

***Pressure BioSciences, Inc.***

***Pressure Cycling Technology (PCT)***

***A Novel, Enabling Platform  
Revolutionizing Biomarker Discovery***

***BioPharm America 2010  
Boston, MA***

***September 16, 2010***



## *Forward Looking Statements*

This presentation may contain forward looking statements that reflect management's current views and opinions as to the status of the Company's products, technology and other future events and operations. These statements are neither a promise nor guarantee, but involve risks and uncertainties that could cause actual results to differ materially from those anticipated or indicated. Investors are cautioned that any forward looking statements should be considered in light of such risks and uncertainties including, without limitation, those detailed in the Company's filings with the Securities and Exchange Commission.



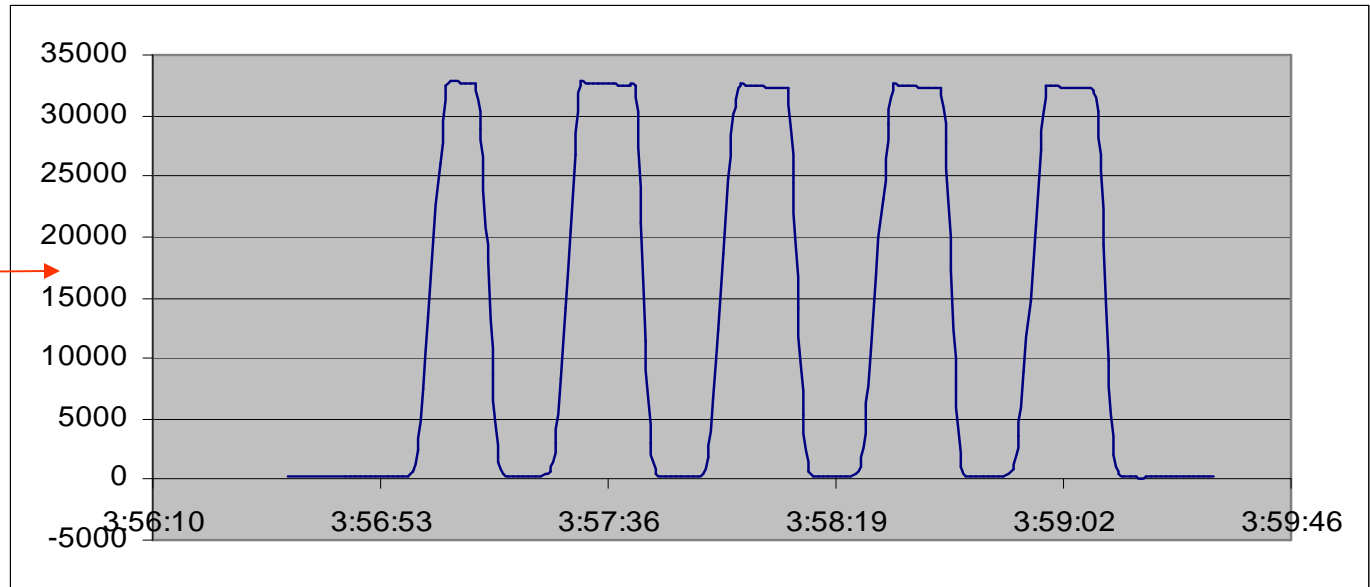
## *Pressure BioSciences, Inc.*

- Formed Sept 2004 - Sale of Boston Biomedica (BBI)
- NASDAQ CM: PBIO
- Started Operations in February 2005
- Began Instrument/Consumables Sales in Late 2007
- Fourteen (14) Employees
- Strong Management Team and Board of Directors
- 24 Issued Patents...Many More in Pipeline
- Focused on the Development and Commercialization of a Powerful, Proprietary, Enabling Platform  
*Pressure Cycling Technology (“PCT”)*



# Pressure Cycling Technology (PCT)

16,000 PSI  
Marianas  
Trench

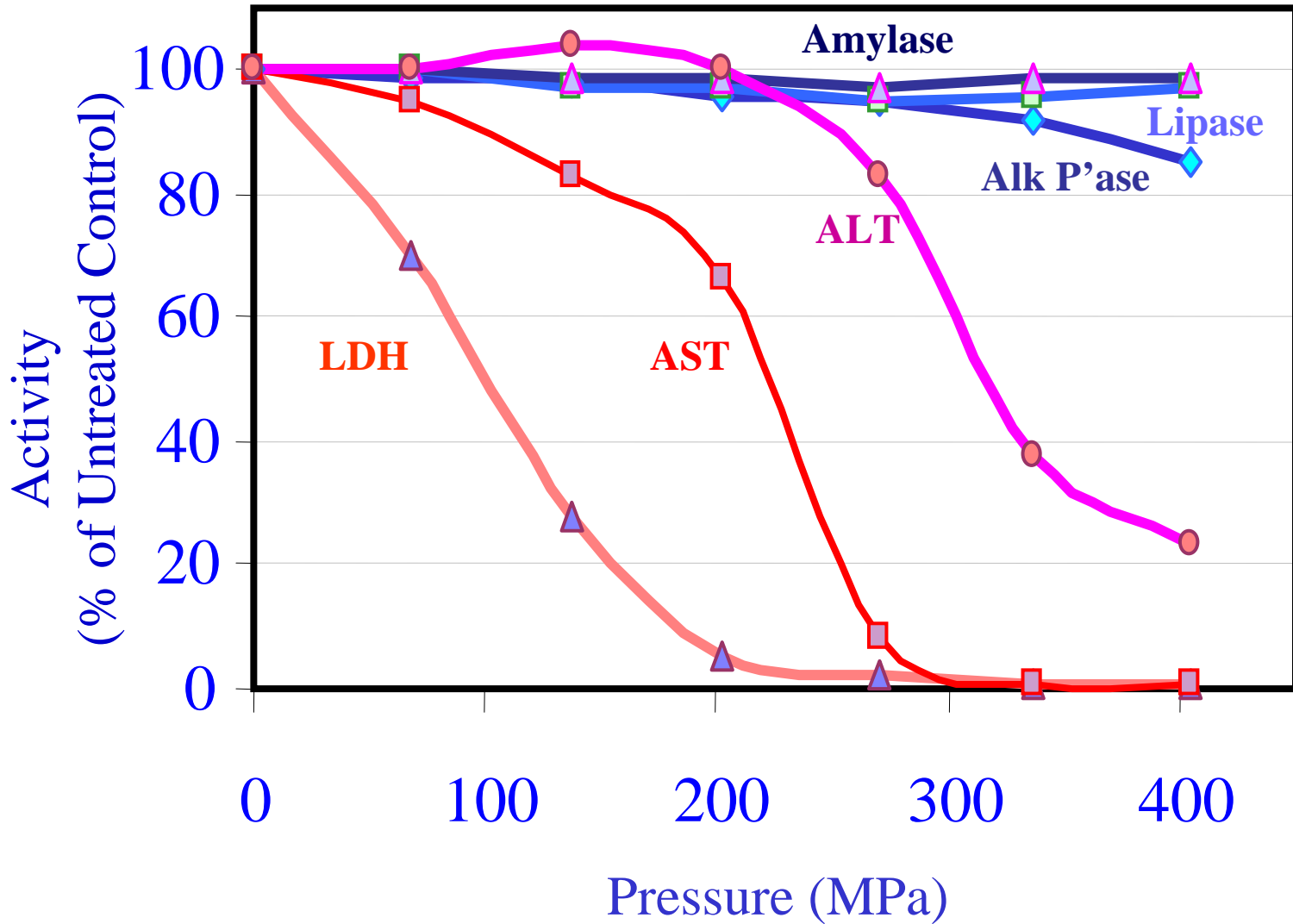


Cycles of hydrostatic pressure between ambient and ultra high levels, which allow for the precise control of biomolecular interactions

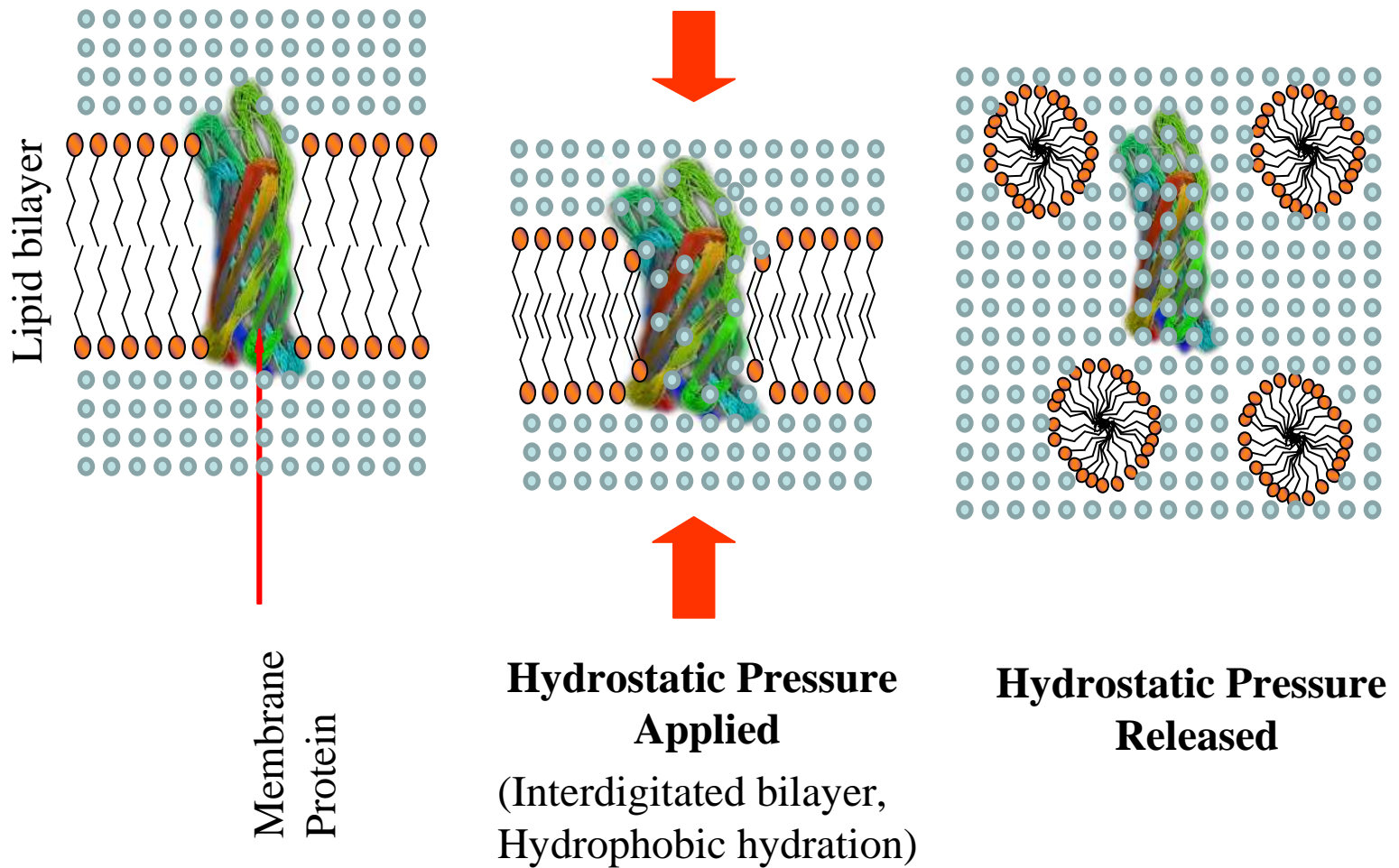
## *Biomolecules Under Pressure - Proteins*

- Pressure promotes **dissociation** of oligomeric proteins
- Pressure promotes protein **unfolding and re-folding**
- Unfolding is driven by pressure induced **hydration**
- Pressure can lead to **protein denaturation**
- Pressure activates most **hydrolytic enzymatic reactions**
- Pressure **protects proteins** from thermal denaturation
- Pressure denatured proteins **retain a relatively compact structure with water molecules penetrating their core**

## Effect of High Pressure on Enzymatic Activity



# High Pressure Destabilizes Biological Membrane Equilibria



# *“Garbage In...Garbage Out”*



Scientific research can be broken down to three key elements:  
Sample Preparation, Analysis, and Interpretation  
**“Discovery Starts with Sample Preparation”**

# Sample Preparation

## Value Proposition

- Analysis of DNA, RNA, Proteins, Lipids, and Small Molecules, is Crucial to the Development of New Diagnostics & Therapeutics
- Sample Preparation Crucial but Challenging...*often a bottleneck/barrier*
- PCT Offers Clear Advantages Over Competitive Products
- PCT Offers New Thermodynamic Mechanisms to Sample Preparation
- Early Traction....*over 115 PCT System installs over past three years, in top pharma, biotech, academic, and government US labs*
- PCT Precisely Controls Test Variables
- PCT Lends Itself Well to **Standardization**
- **Discovery Starts with Sample Preparation**

# Current Sample Preparation Methods

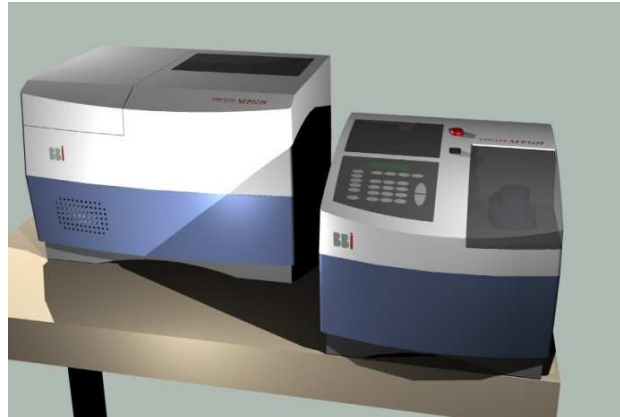
- Mortar & Pestle
- Dounce homogenizer (glass on glass)
- Potter-Elvehjem homogenizer (Teflon on glass)
- Enzymatic Digestion
- Polytron shearing homogenizers
- Blenders
- Bead Beating
- Sonication
- Repeated Freeze/Thaw cycles
- French Press ( $\leq 2000$  PSI)



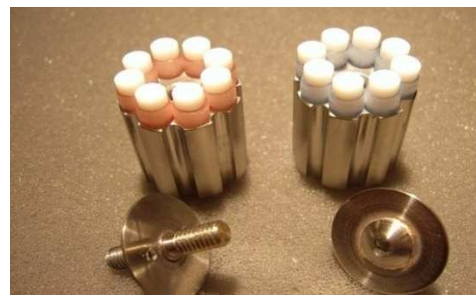
# PCT – Sample Preparation System



Barocyler™  
NEP2320



Barocyler™ NEP3229



# The PCT Shredder

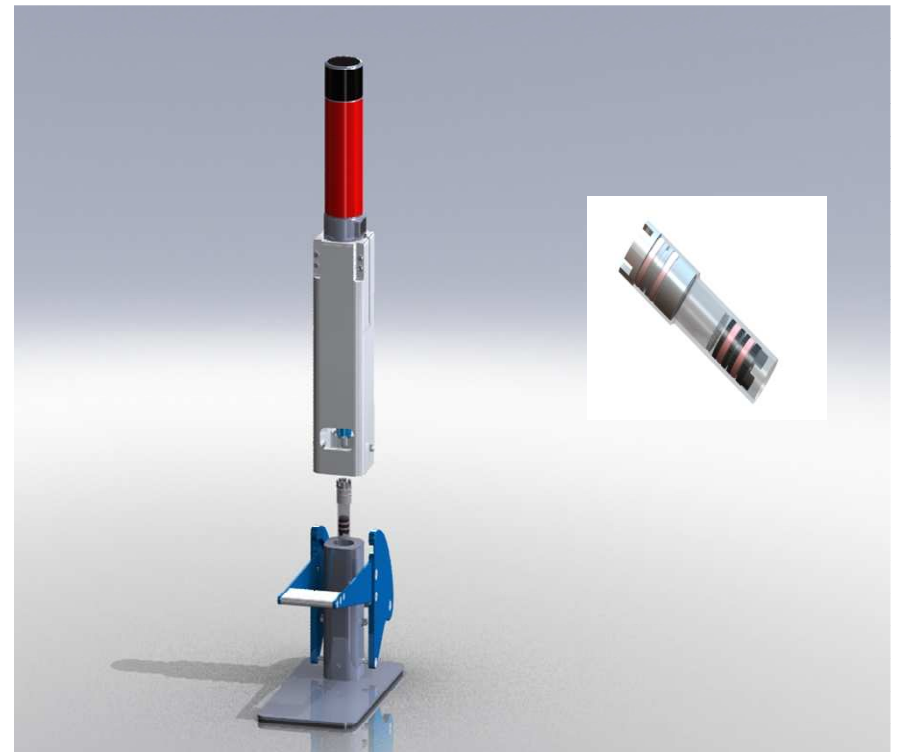
## PCT Shredder (1<sup>st</sup> GEN)

- NiCad Batteries
- Pressure set by pushing Driver
- Pressure levels -15,30,45 lbs Force
- PCT PULSE or Shredder Tubes



## PCT Shredder (3<sup>rd</sup> GEN)

- Long lasting lithium rechargeable batteries
- Paddle for convenient pressure setting
- Pressure levels -15, 30,45 lbs Force
- PCT PULSE or Shredder Tubes
- Heavy Duty and Robust Driver



# Barocyler HUB440

## First Programmable Bench-top High Pressure Generator (500 – 60,000 psi)

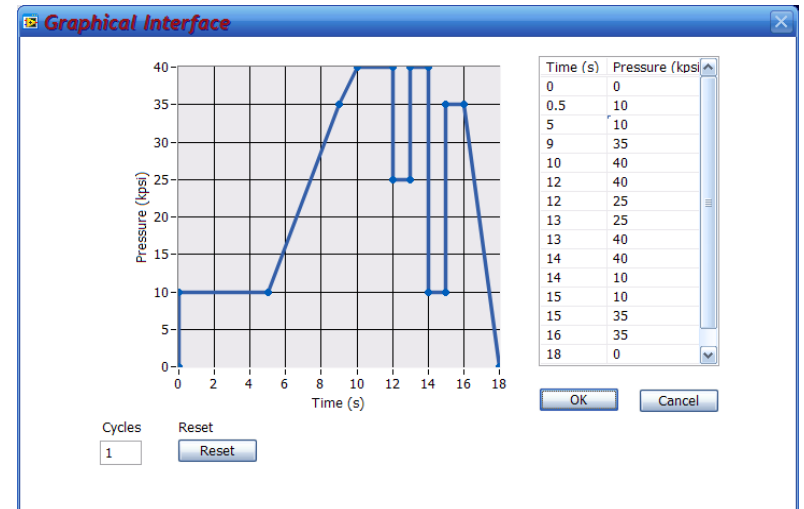
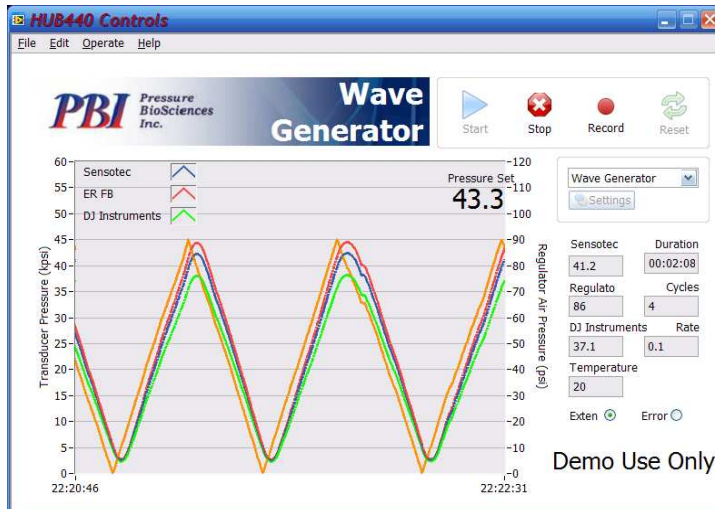
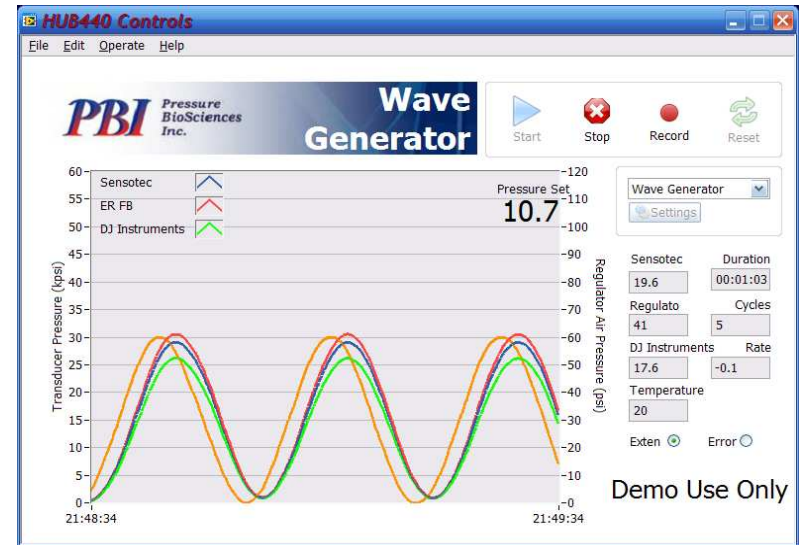
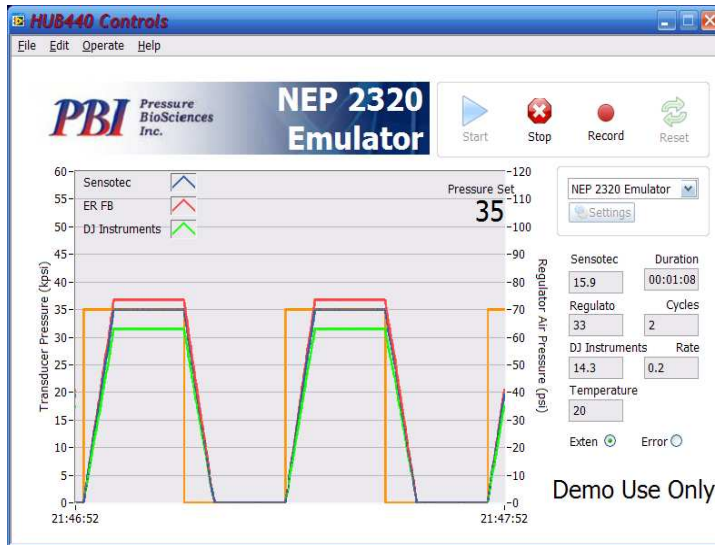
- USB-powered computer interface
- Precise control of low - high pressure/time
- Data logging (Labview software)
- Interface with spectroscopy, HPLC, and other analytical instrumentation

### Applications

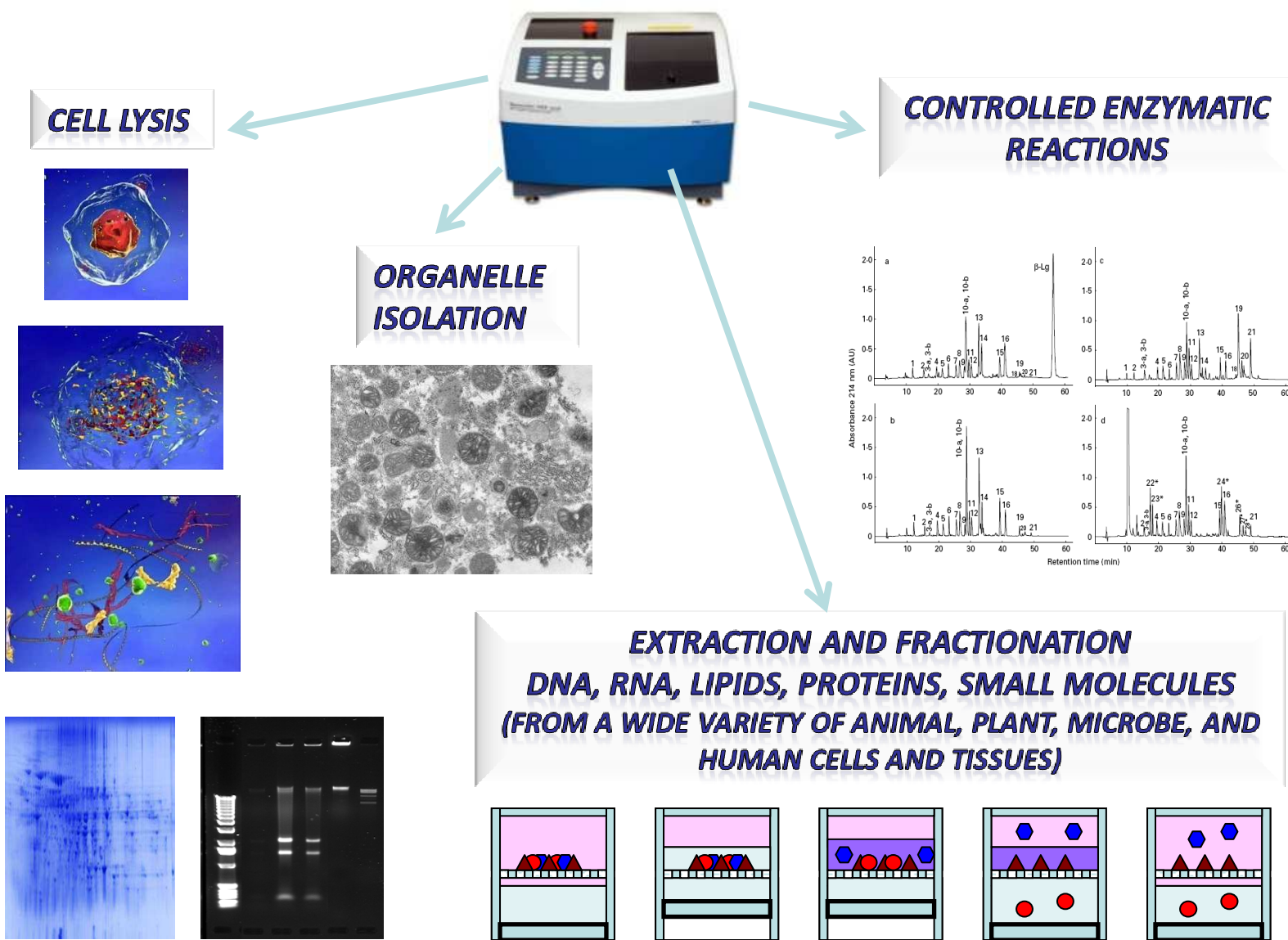
Allows exquisite control of low to ultra-high pressure perturbation of macromolecular structure for magnetic resonance and optical spectroscopy, chemical synthesis, and material science.



# HUB Controls v.1.01 User Interface



# PCT Sample Preparation Applications



# *Lawrence Berkeley Nat'l Laboratory*

“...we have chosen PCT because of greater NA and protein yields from low concentrations of microorganisms, as compared to other methods.”

Dr. Janet Jansson, Sr. Staff Scientist



# High Pressure Symposium: Harvard Medical School

“Applications of Ultra-high Pressure in Biotechnology – May 21, 2010”

•Harvard Medical School	Kuo	Conference Chair
•Harvard School of PH	Ivanov	Membrane Proteins
•The Barnett Institute (NEU)	Szabo	De-glycosylation
•University of Montreal	Bonneil	Enhanced Proteolysis
•FDA	Kehtie	Influenza Vaccine QC
•USAMRIID	Powell	Biological Threats
•Pacific NW Nat'l Labs	Lopez-Ferrer	In-line Protein Digest
•University of Missouri	Pevsner	Colon Cancer
•University of N. Texas	Budowle	Forensic Sciences
•Med Diagnostic Labs	Tilton	Ticks (Lyme Disease)
•Harvard Medical School	Smejkal	Paleontology
•Target Discovery	Schneider	Ovarian Cancer
•AFIP	Fowler	FFPE Proteomics

Invited guests include local scientists, customers, potential customers, potential strategic partners, research analysts, print media, electronic media, investors, shareholders, & thought leaders.

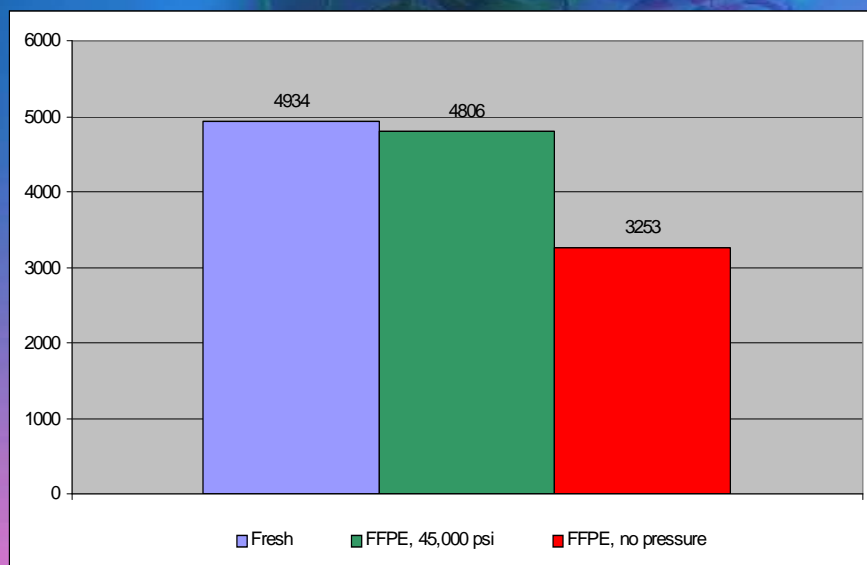
# ***Recovery and ID of Proteins from FFPE Samples***

**FFPE Samples Are An Enormous Untapped Repository (~ 1 Billion) for Discovery, Validation, & Measurement of Disease Detection and Progression, Companion Diagnostics, Drug Response, and Toxicity**

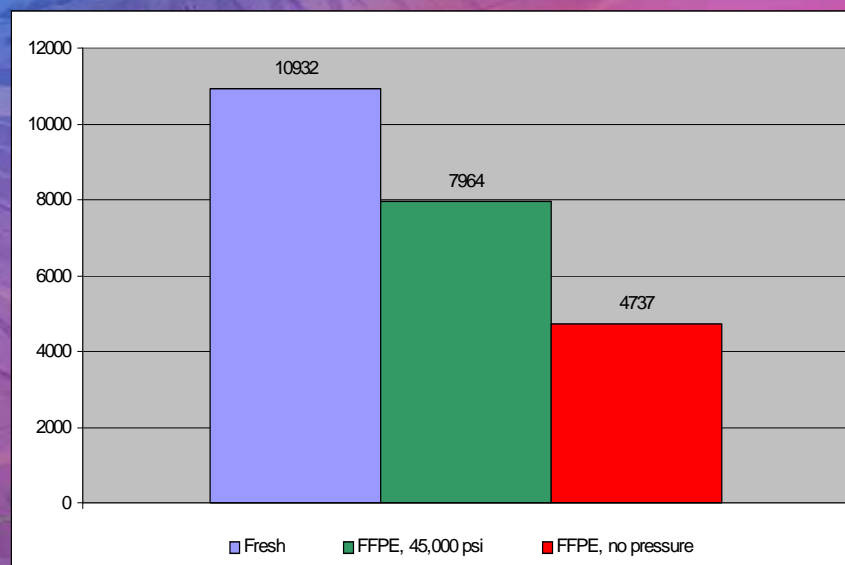
## **Literature Search: Recent Studies Utilizing FFPE**

- **Lung Cancer Metastasis Biomarkers**
- **Breast Cancer Biomarker Expression**
- **Human Epidermal Growth Factor Receptor 2 (HER2)**
- **Frozen & FFPE Colon Adenoma Tissue - 92% Protein Overlap**
- **Measurement of EGFR Protein and Activation**
- **Nasopharyngeal Carcinoma (NPC) – Expression Levels**
- **p53 expression**

# Pressure Cycling Technology (PCT) Significantly Improves Recovery of Proteins/Peptides from Formalin Fixed Paraffin-Embedded (FFPE) Tissue



Unique Proteins Identified



Unique Peptides Identified

Data Courtesy of Dr. C. Fowler (AFIP)

"Our initial data show that for aorta samples, which are traditionally difficult to lyse, a greater amount of protein is recovered following pressure-enhanced FFPE extraction, compared to the standard non-pressure method."

Dr. J. Van Eyk, Director, The Hopkins NHLBI Proteomics Center, John Hopkins

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**Please Visit Us At Our  
Exhibit Booth**

**PBI**