2.8	3.7
Y0142_yopG	1.9	1.7	2.8	3.7
Y0143_yopG	1.9	1.7	2.8	3.7
Y0144_yopG	1.9	1.7	2.8	3.7
Y0145_yopG	1.9	1.7	2.8	3.7
Y0146_yopG	1.9	1.7	2.8	3.7
Y0147_yopG	1.9	1.7	2.8	3.7
Y0148_yopG	1.9	1.7	2.8	3.7
Y0149_yopG	1.9	1.7	2.8	3.7
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Y0175_yopG	1.9	1.7	2.8	3.7
Y0176_yopG	1.9	1.7	2.8	3.7
Y0177_yopG	1.9	1.7	2.8	3.7
Y0178_yopG	1.9	1.7	2.8	3.7
Y0179_yopG	1.9	1.7	2.8	3.7
Y0180_yopG	1.9	1.7	2.8	3.7
Y0181_yopG	1.9	1.7	2.8	3.7
Y0182_yopG	1.9	1.7	2.8	3.7
Y0183_yopG	1.9	1.7	2.8	3.7
Y0184_yopG	1.9	1.7	2.8	3.7
Y0185_yopG	1.9	1.7	2.8	3.7
Y0186_yopG	1.9	1.7	2.8	3.7
Y0187_yopG	1.9	1.7	2.8	3.7
Y0188_yopG	1.9	1.7	2.8	3.7
Y0189_yopG	1.9	1.7	2.8	3.7
Y0190_yopG	1.9	1.7	2.8	3.7
Y0191_yopG	1.9	1.7	2.8	3.7
Y0192_yopG	1.9	1.7	2.8	3.7
Y0193_yopG	1.9	1.7	2.8	3.7
Y0194_yopG	1.9	1.7	2.8	3.7
Y0195_yopG	1.9	1.7	2.8	3.7
Y0196_yopG	1.9	1.7	2.8	3.7
Y0197_yopG	1.9	1.7	2.8	3.7
Y0198_yopG	1.9	1.7	2.8	3.7
Y0199_yopG	1.9	1.7	2.8	3.7
Y0200_yopG	1.9	1.7	2.8	3.7
Y0201_yopG	1.9	1.7	2.8	3.7
Y0202_yopG	1.9	1.7	2.8	3.7
Y0203_yopG	1.9	1.7	2.8	3.7
Y0204_yopG	1.9	1.7	2.8	3.7
Y0205_yopG	1.9	1.7	2.8	3.7
Y0206_yopG	1.9	1.7	2.8	3.7
Y0207_yopG	1.9	1.7	2.8	3.7
Y0208_yopG	1.9	1.7	2.8	3.7
Y0209_yopG	1.9	1.7	2.8	3.7
Y0210_yopG	1.9	1.7	2.8	3.7
Y0211_yopG	1.9	1.7	2.8	3.7
Y0212_yopG	1.9	1.7	2.8	3.7
Y0213_yopG	1.9	1.7	2.8	3.7
Y0214_yopG	1.9	1.7	2.8	3.7
Y0215_yopG	1.9	1.7	2.8	3.7
Y0216_yopG	1.9	1.7	2.8	3.7
Y0217_yopG	1.9	1.7	2.8	3.7
Y0218_yopG	1.9	1.7	2.8	3.7
Y0219_yopG	1.9	1.7	2.8	3.7
Y0220_yopG	1.9	1.7	2.8	3.7
Y0221_yopG	1.9	1.7	2.8	3.7
Y0222_yopG	1.9	1.7	2.8	3.7
Y0223_yopG	1.9	1.7	2.8	3.7
Y0224_yopG	1.9	1.7	2.8	3.7
Y0225_yopG	1.9	1.7	2.8	3.7
Y0226_yopG	1.9	1.7	2.8	3.7
Y0227_yopG	1.9	1.7	2.8	3.7
Y0228_yopG	1.9	1.7	2.8	3.7
Y0229_yopG	1.9	1.7	2.8	3.7
Y0230_yopG	1.9	1.7	2.8	3.7
Y0231_yopG	1.9	1.7	2.8	3.7
Y0232_yopG	1.9	1.7	2.8	3.7
Y0233_yopG	1.9	1.7	2.8	3.7
Y0234_yopG	1.9	1.7	2.8	3.7
Y0235_yopG	1.9	1.7	2.8	3.7
Y0236_yopG	1.9	1.7	2.8	3.7
Y0237_yopG	1.9	1.7	2.8	3.7
Y0238_yopG	1.9	1.7	2.8	3.7
Y0239_yopG	1.9	1.7	2.8	3