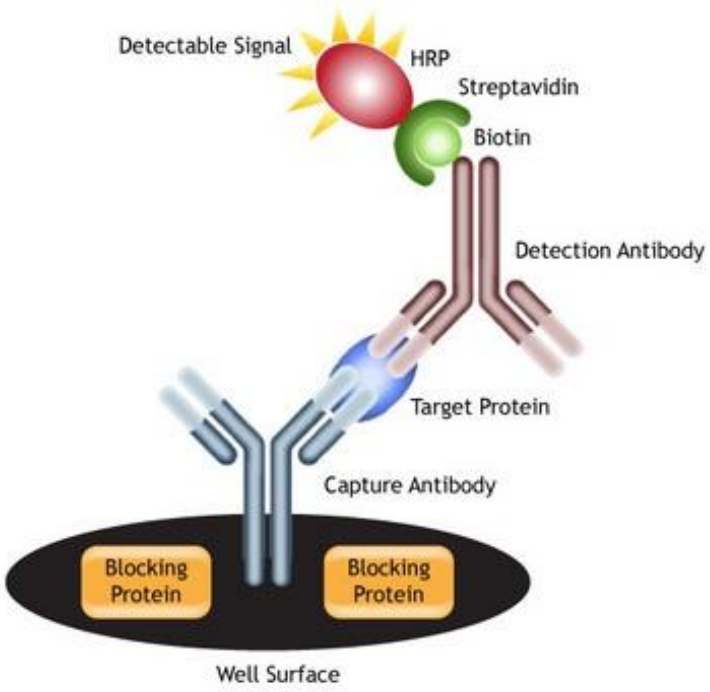


QUANSYS BIOSCIENCES

Competitive and Sandwich ELISAs

Sandwich ELISAs

- Primary or capture antibody is applied to the well.
- The sample containing the antibody's target (antigen) is added.
- The complimentary or match pair antibody which is labeled is added to the well.
- If the antigen was present in the sample, it has then bound to the capture antibody and hence the detection binds, building the antibody/antigen complex.
- The labeled detection will then be read or detected.



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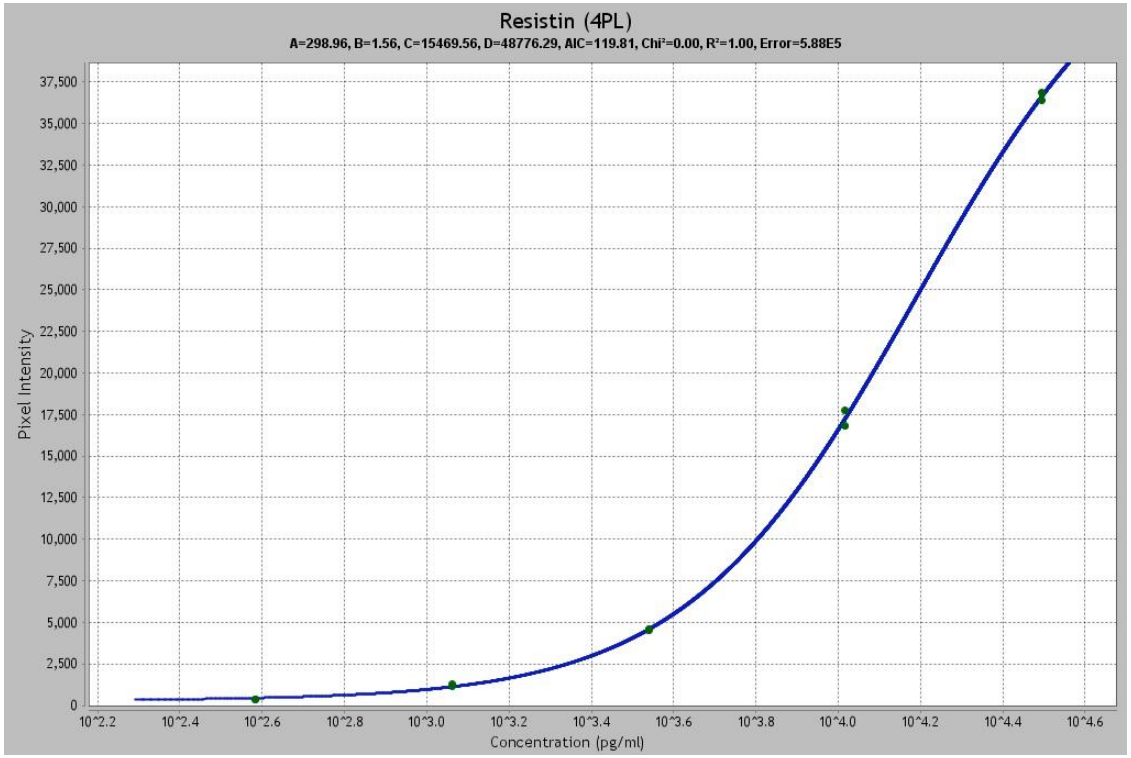
Sandwich ELISAs

- Pros:
  - Sensitive
  - Standardized method
- Cons:
  - More steps in Assay
  - Two antibodies
  - Only applicable on large molecules



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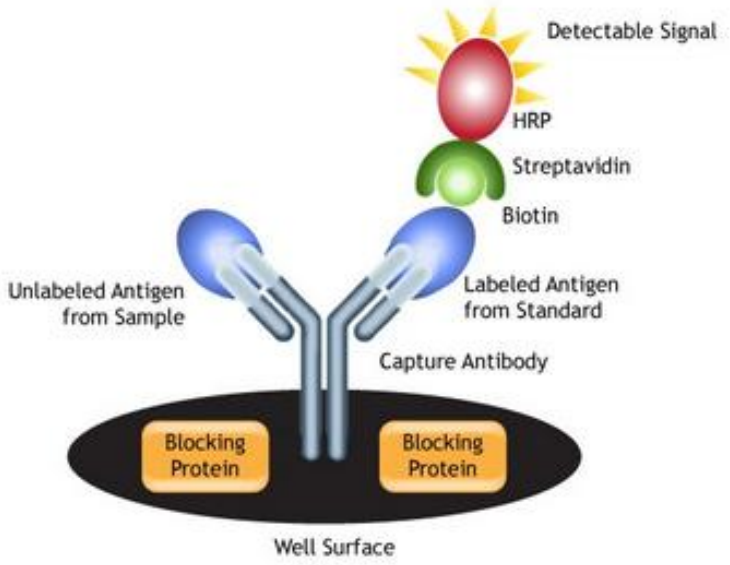
Competitive ELISAs

- Primary or capture antibody is applied to the well.
- The sample containing the antibody's target (antigen) is added.
- In the next step, labeled antigen is added.
- Whatever binding sites remain, they will be occupied by the labeled antigen
- The detector (SHRP) is added.
- Where no antigen was present in the sample, the labeled antigen will have bound and create a signal.
- If the sample contained antigen, no labeled antigen will bind, causing no signal. -inverse curve



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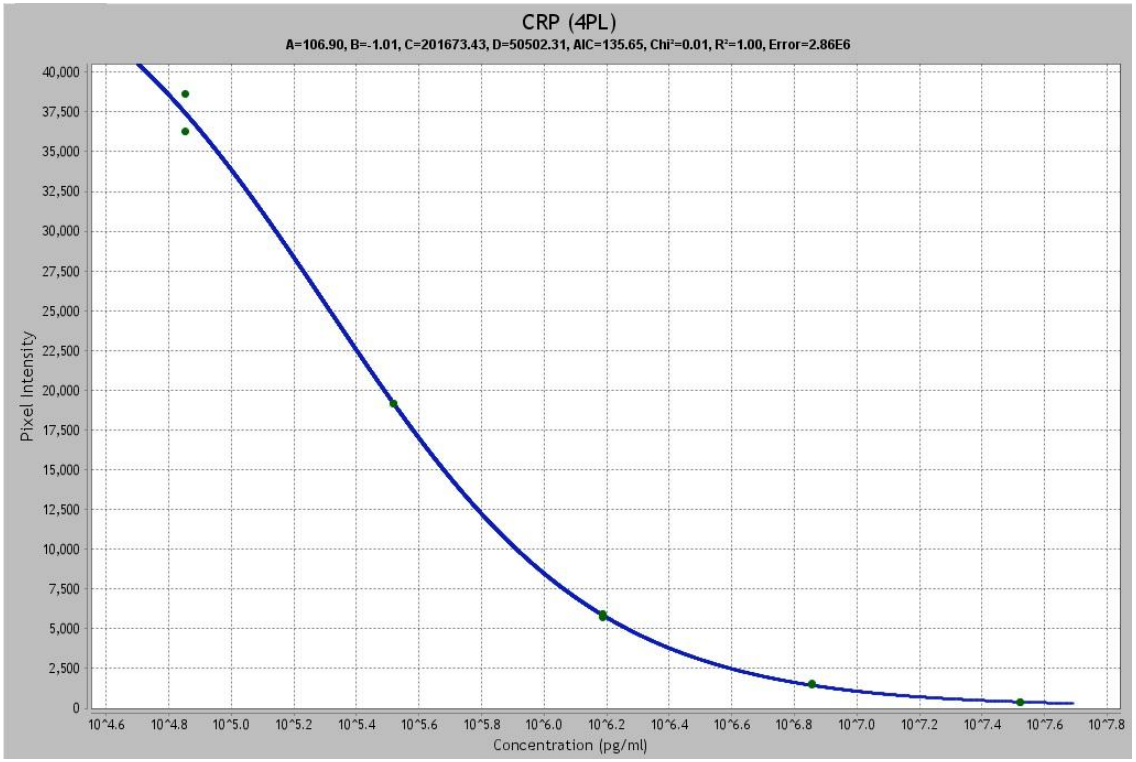
Competitive ELISAs

- Pros:
  - Needs one antibody
  - Small molecules
- Cons:
  - Less sensitive



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Development Strategy

- Solution: Combine both sandwich and competitive ELISA in the same panel of assays so a single dilution is used.
- Benefits:
  - All assays are within one well.
  - Panels with different ranges (ug/ml and ng/ml) can be multiplexed by converting sandwich ELISAs to competitive to alleviate multiple dilutions
  - When only one antibody is available for an assay in a panel, the assay no longer becomes a show-stopper
  - Small and large molecule assays can still be in the same panel

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CASE STUDY # 1

- Validation of Custom Multiplex Array Against Individual ELISAs
  - Simon Fraser University, BC, Canada
  - Female Reproductive Health: Urine samples from Guatemala
  - Adiponectin, Cortisol, E1G, FSHb, HCGb, and C-Peptide
  - 4 sandwich ELISAs and 2 competitive ELISAs in one well
  - Testing in parallel to Bayer ACS:180 Clinical Analyzer
    - Pearson Correlation Coefficient: (≥0.75)
  - Am J Hum Biol. 2012 Jan-Feb;24(1):81-6. doi: 002/ajhb. 21229. Epub 2011 Nov 28.

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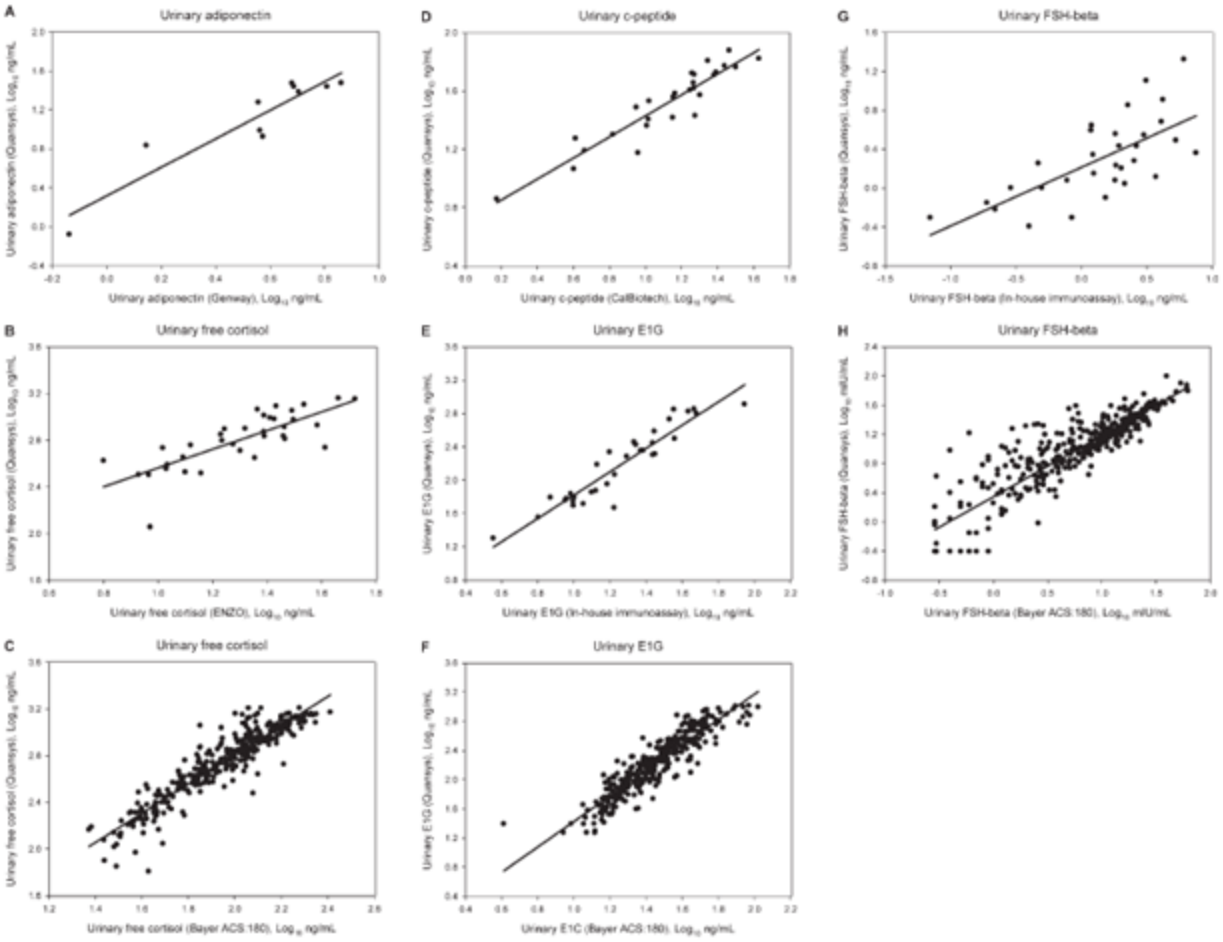
CASE STUDY # 1

SFU

- Correlation between two methodologies



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CASE STUDY # 1

SFU

- Assay Performance: Sensitivity and Reproducibility

	Quansys Multiplex			Traditional ELISAs		
	Sensitivity	Intra-Assay CV	Inter-Assay CV	Sensitivity	Intra-Assay CV	Inter-Assay CV
Adiponectin	0.023 ng/ml	10%	6.90%	0.156 ng/ml	4.4%	6.2%
Free Cortisol	0.343 ng/ml	7.30%	8.50%	0.057 ng/ml	10.5%	13.4%
C-Peptide	0.090 ng/ml	9.30%	7.70%	2 ng/ml	3.9%	8.5%
E1G	0.252 ng/ml	9.70%	8.20%	1.45 ng/ml	7.9%	8.5%
FSHb	0.017 ng/ml	7.20%	7.30%	0.143 ng/ml	3.8%	6.5%
HCGb	0.035 ng/ml	7.10%	7.50%	0.003 ng/ml	3.5%	5.8%

- Summary: “This multiplex technology provides a more economic, rapid, and ecologically sound alternative to individual assays for studies requiring the measurement of multiple biomarkers per biospecimen.”

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Questions?

Thank You!

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