



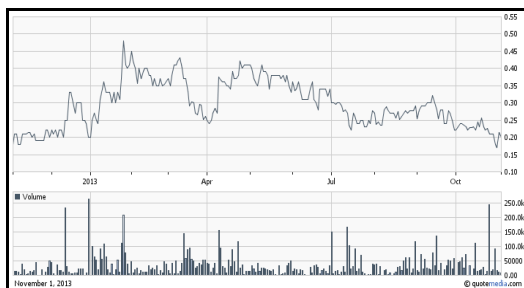
**Pressure BioSciences, Inc.**

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Ticker (Exchange)	PBIO (OTC)
Recent Price (11/01/2013)	\$0.20
52-week Range	\$0.15 - \$0.51
Shares Outstanding	~11.4 million*
Market Capitalization	~\$2.3 million
Daily Avg. 3-month Volume	33,808
Insider Ownership +>5%	15%
Institutional Ownership	—
EPS (Qtr. ended 06/30/2013)	(\$0.10)
Employees	10

\*As of August 14, 2013.

**Pressure BioSciences, Inc. (PBIO-OTC)**



**Company Description**

Pressure BioSciences, Inc. (“Pressure BioSciences” or “the Company”) develops and commercializes proprietary laboratory instruments and consumable products. Its product portfolio is based on its patented pressure cycling technology (PCT) platform, which is able to apply and release high levels of **hydrostatic†** pressure in a safe and controlled manner. While this technology has applications across many industries, Pressure BioSciences is initially using PCT to develop products that improve **sample preparation**—the process of preparing a sample for analysis. In life science research, sample preparation is integral to tens of thousands of laboratories worldwide. Pressure BioSciences’ line of Barocycler® products enables scientists to harness the power of PCT in the lab. **Biological samples** (e.g., cells and tissues from humans, animals, plants) placed in a Barocycler are exposed to programmable cycles of low-to-intense pressure—up to 58,000 pounds per square inch (psi). This act of “cycling” pressure can accelerate and control physical and chemical processes, including the breakup of cellular structures and release of biomarkers from a sample for subsequent testing and analysis. Pressure BioSciences’ portfolio also includes Shredder instruments (for rapidly shredding or grinding samples), sample preparation kits and reagents, and related consumable products (e.g., tubes to hold the samples during the PCT process).

**Key Points**

- The sample preparation market could reach \$5.4 billion in 2013, with potential to more than double to \$11.5 billion by 2018 due to new technology and growing demand.
- The Company has initially focused on PCT tools to enhance **biomarker** discovery, a key part of vaccine, diagnostic, and therapeutic R&D; and forensic analysis, which may improve DNA evidence processing and lead to higher arrest/conviction rates.
- PCT has been shown to extract a higher quality and quantity of proteins, nucleic acids (DNA/RNA), and small molecules from samples versus competitive laboratory products and processes. This is expected to have positive implications in biomarker discovery, forensics, and many other areas of life science research.
- The Company has installed 235 units to date. Customers include large pharma/biotech companies (Bristol-Myers, Amgen, Merck), U.S. government agencies (FBI, FDA, NIH, USDA), and universities (Harvard School of Public Health, Stanford, UCLA). Many users have performed studies using PCT. In total, over 100 papers have been published on diverse applications for PCT.
- Pressure BioSciences emphasizes continued innovation, with multiple novel products in its R&D pipeline, including a high-throughput PCT system designed to meet the automated processing needs of larger clinical diagnostics labs and R&D organizations.
- The Company has 24 issued and 6 pending patents globally that protect aspects and applications of its PCT platform.
- At June 30, 2013, the Company’s cash position was \$113,000.

†BOLD WORDS IN CONTEXT ARE REFERENCED IN THE GLOSSARY ON PAGES 73-75. See inside for applicable disclosures.

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## Executive Overview

Pressure BioSciences, Inc. (“Pressure BioSciences” or “the Company”) is focused on the research, development, and commercialization of proprietary laboratory instruments and consumable products based on its patented pressure cycling technology (PCT) platform. PCT is an enabling technology that is able to create, apply, and release high levels of hydrostatic pressure—currently up to 58,000 pounds per square inch (psi)—quickly and in a safe and controlled manner. As a frame of reference, in the deepest part of the Mariana Trench, which is approximately 35,800 feet under the ocean’s surface and the deepest point on Earth, the pressure is roughly 17,920 psi or nearly nine tons per square inch—roughly the weight of an elephant balanced on a postage stamp. Not only has Pressure BioSciences developed a means to harness high pressure in a compact instrument that fits comfortably on a laboratory workbench, but the Company has also incorporated and patented its unique ability to rapidly cycle pressure on and off.

Pressure is force applied to a surface area. It is a fundamental variable in thermodynamics, and can be used to manipulate matter between amorphous and various crystalline solids, liquid, gas, and plasma physical states, without requiring changes in temperature. Easy access to pressure cycling capabilities can provide a powerful and important tool for many areas of scientific research and materials processing.

PCT is expected to significantly benefit research and routine processing applications across many industries, including the life sciences, diagnostics, pharmaceutical, biotechnology, forensics, food, anti-bioterror, agriculture, automotive, and oil and gas industries, among many others. Extensive demonstrations and data collected internally and by third-party researchers have confirmed the potential of PCT across many of these areas. While Pressure BioSciences could ultimately develop products specially designed for each market, management has evaluated its core competencies and intellectual property strengths and thus selected sample preparation—a critically important process that dramatically affects outcomes in life sciences research and patient diagnostics—as its initial target market. The Company has 24 issued and 6 pending patents worldwide protecting the application of PCT in sample preparation and other areas.

### First Application for Pressure Cycling Technology (PCT): the Sample Preparation Market

Pressure BioSciences is initially using PCT to develop products that improve “sample preparation,” or the process of preparing a biological sample (such as cells and tissues from human, animal, plant, and microbial sources) for analysis. Sample preparation is a crucial laboratory step for most areas of life sciences research and commercial diagnostics applications. Sample preparation is required prior to virtually all scientific analyses, and can be pivotal for protecting such scientific analyses from the ancient conundrum of “garbage in yields garbage out.” Sample preparation processes vary but, within the life sciences, often include breaking up the cellular structures of a sample (“cell lysis”) as well as separating or extracting out various cell components and biomolecules or biomarkers, including proteins, lipids, **small molecules**, and nucleic acids (DNA/RNA). Accurate analysis of these extracted biomolecules is crucial to the development of new diagnostics and therapeutics.

There are a number of tools and techniques currently used in sample preparation, including **bead beaters, sonicators, homogenizers, mortar and pestle grinders, French Press, freezer mills, enzymatic digestion, and chemical dissolution**. While sample preparation is intended to facilitate and improve accurate scientific analysis, conventional methods for performing sample preparation are often complex, time consuming, and error-prone. The University of Florida has identified human error as a major liability in sample preparation. Additionally, it is estimated that laboratory professionals spend as much as 80% of their time preparing samples for analysis (Source: *Lab Manager Magazine*, July 13, 2011). Important cell components can be lost, degraded, damaged, or otherwise affected during the process of breaking up and separating the components of the cell. While scientists can use cutting-edge devices to analyze what remains in the prepared sample, cell components lost or damaged during the sample preparation process will not be found in the final analysis of the sample, hindering the scientists’ ability to obtain relevant and useful results. For this reason, sample preparation is widely considered to be the main bottleneck in life science research.

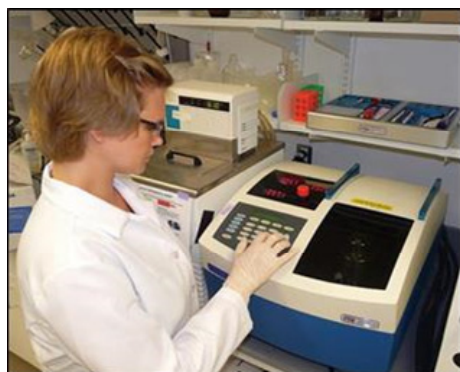
*PCT Is Designed to Substantially Improve Sample Preparation*

Pressure BioSciences believes that its PCT methods offer several important advantages over current sample preparation products and processes. First, researchers at the U.S. Food and Drug Administration (FDA), the Harvard School of Public Health, the Armed Forces Institute of Pathology, the Johns Hopkins School of Medicine, and other institutions have shown that using the PCT platform in their sample preparation workflows resulted in significant advantages, including major reductions in the total time for analysis and improved reliability and reproducibility of testing and quality of results. Next, when used as part of the sample preparation process, PCT was shown to improve the quality and quantity of DNA, RNA, proteins, lipids, and small molecules recovered from a sample versus many available preparation techniques. Third, and perhaps most important of all, PCT has proven to dramatically increase the range of unique versions (often never previously seen) of biomolecules that were liberated and presented for analysis.

PCT helps to make sample preparation reproducible and enables standardization. Conventional bead beaters use small beads to aggressively disrupt the cells and their components in a sample, and mortar and pestle tools require extensive and laborious grinding of samples into smaller pieces. In contrast, PCT uses cycles of hydrostatic pressure to gently, rapidly, and reproducibly liberate and preserve the widest range of DNA, RNA, small molecules, and proteins from almost any biological fluid, cells, or tissue sample. Unlike the application of temperature as a process variable, the application of pressure is homogeneously experienced instantly and equally throughout the sample being treated. Repeated cycles of PCT help to ensure the effective liberation of released biomolecules from within the structural matrix of the sample, in a fashion somewhat analogous to the repeated squeezing of a wet sponge.

It is important to note that PCT is not designed to replace mainstream laboratory analysis instruments used to analyze biological samples; rather, it is designed to enhance the sample preparation process for use in tandem with downstream analyzers to recover greater quantities, higher quality, and previously unavailable biomolecules from the sample.

Figure 1  
BAROCYCLER®



Source: Pressure BioSciences, Inc.

**Pressure BioSciences' Product Portfolio**

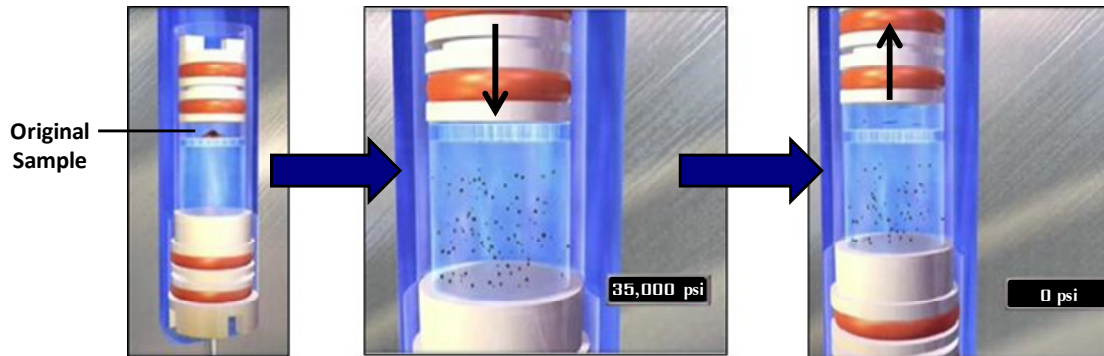
The Company's first commercial application of PCT for sample preparation is the Barocycler® family of bench-top laboratory instruments. Biological samples placed in the pressure chamber of a Barocycler are exposed to programmable cycles of low-to-intense pressure. This act of "cycling" pressure quickly breaks up the cellular structures of a sample for downstream testing and analysis. An example of a Barocycler is shown in Figure 1. The Company's first two Barocycler models—NEP2320 and NEP3229—produce up to 35,000 psi and vary in sample capacity, size, and weight. The 65-pound NEP2320 model lists for around \$30,000 and provides a sample chamber suitable for processing, while the larger NEP3229 model weighs roughly 350 pounds and lists for approximately \$45,000 but has the capacity to simultaneously process up to three times the number of samples as the smaller NEP2320.

To prepare a biological sample using a Barocycler, the user first places the sample in a consumable processing sample tube that is specially designed to withstand high levels of pressure, and fills the remainder of the tube with solutions called "processing buffers." The user then inserts the sample tube into the chamber of the Barocycler and specifies the desired maximum level of pressure and number of cycles to subject the sample to (or can program more finely-grained pressure cycle profiles, if desired), before starting the machine.

Figure 2 (page 5) shows a rendering of the PCT process, with a maximum set pressure of 35,000 psi (NEP3229). As shown in the center of Figure 2, the increase in pressure forces the sample to pass through a disc with small holes (called a "lysis disc"), breaking the sample into smaller pieces that disperse throughout the processing buffers. As the pressure cycles back to ambient psi (about 14.5 psi), fragments of the sample are pulled back through the lysis disc. This process repeats for the number of cycles designated by the user.

Figure 2

HOW PRESSURE CYCLING TECHNOLOGY WORKS



Source: Pressure BioSciences, Inc.

*Next-Generation PCT System Offers Increased Versatility and Higher Margins*

Pressure BioSciences' next-generation PCT platform is the HUB family of Barocyclers. HUB models are designed to serve as a hub for a laboratory's high-pressure needs, with users purchasing accessories as needed to expand the functionality of their HUB unit. Unlike the Barocycler models, which have a set chamber size that can fit a specified number of samples in each test run, HUB models offer flexibility that enables the laboratory's pressure cycling capabilities to grow and adapt with the laboratory's needs. Additionally, the HUB concept allows Pressure BioSciences to focus on the development of accessories that target additional and new markets.

In the coming years, the Company expects the HUB platform to become the central workhorse in its PCT-based instrument line. The HUB technology is in a late-stage R&D phase, and the Company has begun early commercialization, with initial HUB units already purchased and installed in approximately 10 collaborator facilities. The current model, the HUB440, weighs 55 pounds, provides up to 58,000 psi, and lists for roughly \$30,000. Pressure BioSciences is currently working on development of a HUB model that can reach 100,000 psi, which to the Company's knowledge, would be the first device of its kind to do so.

*Consumables and Other Products for Sample Preparation*

In addition to the Barocycler instruments, the Company markets a line of Shredder instruments for rapidly shredding or grinding bulky or particularly tough samples down to manageable sizes for further processing. The Shredder line is detailed on pages 29-31 of the Core Story of this report.

With a keen eye on the perennial "razor and razor blades" business model, Pressure BioSciences also sells many single-use, consumable products, including PULSE Tubes and MicroTubes to hold each sample being processed, as well as a variety of prepackaged sample preparation kits and reagents that provide additional specialized capabilities tailored to particular sample processing challenges. All of the consumables are designed to be used in conjunction with the Barocycler and Shredder products. The Company anticipates that these consumable products could eventually grow to become the leading revenue-generating item and profit center in its PCT products mix. These consumable products are described on pages 31-33 of the Core Story.

**Enabling Platform Technology with Broad Applications**

Pressure BioSciences is initially developing PCT to improve sample preparation. The sample preparation market is among the fastest-growing segments of the life sciences industry. It is forecasted to reach \$5.4 billion in 2013, with potential to more than double to \$11.5 billion in 2018 (Source: BCC Research, *Sample Preparation in Genomics, Proteomics, and Epigenomics: Global Markets*, September 2013). The Company estimates that 500,000 scientists in 80,000 laboratories globally perform sample preparation procedures. The use of PCT to prepare samples could contribute to the development of better therapeutics, diagnostics, and vaccines, as well as enable more accurate research into diseases and disorders, including leading causes of death like heart disease and cancer, and major chronic diseases like obesity, diabetes, and Alzheimer's disease.

Within the expansive sample preparation market, the Company is developing PCT for two key areas: (1) biomarker discovery, a key part of vaccine, diagnostic, and therapeutic R&D (detailed on pages 36-41); and (2) forensics, which may improve processing of DNA evidence and lead to higher arrest/conviction rates (pages 41-48). The ability for PCT to extract unique biomolecules that have potential to be important biomarkers has been studied by respected institutions and scientists, such as the FDA and the Harvard School of Public Health. Pressure BioSciences has partnered with several leading forensic institutions, including the Henry C. Lee Institute of Forensic Science, Florida International University's International Forensic Research Institute (IFRI), and the University of North Texas Health Science Center's Institute of Applied Genetics, to accelerate the development of its technology for this market.

Some additional potential markets for PCT include pathogen inactivation, anti-bioterror applications, protein purification, control of chemical reactions (particularly enzymatic reactions), immunodiagnosics, food safety, vaccine development, and DNA sequencing, including next-generation and whole genome sequencing.

### Key Market Leaders Have Recognized the Benefits of PCT

PCT systems have been used by customers and collaborators across a range of industries. Pressure BioSciences' strategic collaboration program allows researchers and their laboratory teams to lease a Barocycler under favorable terms for three to six months in exchange for collecting and sharing data on their diverse variety of applications for the PCT system. These relationships have led to over 100 publications in peer-reviewed journals as well as presentations at major industry events. The Company anticipates that this investment in strategic collaborations will increase visibility and marketplace adoption of its PCT technology and products, as commercialization is accelerated.

As shown in Figure 3, Pressure BioSciences' customers have included the FDA, the National Institutes of Health (NIH), U.S. Centers for Disease Control and Prevention (CDC), Federal Bureau of Investigation (FBI), and U.S. Department of Agriculture (USDA). Academic customers have included Harvard Medical School, Stanford University, and the University of California at Los Angeles (UCLA). As well, the Company has diagnostic, biotechnology, and pharmaceutical customers, including Amgen, Inc. (AMGN-NASDAQ), Biogen Idec Inc. (BIIB-NASDAQ), Bristol-Myers Squibb Co. (BMY-NYSE), Eli Lilly & Co. (LLY-NYSE), and Merck & Co., Inc. (MRK-NYSE). Many of these entities have performed and published important demonstrations and studies that were crucially enabled by PCT systems.

Figure 3  
A SELECTION OF KEY CUSTOMERS

• Amgen, Inc.	• U.S. Centers for Disease Control and Prevention	• University of California, Los Angeles
• Biogen Idec Inc.	• U.S. Army Medical Research Institute of Infectious Diseases	• University of California, San Diego
• Eli Lilly & Co.	• Dana-Farber Cancer Institute	• University of California, Davis
• Novartis AG	• Sanford-Burnham Medical Research Institute	• Lawrence Livermore National Laboratory
• Monsanto Company	• Thermo Fisher Scientific Inc.	• Stanford University
• Bristol-Myers Squibb Co.	• Target Discovery, Inc.	• University of North Texas
• Merck & Co., Inc.	• U.S. Department of Agriculture	• Mississippi State University
• Centocor Biotech, Inc. (now Janssen Biotech, Inc.)	• Battelle Memorial Institute	• Montana State University
• Takeda Pharmaceutical Company Ltd.	• Harvard Medical School	• Lawrence Berkeley National Laboratory
• Momenta Pharmaceuticals Inc.	• Harvard School of Public Health	• University of Kentucky
• U.S. Army	• Rockefeller University	• Florida International University
• U.S. Federal Bureau of Investigation	• University of New Hampshire	• University of Massachusetts Boston
• U.S. Food and Drug Administration	• Barnett Institute (Northeastern University)	
• U.S. National Institutes of Health		
• Armed Forces Institute of Pathology		

Source: Pressure BioSciences, Inc.

## **Ramping Up Commercialization Efforts**

Pressure BioSciences' chief focus since 2006 has been the continued development of its PCT platform technology and related consumable product line development. While the Company's products and consumables have been available for commercial sale outside of the collaboration program—primarily as a means to support and provide important feedback for continued R&D—Pressure BioSciences has not yet established a dedicated team to directly market and sell its products. Nevertheless, the Company's technology has experienced considerable early traction, with approximately 235 PCT systems already paid for and installed worldwide.

Pressure BioSciences is now gathering financial resources, and shifting focus from R&D to commercializing its product portfolio. The Company has expanded its distribution coverage to approximately 24 countries worldwide (as overviewed in Figure 4 [page 10]), and plans to continue expanding its network. Additionally, Pressure BioSciences plans to build a dedicated sales and marketing team as funding becomes available, which the Company expects can have a significant positive impact on PCT adoption and sales growth.

## **Continued Innovation Using PCT**

Pressure BioSciences strives to continually innovate and improve upon its existing technologies. In total, an estimated \$40 million has been invested in developing the PCT platform, including \$1.2 million in grant funding through the U.S. government's Small Business Innovation Research (SBIR) program as well as an \$850,000 contract from the U.S. Department of Defense (DOD) to develop a PCT-based system that provides for the simultaneous inactivation and processing of pathogenic organisms (specifically viruses and bacteria). To Pressure BioSciences' knowledge, PCT has been shown to be the only technology able to completely kill anthrax while still leaving the sample in a testable state.

In addition to the ongoing product development for the HUB Barocycler, the Company has three other instruments in its product pipeline. This includes development of the Barocycler HT Multiwell, which is designed to be a high-throughput PCT system capable of processing up to 96 (eventually up to 384) samples simultaneously in the industry-standard format for integration with existing lab automation platforms. In contrast, the existing Barocycler products provide capacity for one or up to a maximum run of 48 samples in a manual-handling format. In mid-2013, the Company announced that it achieved proof-of-principle for this initiative, which it identified as a major technical breakthrough and significant growth opportunity for its technology. While the current processing capabilities of its existing products have limited sales to small laboratories and individual scientists, an automation-ready, high-throughput system fulfills the needs of larger research and clinical diagnostics laboratories and companies. Pressure BioSciences believes that the Barocycler HT Multiwell has the potential to fuel growth and increase revenues for existing and new PCT-based applications and products, as well as to facilitate new strategic partnerships. The Company expects beta units to be built and tested by the first quarter 2014, with market-ready units to be available for sale as early as the second quarter 2014.

Additional products in development include specialized systems for protein extraction and digestion, as overviewed on pages 49-53.

## **Corporate Information**

Pressure BioSciences is a spin-off of Boston Biomedica Inc., which was founded in 1978. Richard T. Schumacher founded Boston Biomedica during his tenure as a researcher at the Center for Blood Research (CBR), a laboratory affiliated with Harvard Medical School. He left CBR in 1985 and spent the next two decades building Boston Biomedica to become a preeminent global quality control company for infectious disease testing. In 1996, the company went public on NASDAQ and grew to over 300 employees and 6,000 laboratories across 60 countries. In September 2004, this entity completed the sale of its core business units and began to focus exclusively on the development and commercialization of the PCT platform. Following this change in business strategy, the Company's legal name was changed from Boston Biomedica, Inc. to Pressure BioSciences, Inc. Pressure BioSciences is now traded on the OTCQB tier of the OTC Marketplace under the ticker symbol "PBIO."

The Company leases corporate offices in South Easton, Massachusetts. In addition to serving as Pressure BioSciences' corporate headquarters, these facilities support the manufacture and assembly of the Barocycler HUB440, the Shredder SG3, and MicroTubes. Pressure BioSciences also leases laboratory and office space at the University of Massachusetts in Boston, where it performs R&D activities. At present, the Company employs 10 full-time individuals.



## Growth Strategy

### Current Sources of Revenue and Funding

Pressure BioSciences' current sources of revenue stem from the sale or lease of its Barocycler and Shredder instruments, replacement of instrument parts, and from recurring purchases of consumables required for the PCT process, including single-use processing containers (PULSE Tubes and MicroTubes) and reagent kits. As well, the Company provides extended Barocycler service contracts (priced at a 10% premium to the purchase price of an instrument), which include preventative maintenance, repair or replacement of worn or defective parts, and phone support. Pressure BioSciences may also earn royalties through licensing agreements, such as with Target Discovery, Inc. (<http://www.targetdiscovery.com> [as described on pages 39-40]). Pressure BioSciences is a company of 10 full-time employees, one of whom is currently dedicated to field sales. The Company expects to significantly increase its focus on marketing, sales, and distribution in the near future.

Federal grants have further served as an important source of revenue for Pressure BioSciences. The Company pursues grants for planned R&D initiatives. To date, Pressure BioSciences' R&D has been supported by \$1.2 million in grant funding through the U.S. government's Small Business Innovation Research (SBIR) program. These comprise SBIR Phase I and II grants from the NIH, which support projects to establish the technical merit and feasibility of ideas that could lead to marketed products or services. Funding received from grants offset laboratory expenses incurred in developing applications for PCT, covering expenses that would typically have been paid by the Company. All existing grants are completed.

Additionally, in 2011, the Company was awarded an \$850,000 contract from the DOD to develop a PCT-based system to improve the processing of pathogenic organisms (specifically viruses and bacteria). The PCT platform has been shown to inactivate and extract live attenuated virus and bacteria that are used as models of important biological threat agents. To Pressure BioSciences' knowledge, PCT is the only technology that has been shown to be able to completely kill anthrax while still leaving the sample in a testable state. As well, the Company has rights to market and sell the final product platform in the non-military arena, intending to make this new system available for pharmaceutical, biotechnology, and academic users. The DOD contract funded studies through September 2013.

Key to Pressure BioSciences' future growth is the development of a sustainable fiscal road map through a combination of financings, continued reductions in costs, and measurable increases in revenue. As of June 30, 2013, Pressure BioSciences' cash position was \$113,000. The Company is pursuing an investor awareness strategy and has hired corporate advisory firm Merriman Capital, Inc. to assist in these efforts. From February 2012 to date, the Company raised roughly \$4.5 million in gross proceeds. Pressure BioSciences reports that it has completed eight raises in the last four years, seven of which were completed internally at or above market value.

### Commercialization Strategy

The Company's most significant strategy for growth is its aggressive product commercialization for PCT products. Key to this approach is bolstering sales of its core products and technologies through a focus on expanding distribution and establishing a dedicated sales and marketing team, as described below and continued on page 10.

**Global Distribution Network.** During 2012, Pressure BioSciences expanded its distribution coverage from 3 to 24 countries worldwide across Europe, Asia, and Australia. These agreements are summarized in Figure 4 (page 10). Most recently, in late 2012, the Company entered into distribution agreements with Constant Systems Ltd. and Cole-Parmer Corporation. Pressure BioSciences aims to add another major distribution partner during 2013 or early 2014.

Figure 4

A SELECTION OF PRESSURE BIOSCIENCES' DISTRIBUTION AGREEMENTS

Date	Company	Agreement Terms
June 2008	Veritas Corporation	Exclusive distribution rights to Pressure Biosciences' products in Japan through December 31, 2013
Oct. 2011	IUL Instruments GmbH	Exclusive distribution rights to Pressure BioSciences' products in Germany and Switzerland through March 31, 2014
Nov. 2011	Oroboros Instruments Corp.	Non-exclusive global distribution rights to the Shredder SG3 System and related products through December 31, 2013
Jan. 2012	Digilab, Inc.	Global co-marketing and co-selling agreement for each company's respective product lines
May 2012	LEAP Technologies	Strategic co-development, co-marketing, and co-selling agreement for each company's respective product lines
July 2012	Yinzhou Police Equipment & Technology Co., Ltd	Exclusive rights to distribute the PCT product line and non-exclusive distribution rights for the SG3 and related products within the Chinese forensic market
July 2012	BMN MSI., Ltd.	Exclusive rights to distribute the PCT product line and non-exclusive rights to distribute the Shredder SG3 and associated consumables in Vietnam, Cambodia, and Laos
July 2012	iScience Technology	Exclusive rights to distribute the PCT product line and non-exclusive rights to distribute the Shredder SG3 and associated consumables in Australia and New Zealand
Mar. 2012	LA Biosystems BV	Exclusive rights to market and sell PCT products and consumables as well as non-exclusive rights to market and sell the Shredder SG3 and associated consumables in Belgium, the Netherlands, and Luxembourg
Oct. 2012	Cole-Parmer Corporation	Non-exclusive, worldwide distribution rights to the PBI Shredder SG3 System and related consumables through December 2014
Nov. 2012	Constant Systems Ltd	Non-exclusive distribution rights to certain of Pressure BioSciences' PCT SPS product line in England, Scotland, Wales, Ireland, Spain, Portugal, Italy, Norway, Sweden, Finland, Denmark, and Singapore, through December 31, 2013

Source: Pressure BioSciences, Inc.'s Press Releases and Form 10-K dated May 15, 2013.

**Establishing a Dedicated Sales and Marketing Team.** Pressure BioSciences' domestic sales force currently consists of one full-time sales director and one part-time salesperson. To date, sales have been achieved largely without support of a significant sales and marketing effort, with a focus on trade shows and sponsorships, advertising, search engine optimization, creation and dissemination of a company newsletter, lectures, seminars, and demonstrations. Pressure BioSciences has plans to formally establish a sales team (contingent on obtaining adequate financial resources), which the Company believes could augment sales of its PCT products and significantly increase market penetration. Richard T. Schumacher, the Company's president and CEO, has years of experience and expertise in this process. Previously, while serving as founder and CEO of Boston Biomedica, Mr. Schumacher oversaw the growth of Boston Biomedica's marketing and sales team from three people and several million in revenue (early 1990s) to over 30 people and nearly \$30 million in revenue by 2000.

**Continued Product Development.** In addition to marketing and commercializing its existing products, Pressure BioSciences is also committed to continued innovation. The Company is developing three novel products to support future revenues in key markets: (1) a high-throughput system called the Barocycler HT Multiwell; (2) the Barocycler FFPE Protein Extraction Instrument System; and (3) the Company's first high-performance liquid chromatography (HPLC) system, called the XstreamPCT™ HPLC Digestion Module. These technologies could bolster Pressure BioSciences' product portfolio and are expected to result in increased revenues from both instrument and consumables sales.

## Milestones

Pressure BioSciences has achieved a number of significant milestones in the past 12 months, including those highlighted below.

### Continued Product Development

- Achieved proof-of-principle objectives with a new high-throughput design for its patented PCT platform, which the Company believes opens up opportunities for growth in existing and new PCT-based applications and products, new strategic partnerships, and increased revenue potential
- Announced that its PCT platform was featured in a breakthrough method developed by researchers at Harvard Medical School for analyzing lipids (fats) in fecal material, which was published in the peer-reviewed research journal *Analytical Chemistry*
- Announced that data presented at the 2013 American Academy of Forensic Sciences (AAFS) by Florida International University (FIU) researchers revealed further advances in methods using the Company's PCT platform to create a better DNA profile from rape kits
- Announced that data presented at the US HUPO 9<sup>th</sup> Annual Conference from three separate research groups—ETH Zurich and the University of Zurich; the FDA and NIH; and the Johns Hopkins University, Louisiana State University, and Wake Forest University Schools of Medicine—indicated that sample preparation methods based on the Company's PCT platform resulted in improved quality and/or efficiency of test results
- Announced that multiple presentations at international scientific conferences confirmed the advantages of PCT, with researchers reporting that the PCT significantly improved the detection of DNA and proteins in biomarker discovery, forensics, environmental, and biodefense studies

### Expanded Distribution

- Expanded sales reach to 24 countries across Europe, Asia, and Australia
- Announced a strategic co-marketing, co-selling, and distribution agreement with Constant Systems Ltd., which expanded distribution in 12 countries in Europe and Asia
- Added Cole-Parmer as a global distributor for Pressure BioSciences' Shredder SG3 sample preparation device

### Appointed New Leadership

- Appointed Dr. Mickey Urdea to develop and lead the Company's Scientific Advisory Board as well as to join Pressure BioSciences' Board of Directors (biography provided on page 16)

### Obtained Additional Funding to Support Ongoing R&D and Initial Marketing and Sales Initiatives

- Completed a two-tranche Series G private placement in November 2012 for \$726,598 in gross proceeds
- Closed over \$2 million in total gross proceeds during a multi-tranche private placement between February and June 2013

## Intellectual Property

Pressure BioSciences relies on a combination of intellectual property (IP) as well as trade secrets, know-how, and technological innovations to protect its competitive position in the market. The Company's IP portfolio includes 24 issued patents relating to its pressure cycling technology (PCT). These patents, listed in Figure 5, encompass multiple sample preparation applications. The expiration dates for these patents occur between 2015 and 2027. Additionally, Pressure BioSciences reports that it has six pending patents and additional patents in development.

Figure 5  
GLOBAL PATENT PORTFOLIO

Patent No.	Title	Region	Date Issued
US 6,036,923	Pressure cycling reactor and methods of controlling reactions using pressure	U.S.	03/14/2000
US 6,111,096	Nucleic acid isolation and purification	U.S.	08/29/2000
US 6,120,985	Pressure-enhanced extraction and purification	U.S.	09/19/2000
US 6,127,534	Pressure modulated ion activity	U.S.	10/03/2000
US 6,245,506	Integrated sequencing device	U.S.	06/12/2001
US 6,258,534	Pressure-controlled nucleic acid hybridization	U.S.	07/10/2001
US 6,270,723	Rapid cryobaric sterilization and vaccine preparation	U.S.	08/07/2001
US 6,274,762	Pressure-enhanced extraction and purification	U.S.	08/14/2001
US 6,448,065	Integrated sequencing device	U.S.	09/10/2002
US 6,569,672	Pressure cycling reactor	U.S.	05/27/2003
US 6,635,469	Pressure-mediated binding of biomolecular complexes	U.S.	10/21/2003
US 6,696,019	Rapid cryobaric sterilization and vaccine preparation	U.S.	02/24/2004
US 6,753,169	Pressure-controlled nucleic acid hybridization	U.S.	06/22/2004
US 7,626,017 B2	Pressure-enhanced extraction and purification	U.S.	12/01/2009
JP 3781780	Pressure-mediated binding of biomolecular complexes	Japan	03/17/2006
JP 4308320	Pressure cycling reactor and methods of controlling reactions using pressure	Japan	5/15/2009
EP 0814900	Pressure cycling reactor	Europe	9/12/2001
EP 0924991	Pressure-mediated binding of biomolecular complexes	Europe	10/4/2001
EP 1112091	Rapid cryobaric sterilization and vaccine preparation	Europe	9/17/2003
AU 745925	Pressure-enhanced extraction and purification	Australia	7/25/2002
AU 2002259017	Multichamber device and uses thereof for processing of biological samples	Australia	11/29/2007
AU 2002367749	Rapid sterilization and vaccine preparation	Australia	1/17/2008
CA 2259318	Pressure-mediated binding of biomolecular complexes	Canada	6/26/2007
CA 2301067	Rapid cryobaric sterilization and vaccine preparation	Canada	9/2/2008

Source: Pressure BioSciences, Inc.

## Key License Agreements

PCT is an important component of Pressure BioSciences' business. The Company's IP in this area spans several license agreements, as overviewed below.

- In 1998, Pressure BioSciences acquired BioSeq, Inc. and its related technology and agreements. BioSeq was a company developing PCT after acquiring the original technology from BioMolecular Assays, Inc.

*Terms:* Pressure BioSciences is required to pay BioMolecular Assays a 5% royalty on sales of the Company's products or services that use the original PCT as well as 5% of proceeds from any sale, transfer, or license of all or any portion of the original PCT. Both of these payment obligations end in 2016. In 2011 and 2012, Pressure BioSciences incurred roughly \$23,634 and \$21,090, respectively, in royalty expenses associated with its obligation to BioMolecular Assays. Pressure BioSciences also licensed certain limited rights to the original PCT back to BioMolecular Assays solely for molecular applications in scientific R&D and in scientific plant R&D in exchange for a 20% royalty for any license or other fees and royalties received in connection with the sale, assignment, license, or other transfer of any rights granted to BioMolecular Assays under the license until the patents expire (expected in 2016). To date, no royalty payments have been received from BioMolecular Assays.

- In 2008, Pressure BioSciences entered into an exclusive patent license agreement with the Battelle Memorial Institute for a method and system for improving the analysis of protein samples. The patent application, filed in 2008, encompasses an automated system using pressure and a pre-selected agent to obtain a digested sample in less time than current methods while maintaining the sample's integrity.

*Terms:* Pressure BioSciences owes royalty payments on net sales of "licensed products" and is obligated to make minimum royalty payments for each year that it retains the rights (\$7,500 and \$10,000 in 2011 and 2012, respectively). The Company is required to have its first commercial sale of the licensed products within one year after the related patent is issued.

## Company Leadership

Pressure BioSciences' leadership is composed of individuals with broad experience in the life science industry. Its chief executive officer (CEO), Richard T. Schumacher, intimately understands the business and market, having been with the Company since he founded its legacy business, Boston Biomedica, Inc., in 1978. He is supported by individuals with considerable R&D experience, including with high-pressure technologies as well as **proteomic**, **genomic**, and biochemical analysis.

### Management

Figure 6 summarizes the Company's executive leadership, followed by brief biographies.

Figure 6  
MANAGEMENT

Richard T. Schumacher	President, Chief Executive Officer, and Director
Edmund Y. Ting, Sc.D.	Senior Vice President of Engineering
Nathan P. Lawrence, Ph.D.	Vice President of Sales, Marketing, and Business Development
Alexander Lazarev, Ph.D.	Vice President of Research and Development
Richard P. Thomley, CPA	Acting Chief Financial Officer

*Source: Pressure BioSciences, Inc.*

#### *Richard T. Schumacher, President, Chief Executive Officer, and Director*

Mr. Schumacher, the founder of Pressure BioSciences, has served as a director of the Company since the formation of its legacy business, Boston Biomedica, Inc., in 1978. He has served as CEO of Pressure BioSciences since April 2004 and as president since September 2004. Mr. Schumacher previously served as CEO and chairman of the Board of Boston Biomedica from 1992 to February 2003 and as president from 1986 to August 1999. He served as the Director of Infectious Disease Services for Clinical Sciences Laboratory, a New England-based medical reference laboratory, from 1986 to 1988. From 1972 to 1985, Mr. Schumacher was employed by the Center for Blood Research, a non-profit medical research institute associated with Harvard Medical School. He received a B.S. in zoology from the University of New Hampshire.

#### *Edmund Y. Ting, Sc.D., Senior Vice President of Engineering*

Dr. Ting joined Pressure BioSciences as senior vice president of engineering in April 2006. Prior to joining the Company, Dr. Ting served as the chief research officer of Avure Technologies, a worldwide manufacturer of high-pressure hydrostatic processing equipment for the food and materials processing industry, where he worked from 2001 to 2006. From 1990 to 2001, Dr. Ting was employed by Flow International Corporation, a world leader in the ultra-high-pressure waterjet cutting technology market and the parent company of Avure Technologies, until November 2005. Dr. Ting's last position at Flow International was vice president of engineering research and development. He was also a research scientist and a group leader at Grumman Aerospace Corporation. Dr. Ting received a B.S. in mechanical engineering from Northeastern University and an Sc.D. in materials science and engineering from Massachusetts Institute of Technology.

*Nathan P. Lawrence, Ph.D., Vice President of Sales, Marketing, and Business Development*

Dr. Lawrence joined Pressure BioSciences in August 2005 as director of research and development until his promotion to vice president of sales, marketing, and business development in April 2006. Dr. Lawrence was responsible for the development of protocols based on PCT. From 2004 through 2005, Dr. Lawrence worked for 454 Life Sciences, a subsidiary of Roche Diagnostics Corporation that specializes in high-throughput DNA sequencing, in product development. Prior to 454 Life Sciences, Dr. Lawrence was director of R&D for Boston Biomedica. He was responsible for the development of PCT, as well as the development of nucleic acid-based diagnostic assays. Prior to joining Boston Biomedica, Dr. Lawrence held several positions with increasing responsibility in research and development and manufacturing at Becton, Dickinson and Company (BDX-NYSE) and Gene-Trak Systems. Dr. Lawrence holds a B.A. from the University of Miami, an M.S. from Southern Connecticut State University, and a Ph.D. from Yale University.

*Alexander Lazarev, Ph.D., Vice President of Research and Development*

Dr. Lazarev joined Pressure BioSciences in April 2006 as director of R&D and was promoted to vice president of R&D in March 2007. Prior to joining Pressure BioSciences, Dr. Lazarev worked as a visiting scientist at the Barnett Institute of Chemical and Biological Analysis at Northeastern University, and served as a director of new technology development at Proteome Systems, Inc., where he was involved in R&D of innovative proteomic analysis applications. Dr. Lazarev previously held senior research positions at Genomic Solutions, Inc. and PhytoChem Technologies, Inc. Most of Dr. Lazarev's scientific career has been dedicated to development of methods and applications for biochemical analysis. He has been elected as an executive board member of the MASSEP.org, a nonprofit scientific discussion forum dedicated to the promotion and improvement of chromatography and other analytical technologies. Dr. Lazarev earned undergraduate and graduate degrees at the University of Kazan (Russian Federation).

*Richard P. Thomley, CPA, Acting Chief Financial Officer*

On September 27, 2013, the Company's Board of Directors appointed Mr. Thomley as acting chief financial officer (CFO). Mr. Thomley has been an independent financial consultant to the Company since March 2013, during which time he has been involved in all areas of financial responsibilities, including SEC reporting. Before that, Mr. Thomley was the director of finance of Kiva Systems, Inc. from June 2007 until January 2012. Mr. Thomley was the chief accounting officer at Spire Corp. from January to June 2007. He served as the corporate controller for Implant Sciences, Inc. from November 2004 until January 2007. Mr. Thomley was CFO at SynQor, Inc. from July 2000 to November 2004. He has also held senior management positions in finance at Genesis Technical and Financial, Catamount Manufacturing, Inc., ChemDesign Corp., and Ansul Fire Protection Co. He has over 30 years of experience in U.S. GAAP accounting, internal management, financial reporting and analysis, and SEC reporting. Mr. Thomley is a Certified Public Accountant (CPA).

## Board of Directors

In the past year, Pressure BioSciences has focused on strengthening its Board of Directors, which oversees the conduct of and supervises the Company's management. Pressure BioSciences' Board of Directors includes seasoned Wall Street veterans as well as experienced executives in the life sciences field. Figure 7 provides a summary of Board members, followed by detailed biographies.

Figure 7  
BOARD OF DIRECTORS

Jeffrey N. Peterson	Chairman of the Board of Directors
Michael "Mickey" S. Urdea, Ph.D.	Director
Vito Mangiardi, MBA	Director
Kevin Pollack, Esq., MBA	Director
Richard T. Schumacher	President, Chief Executive Officer, and Director

Source: Pressure BioSciences, Inc.

### *Jeffrey N. Peterson, Chairman of the Board of Directors*

Mr. Peterson is the chairman of the Board of Directors. He is also the CEO of Target Discovery, Inc., a personalized medicine diagnostics company. Mr. Peterson also serves as chairman of Target Discovery's majority-owned subsidiary company, Veritomyx, Inc., which develops tools in accurate peptide, protein, isoform, and metabolite identification and characterization. Mr. Peterson brings broad executive general management, multi-functional, multi-business, and international experience to these roles. Prior to Target Discovery, he served as CEO of Sharpe, Peterson, Ocheltree & Associates, an international business development consulting firm assisting Fortune 500 and many smaller firms in business expansion and strategy. He spent nine years in key management roles in Abbott Laboratories' (ABT-NYSE) Diagnostics and International (pharmaceuticals, hospital products, nutritionals, consumer) businesses, last serving as CEO and General Manager of Abbott South Africa, where he doubled the sales and tripled the income of this 50 year-old business in 3.5 years during the tumult of South Africa's political transition. He played an earlier pivotal management role in Abbott's successful introduction and support of multiple new diagnostics instrument and reagent systems in the X-System series, including the IMx (the highest global sales diagnostic system at that time). Mr. Peterson's experience prior to Abbott included 11 years with General Electric Company's (GE-NYSE) Engineered Materials and Plastics businesses, spanning roles in strategic planning, business development, technology licensing, marketing/sales, operations/quality, and R&D. He holds B.S. and M.S. degrees in chemical engineering from MIT, as well as six issued U.S. patents. Mr. Peterson is chair emeritus of the BayBio Institute, a nonprofit organization serving the regional life science community, and serves on the Board of BayBio, the trade association for the life sciences industry in Northern California. He is a cofounder of the Coalition for 21<sup>st</sup> Century Medicine, and of BIO's Personalized Medicine and Diagnostics (PMDx) Working Group.

### *Michael "Mickey" S. Urdea, Ph.D., Director*

Dr. Urdea has devoted his 30-year career to human diagnostics in a variety of capacities. He has been involved in the discovery of new biomarkers, the development of new technologies for biomarker discovery, validation and commercialization, diagnostic test development, manufacturing and marketing, and the management of companies involved in these activities. Dr. Urdea founded and is a managing partner for Halteres Associates, LLC, a biotechnology consulting firm. He also founded and served as CEO of Tethys Bioscience, Inc., a proteomics-based diagnostics company involved in preventative personalized medicine. Tethys' first product was the protein-based PreDx Diabetes Risk Score Test, which was introduced in 2008. Additionally, Dr. Urdea is a founder and chairman of Catalysis Foundation for Health, an organization addressing gaps in global healthcare caused by inefficiencies in disease diagnosis and monitoring. He serves as an expert consultant to the life sciences industry and is on the Scientific Advisory Boards and Boards of Directors of a number of biotechnology, diagnostics, venture capital, and philanthropic organizations. Prior to his current business activities, Dr. Urdea founded the Nucleic Acid Diagnostics group at Chiron Corporation, and with colleagues, invented branched DNA molecules for amplification of signal in



nucleic acid complexes. Application of this technology resulted in the first commercial products for quantification of human hepatitis B, hepatitis C, and human immunodeficiency viruses (HBV, HCV, and HIV, respectively). He then became business head of the Molecular Diagnostics group and chief scientific officer at Bayer Diagnostics. He was also a member of the Bill and Melinda Gates Foundation Diagnostic Forum. Dr. Urdea is an author on nearly 200 peer-reviewed scientific publications, nearly 300 abstracts and international scientific presentations, and more than 100 issued and pending patents. He received a B.S. in biology and chemistry from Northern Arizona University and a Ph.D. in biochemistry from Washington State University.

*Vito Mangiardi, MBA, Director*

Mr. Mangiardi is a strong P&L performer and corporate strategist in general management, operations, sales/marketing, and science. He has held positions as a research chemist for Bio-Rad Laboratories, Inc.; sales and marketing director for Baxter Travenol, Inc.; executive vice president and chief operating officer (COO) for Quintiles Transnational Holdings Inc. (Q-NYSE); president and CEO of Diagnostics Laboratories, Inc., Clingenix, Inc., and Bilcare, Inc.; and president of AAI Pharma, Inc. More recently, he was the COO and deputy director of operations and production at the University of California's Lawrence Berkeley National Laboratory, Joint Genome Institute. Mr. Mangiardi has experience with three startups, two midsize, and several mature companies, and has international experience leading and managing organizations on four continents. He has experience in alliances, acquisitions, due diligence, and post-acquisition assimilation. Mr. Mangiardi has been on the Board of Directors of three companies and has proven success in working with both national and international investment groups to raise funds. Mr. Mangiardi received a B.S. in biology/chemistry from Eastern Illinois University and two MBA degrees from Golden Gate University in general management and in marketing. Mr. Mangiardi is an inventor on four patents and is a member of numerous professional organizations.

*Kevin Pollack, Esq., MBA, Director*

Mr. Pollack is chief financial officer of Lightlake Therapeutics Inc. (LLTP-OTC), a developing biopharmaceutical company aiming to build a platform of biopharmaceutical solutions to common addictions and related disorders. He also serves as president of Short Hills Capital LLC, where he provides a range of advisory services to investors, asset management firms, institutions, and companies. Previously, Mr. Pollack worked in asset management at Paragon Capital LP, a private investment firm focused primarily on investing in U.S.-listed companies. Prior to that, he worked as an investment banker at Banc of America Securities LLC (an investment banking subsidiary of Bank of America Corporation until it was merged with Merrill Lynch), focusing on both corporate finance and mergers and acquisitions (M&A). Mr. Pollack started his career at Sidley Austin LLP (formerly Brown & Wood LLP) as a securities attorney focusing on corporate finance and M&A. He currently sits on the Board of Directors of Lightlake Therapeutics Inc. and on the Board of Directors of MagneGas Corporation (MNGA-NASDAQ), the developer of a technology that converts liquid waste into a hydrogen-based metal working fuel and natural gas alternative. Mr. Pollack graduated magna cum laude from the Wharton School of the University of Pennsylvania and received a dual J.D./M.B.A. from Vanderbilt University, where he graduated with Beta Gamma Sigma honors.

*Richard T. Schumacher, President, Chief Executive Officer, and Director*

Biography provided on page 14.

## Core Story

Pressure BioSciences, Inc. (“Pressure BioSciences” or “the Company”) is focused on the research, development, and commercialization of its patented pressure cycling technology (PCT). PCT is an enabling technology that is able to create, apply, and release high levels of hydrostatic pressure—currently up to 58,000 pounds per square inch (psi)—quickly and in a safe and controlled manner. The technology has been featured in approximately 100 publications and posters by researchers at recognized medical and research institutions and U.S. government agencies, including UCLA, Brigham and Women’s Hospital, Harvard Medical School, Johns Hopkins University, Georgetown University, the FDA, FBI, NIH, and the U.S. Army Medical Research Institute of Infectious Diseases. The Company has 24 issued patents and six pending patents globally, which cover multiple applications of PCT.

Pressure BioSciences is initially developing the PCT platform as a tool for preparing samples in life science research, which includes pharmacology, biomarker discovery and diagnostics, cancer studies, physiology, microbiology, genomics, bioinformatics, biotechnology, and stem cell research, among other areas. Sample preparation, the process of preparing a sample for scientific analysis, is a crucial laboratory step for life sciences research. It can include breaking up and separating the components of the cell. Important cell components can be lost, degraded, damaged, or otherwise adversely affected during the sample preparation process. For this reason, sample preparation using existing techniques is considered to be a bottleneck in life science research. Given the breadth of the life sciences market, the Company has elected to focus initially on sample preparation in biomarker discovery and forensic applications.

Pressure BioSciences has harnessed the PCT platform to develop a line of laboratory instruments under the Barocycler® brand. In a laboratory setting, scientists can use a Barocycler instrument to quickly break up the cellular structures of a specimen in an effort to release nucleic acids, proteins, lipids, and small molecules for further testing and analysis. Some of these may not be able to be liberated and produced for analysis by any other technology. The Company’s current product models, which are designed to fit on a laboratory workbench, are capable of producing up to 58,000 psi. To date, Pressure BioSciences has installed approximately 235 Barocycler instruments worldwide (see Figure 3 [page 6] for a selection of marquee users).

It is important to note that PCT is applicable to a variety of industries with hundreds of relevant applications beyond the life science research industry. Pressure BioSciences reports that its products have been used by a Detroit automaker to evaluate compression in an engine as well as by Mississippi State University to study bacteria encountered during oil drilling activities, in an attempt to help the oil and gas industry determine oil quality at earlier stages. A selection of relevant applications for PCT is presented in Figure 23 (page 35).

Pressure BioSciences’ research and development (R&D) to date has been supported by approximately \$1.2 million in grant funding through the U.S. government’s Small Business Innovation Research (SBIR) program. Additionally, in 2011, the Company received an \$850,000 contract from the U.S. Department of Defense (DOD) to develop a PCT-based system that improves the inactivation and simultaneous processing of pathogenic organisms (specifically of viruses and bacteria), such as anthrax.

## Sample Preparation Overview

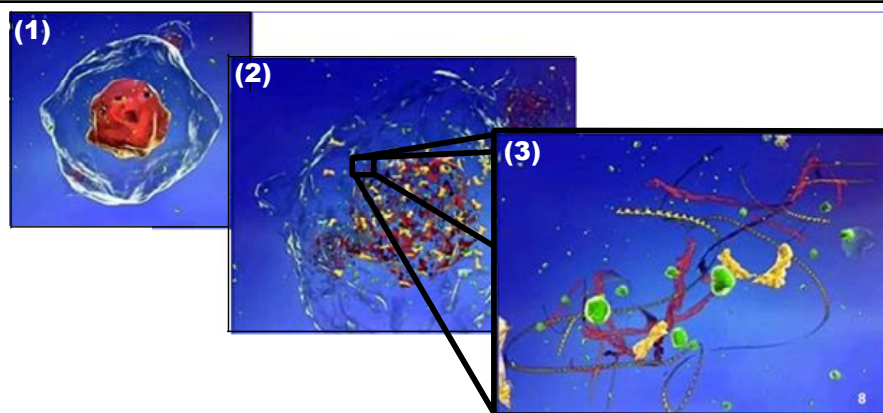
Scientific research has advanced from cellular biology (studying the structure and composition of whole cells) to molecular biology—studying the structure and function of the macromolecules found within cells (e.g., proteins and nucleic acids). Genomics (the study of genes and their function) and proteomics (the study of proteins) are fields of molecular biology. Preparing a sample for scientific analysis, or “sample preparation,” is an important laboratory step for genomic, proteomic, lipidomic, and small molecule studies as well as other areas of life science research. There are two key goals of sample preparation: (1) to remove any interference that may affect the results of the scientific analysis; and (2) to increase abundance of targeted **analytes**, thus improving detectability and ultimately the utility of the scientific analysis.

Sample preparation can encompass a wide range of activities, from sampling and collecting to **fractionating**, isolating, extracting, and concentrating. Cell lysis—the act of breaking down a cell—is the first laboratory step in this process. A range of tools and techniques are used to break up cells, including bead beaters, sonicators, homogenizers, mortar and pestle grinders, French Press, freezer milling, enzymatic digestion, and chemical dissolution. The process of sample extraction entails separating a substance—such as nucleic acids (DNA, RNA), proteins, and small molecules—from the plant or animal cells and tissues that are being studied. Sample extraction is generally regarded as the key part of sample preparation.

Figure 8 provides an illustration of cell lysis. On the left side in Box 1 is a representation of a whole cell, which can be human, animal, plant, microbial, etc. The cell is composed of thousands of different proteins and contains RNA, DNA, lipids, and small molecules. The outside layer is a membrane called the lipid bilayer, which is composed of opposed layers of lipids containing many intertwined membrane proteins. In order to study the individual components of the cell membrane, it has to be broken up (“lysed”) into smaller pieces and separated. The middle image (“2”) in Figure 8 shows the representative cell being broken up. The right side (“3”) of Figure 8 is a magnification of a small section of the broken up cell. Once lysed, researchers can study individual components of the cell, such as specific membrane proteins, and further, individual components of proteins and nucleic acids.

Figure 8

IMPORTANT BIOMOLECULES ARE RELEASED BY CELL LYSIS



Source: Pressure BioSciences, Inc.

Studying proteomics, genomics, and lipidomics within cells can help answer important questions about diagnosis, treatment, and prognosis of many forms of disease and disorders. As an example, differences in membrane proteins from prostate cancer cells may help physicians identify whether aggressive intervention such as a radical prostatectomy is clearly indicated versus whether the patient would have the greatest benefit from avoiding invasive procedures and simply continuing to monitor this typically indolent cancer in a “watchful waiting” mode.

Currently, cancer patients may try a therapy for months before an assessment is completed to ascertain whether the therapy is beneficial. In contrast, tests could be developed to detect for specific proteins or mutations that help select the best therapy for individuals and their specific cancer types, and which monitor treatment response and prognostic outlook. This latter process is called “personalized medicine,” and is often heralded as the treatment paradigm of the future.

#### *Limitations of Current Sample Preparation Techniques*

While technology has vastly improved for analyzing samples, technology for preparing samples prior to their analysis has lagged behind. Sample preparation can be both a complex and time-consuming process. The University of Florida has identified human error as a major concern in sample preparation. Additionally, it is estimated that laboratory professionals spend as much as 80% of their time preparing samples for analysis (Source: *Lab Manager Magazine*, July 13, 2011). As a result, sample preparation remains the main bottleneck for discoveries in genomic and proteomic research (Source: BCC Research, *Sample Preparation in Genomics, Proteomics, and Epigenomics: Global Markets*, September 2011). Important cell components may be lost, degraded, damaged, or otherwise adversely affected during the process of breaking up and separating the components of the cell, hindering a scientist’s ability to obtain relevant and meaningful results. If the cellular substructures are not completely broken up, important proteins are inadvertently removed from the sample or otherwise obscured from analysis. If cellular structures are too aggressively broken up, important proteins are reduced to nonfunctional fragments of little or no value for subsequent analysis. Regardless of the advanced technology used to study the sample after it is prepared, important cell components lost or obscured through the sample preparation process will not be found in the analysis of the sample.

Methods that can be used to decrease sample preparation errors include using automated sample processing devices (to avoid human error), increasing the homogeneity of the raw material under investigation, and decreasing the total elapsed time between sample collection/arrival to when it is evaluated. PCT offers critical improvements and risk mitigation in all three of these dimensions. The impending extension of the PCT platform into the Barocycler HT Multiwell instrument is expected to deliver further improvements and risk mitigation by integrating smoothly with automated sample handling and pipetting systems that are already standard equipment in labs for high-throughput processing and minimization of human error through manual handling and pipetting steps.

#### *Market Opportunity*

The sample preparation market encompasses products, technologies, and services used to prepare samples for genomics, epigenomics, lipidomics, and proteomics. These products and services are used by academic and government research institutions, biotechnology and pharmaceutical companies, diagnostics manufacturers and clinical laboratories, and other public and private laboratories. The Company estimates that there are over 500,000 researchers and over 80,000 research laboratories working with biological samples worldwide.

The sample preparation market was valued at approximately \$4.6 billion in 2012, and is forecast to reach \$5.4 billion in 2013. The market is expanding rapidly, with potential to more than double to \$11.5 billion in 2018 (Source: BCC Research, *Sample Preparation in Genomics, Proteomics, and Epigenomics: Global Markets*, September 2013). Growth drivers for this market include increasing R&D investments for drug discovery and new drug development, growing demand for genetically modified crops and organisms, and the emergence of new technologies (Source: Transparency Market Research). The current transformation of healthcare and medical practice from a standardized “one size fits all” model into the new “personalized medicine” paradigm is being driven by a revolution in biomarker discoveries converted into diagnostic tools to inform these treatment guidance decisions and provides a perfect example of such a major emerging technology and market shift.

Pressure BioSciences believes that its Barocycler instruments and PCT consumable products fill an important and growing need in the sample preparation market. The Company has identified several additional benefits of commercializing PCT technology for the sample preparation market, as listed on page 21:

- the sample preparation market has a large unsatisfied need for better technology solutions;
- it is composed mostly of research laboratories, which are subject to minimal governmental regulation;
- it offers many applications with low technical risk for rapid development of the Company's products;
- it is compatible with Pressure BioSciences' technical core competency; and
- the Company has a strong patent and proprietary knowledge position in this area.

## Product Portfolio

### PRESSURE CYCLING TECHNOLOGY (PCT): HOW IT WORKS

Pressure BioSciences has developed and patented a novel, enabling technology platform, called pressure cycling technology (PCT). PCT or “barocycling” entails rapidly cycling pressure between standard atmospheric pressure (or ambient pressure) to high and ultra-high levels of hydrostatic pressure. The physical manipulations and state changes that PCT delivers are enabling applications that were previously unattainable. One example is the use of PCT to break up biological samples (through cell lysis) from human, animal, plant, and microbial sources prior to analysis. In this application, PCT has been shown to release higher quality, greater quantities, and (most importantly) previously unobserved species of biomolecules from samples—including nucleic acids, small molecules, lipids, and proteins—versus the many available alternative technologies.

The core of the Company’s PCT platform is the internally developed pressure intensifying systems—called the Ting Intensifier—that is able to increase pressure at a 440:1 ratio, and the novel sample containment systems. For example, when connected to a standard 85 psi air compressor—which is widely used by scientific laboratories, in dental offices, and by consumers—the intensifier can amplify the pressure to roughly 37,400 psi. The current intensifier, which is electronically controlled and has a maximum outlet pressure of 4 kbar (~58,000 psi), was designed and built by Dr. Edmund Ting, who currently serves as senior vice president of engineering at Pressure BioSciences and Dr. Alexander Lazarev, who serves as vice president of research and development at Pressure BioSciences (biographies on pages 14-15, respectively). Dr. Ting and Dr. Lazarev are recognized worldwide as high-pressure equipment and processing experts.

Pressure BioSciences’ PCT platform has shown to have the qualities outlined below.

- (1) *Fast*. PCT is able to release nucleic acids, small molecules, and proteins from a wide variety of cells and tissues within minutes, providing ~100X reduction in time versus current methods.
- (2) *Efficient*. Current PCT models can extract up to 48 samples simultaneously.
- (3) *Versatile*. PCT can process animal, plant, microbial, and non-biological samples. As well, it allows the researcher to define the specific parameters of each test run (e.g., the level of pressure, the number of cycles, detailed time/pressure profiles).
- (4) *Reproducible*. PCT is able to achieve consistent sample extraction, with a level of reproducibility that exceeds other sample preparation methods.

Pressure BioSciences believes that the versatility, speed, throughput, and reproducible control and output of its PCT system far exceeds other current extraction methods, including bead beaters, sonicators, homogenizers, and mortar and pestle grinders.

The Company has harnessed PCT to develop a line of laboratory instruments, called Barocyclers, which are capable of cycling pressure at controlled temperatures and in specific timed intervals. Currently, Pressure BioSciences has several Barocycler models available for sale, as overviewed on pages 25-28. The pressure chamber of a Barocycler instrument is composed of stainless steel (roughly a half-inch thick) and filled with water. The Company’s Barocycler machines pressurize the water to produce hydrostatic pressure. Water is used due to its low compressibility. At 35,000 psi, water compresses by just 8% (Source: [www.thefabricator.com](http://www.thefabricator.com), a production of the Fabrication and Manufacturers’ Association, International). The Company reports that, because its technology delivers pressure through nearly incompressible water, it believes that any malfunctions avoid the risk of explosive failures as the pressurized water would simply escape from a safety port out of the back of the machine.

Barocycler instruments can currently deliver up to 58,000 psi of hydrostatic pressure; with the Company anticipating being able to release equipment that can deliver up to 100,000 psi by the first quarter 2014. As a frame of reference, in the deepest part of the Mariana Trench—approximately 35,800 feet under the ocean’s surface and the deepest point on Earth—the pressure is about 17,920 psi or nearly nine tons per square inch (Source: NASA). The National Oceanic and Atmospheric Administration (NOAA) likens this to the weight of an elephant balanced on a postage stamp. Pressure BioSciences reports that its technology can triple that pressure within seconds, very safely and under careful control.

Figure 9 overviews Pressure BioSciences’ current product offerings, followed by summaries of each product family. In addition to instruments, the Company has a consumables product line, which includes PULSE™ (Pressure Used to Lyse Samples for Extraction) Tubes, MicroTubes, reagents, and specialized kits. Samples to be processed in a Barocycler are first placed in PULSE™ Tubes or MicroTubes. These consumable products are designed for single use, and act to minimize the risk of cross contamination between samples as well as to reduce the user’s risk of exposure to pathogens/toxins. Pressure BioSciences’ consumable products are further detailed on pages 31-33.

Figure 9  
PRODUCT OFFERINGS

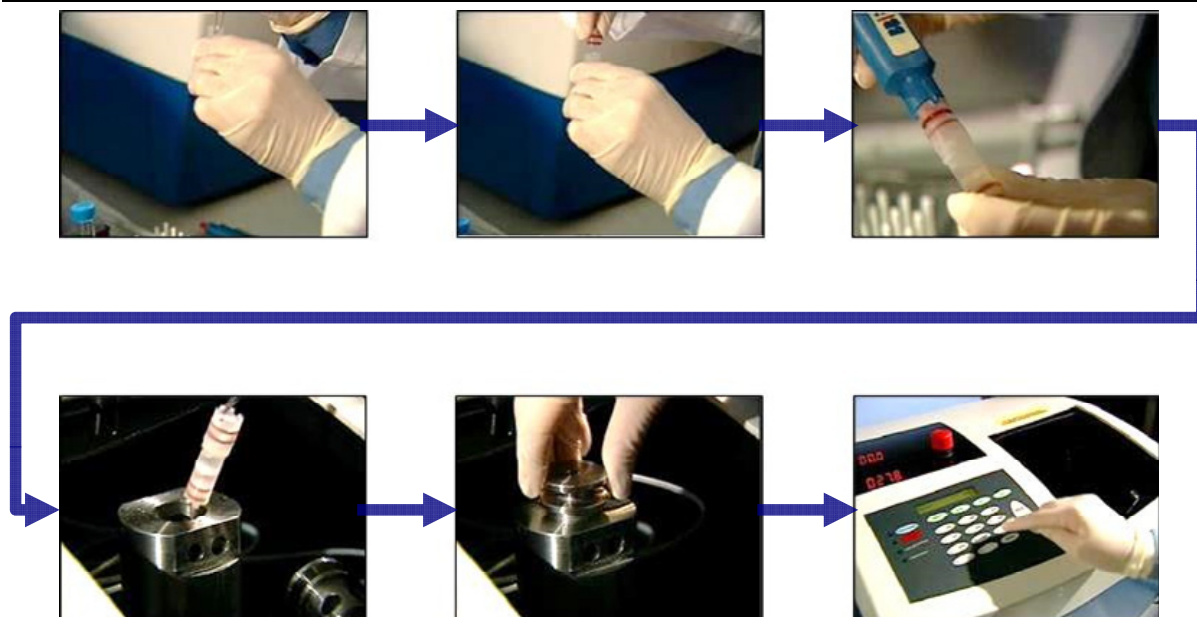
Barocycler Instruments	PULSE Tubes	Kits and Reagents
<ul style="list-style-type: none"> <li>○ Barocycler NEP3229</li> <li>○ Barocycler NEP2320</li> <li>○ Barocycler HUB440</li> </ul>	<ul style="list-style-type: none"> <li>○ FT500 PULSE Tubes</li> <li>○ FT500-ND PULSE Tube</li> </ul>	<ul style="list-style-type: none"> <li>○ Mitochondria Isolation Kit: Rat Muscle</li> <li>○ Mitochondria Isolation Kit: Rat Lung</li> <li>○ ProteoSolve-TD1 and TD2 Kits</li> <li>○ ProteoSolve-CE Native Kit</li> <li>○ ProteoSolve-CE Stringent Kit</li> <li>○ ProteoSolve-SB Kit</li> <li>○ ProteoSolve-IEF Reagent</li> </ul>
Shredders	PCT MicroTube Adapter Kit	
<ul style="list-style-type: none"> <li>○ SHREDDER SG3</li> <li>○ Shredder PULSE Tubes</li> </ul>	<ul style="list-style-type: none"> <li>○ MicroTubes</li> <li>○ MicroCaps</li> </ul>	

*Source: Pressure BioSciences, Inc.*

### PCT Sample Preparation Process

The combination of Pressure BioSciences’ consumable products and the Barocycler instruments comprise what Pressure BioSciences calls its PCT Sample Preparation System (the “PCT System”). To prepare a sample in the PCT System, the sample is placed into the sample chamber of a PULSE Tube. The sample can be cells or tissues, from a bodily fluid to a tumor biopsy to an entire tick or mosquito. Then, a movable piece called a “ram” is assembled inside the tube and a processing buffer is added to the PULSE Tube’s fluid retention chamber. The tube is closed before being placed into the reaction chamber of the Barocycler, which is filled with water. The reaction chamber is then sealed with a cap that locks into place and a safety door is closed. The user sets the desired pressure level, time required at high and ambient pressures, and number of pressure cycles—or alternatively selects a custom programmed protocol—and then starts the machine, as illustrated in Figure 10 (page 24).

Figure 10  
PREPARATION FOR THE PCT PROCESS

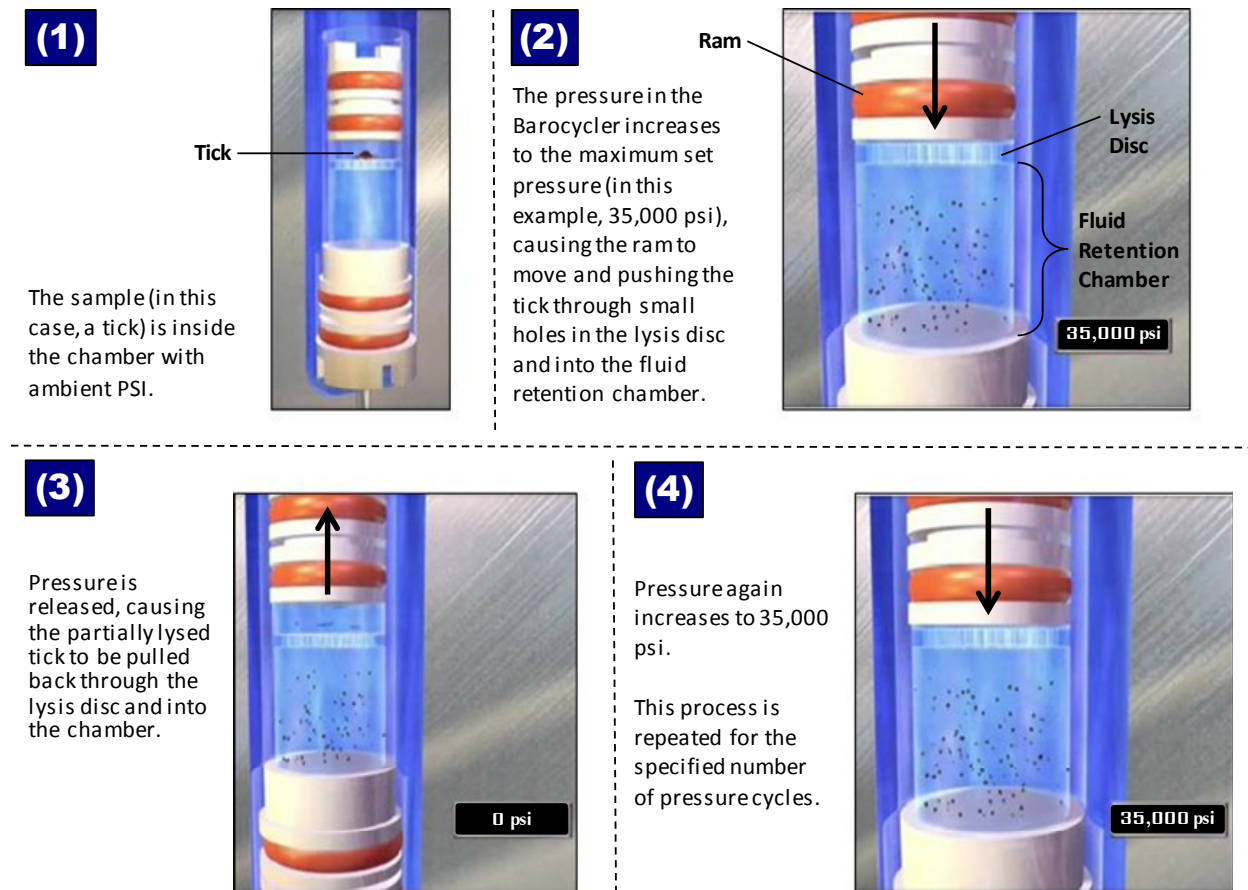


Sources: Pressure BioSciences, Inc. and Crystal Research Associates, LLC.

As illustrated in Figure 11 (page 25), the Barocycler increases the pressure in the chamber from ambient to the desired maximum pressure. The increasing pressure causes the ram to move, pushing the sample material from the sample chamber through small holes in the lysis disc and into the fluid retention chamber of the PULSE Tube, where it rapidly mixes with the added processing buffers. When pressure is released, the partially lysed sample is pulled back through the lysis disc and into the chamber. This process is repeated for the specified number of pressure cycles. The combination of physical passage through the lysis disc, rapid pressure changes, and other biophysical mechanisms that occur during the PCT process causes the cells and tissues of the sample to break up, releasing nucleic acids, proteins, lipids, and small molecules. The entire PCT process typically lasts several minutes. When the PCT process is finished, the PULSE Tube can be taken out of the Barocycler reaction chamber. The user then removes the **supernatant** from the PULSE Tube, which contains the extracted biomolecules that can then be used for subsequent testing.



Figure 11  
ILLUSTRATION OF THE PCT PROCESS



Note: In (1) and (3), the ambient pressure is 15 PSI = 0 PSIG.

Sources: Pressure BioSciences, Inc. and Crystal Research Associates, LLC.

### Barocycler Instruments

The Barocycler instrument is designed to fit conveniently on a laboratory bench top, or inside a biological safety cabinet, or on the shelf of a laboratory cold room. Additionally, each instrument is equipped with an external chiller hook-up (to control temperature during the PCT process) as well as automatic fill and dispensing valves. The machines also have an integrated micro-processor keypad that enables the user to program up to 99 custom protocols. The user can set the desired pressure, number of cycles, cycle profile, add specific chemistry reagents, and set specific temperatures for each protocol.

An additional advantage of Pressure BioSciences' Barocycler products is the ability to regulate temperature. Other homogenization procedures, such as beat beaters that rely on collisional friction to break up cells, generate large amounts of heat throughout the process. The heat accumulation must be managed by alternating the samples between the homogenization operation and ice baths. In contrast, a Barocycler product can control the temperature continuously throughout the process as there is a heating/cooling jacket around the pressure chamber. Pressure BioSciences reports that its technology can maintain sample temperature within a tight range of a few degrees.

The Company's NEP2320 and NEP3229 Barocycler products are manufactured and assembled by Source Scientific, LLC ([www.sourcescientific.com](http://www.sourcescientific.com)), a BIT Group company that develops and manufactures medical instruments.

*Barocyler NEP3229*

Pressure BioSciences' original Barocyler model, the NEP3229, is capable of producing up to a working maximum pressure of 35,000 psi. Pressure BioSciences believes that the Barocyler NEP3229 fills an important and growing need in the genomics, transcriptomics, lipidomics, and proteomics sample preparation market for a small, more affordable instrument that provides safety, quality, versatility, and reproducibility. It can process up to three samples simultaneously using PULSE Tubes, or up to 48 samples concurrently when using the Company's MicroTubes. The NEP3229 model contains two key components: (1) the primary machine that contains the stainless steel pressure chamber, a microprocessor, and an easy-to-use keypad; and (2) the 1.5 horsepower hydraulic motor and pump assembly, which serves as the machine's pressure source. While the primary machine can fit on a bench top, the pump and motor is designed to be placed on the floor. In total, the NEP3229 model weighs roughly 350 pounds. It has a list price of approximately \$45,000.

Figure 12  
BAROCYCLER NEP3229



Main unit  
with  
pressure  
chamber  
and user  
interface

Pump and  
motor

Source: Pressure BioSciences, Inc.

*Barocyler NEP2320*

Figure 13  
BAROCYCLER NEP2320



Source: Pressure BioSciences, Inc.

The Barocyler NEP2320 is a smaller, more portable version of the NEP3229 unit, weighing roughly 65 lbs. (80 lbs. with accessories). The NEP2320 model was originally designed to serve as a demonstration instrument due to its compact size. However, the Company reports that the model has been positively received by customers, and represents as much as 85% of Barocyclers sold. Unlike the larger NEP3229 model, the NEP2320 is pneumatic (versus hydraulic) in that it creates pressure cycles using compressed air. The NEP2320 unit can be attached to a standard 85 psi air compressor, which is widely used by scientific laboratories, in dental offices, and by consumers, or can even be connected to bottled gas. This Barocyler configuration routinely produces up to 35,000 psi, and with software and some hardware changes, can produce up to 45,000 psi. The NEP2320 model can process one sample at a time using a PULSE Tube, or up to 12 samples simultaneously using MicroTubes. The NEP2320 model lists at roughly \$30,000.

*HUB Barocyclers*

Pressure BioSciences’ next-generation PCT platform is the HUB family of Barocyclers. These high-pressure devices are designed for research and manufacturing applications. To the Company’s knowledge, the mid-range Barocycler HUB440 (shown on the right side of Figure 14) is the first portable, ready to use pressure generator designed and suitable for the laboratory bench. The HUB440 model is capable of creating and controlling hydrostatic pressure from 500 psi to 58,000 psi. The HUB design is compact and portable, with the HUB440 model weighing approximately 55 lbs.

Figure 14

NEXT-GENERATION PCT TECHNOLOGY: HUB BAROCYCLERS

Model	Maximum Pressure	Year Developed
HUB160	20,000 psi	2012
HUB440	58,000 psi	2012
HUB880*	100,000 psi	<i>In development</i>

\* Note: This model is in the development stage.

Key Features	
▪ Compact	▪ Computer controlled
▪ Portable	▪ LabVIEW software driven
▪ Versatile	



Source: Pressure BioSciences, Inc.

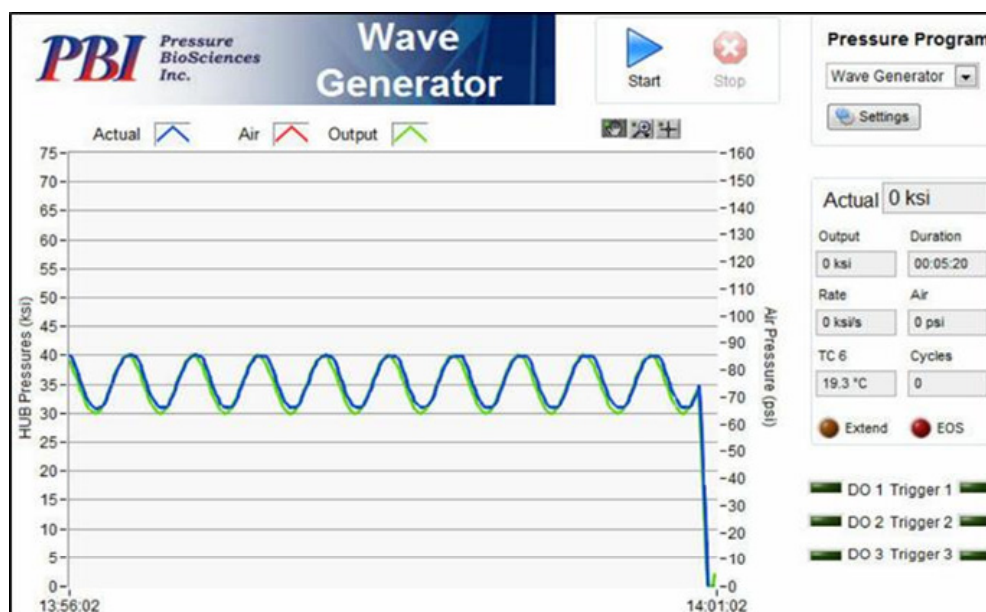
HUB models are designed to serve as a “hub” for a laboratory’s high-pressure needs, with users purchasing specific sample interface accessories as needed to expand the functionality of their HUB unit. The goal of the HUB is to create a modular system that can grow and adapt to a laboratory’s needs. HUB models therefore do not include the stainless steel sample chamber built into other Barocycler models, and instead focus on pressure delivery and cycling controls. In effect, the HUB unit functions as the heart and the brain of a PCT system, and can be interfaced with a variety of “hands and feet” appendages for delivery of the desired pressure cycling regime to a targeted sample. The Company believes that this configuration offers the customer far greater growth potential and application flexibility than the NEP2320 and 3229 instruments, while offering the Company the potential for higher revenue and better gross margins.

Unlike the original Barocycler models, which have a set chamber size and can fit a specified number of samples in each run, HUB models allow a laboratory to purchase a larger size stainless steel chamber to use with its HUB unit to process a greater number of specimens simultaneously. This modularity also allows the HUB pressure system to be more easily paired and interfaced with other devices used in laboratories, such as a **nuclear magnetic resonance (NMR)** or **electron paramagnetic resonance (EPR)** instrument, while continuing to deliver pressure cycles up to 58,000 psi. In the coming years, the Company expects the HUB platform to become the main product family in its PCT-based instrument line.

The HUB can be easily programmed and operated from a standard netbook/laptop computer. Pressure BioSciences has written custom software based on the specialty LabVIEW software platform, which makes the HUB models highly adaptable and versatile in many standard laboratory settings. The Company owns the necessary rights under a license to use National Instruments Corp.’s LabVIEW software. A screenshot of the LabVIEW software is shown in Figure 15 (page 28).

Specifically, the HUB models can be programmed to meet the user's exact specifications for each protocol. Whereas the original Barocycler models can hold up to 99 programmed protocols, the HUB's operator can design, control, and customize an unlimited number of procedures by entering the design specifications for each procedure via the LabVIEW computer software. As an example, the user could program the HUB to begin at 20,000 psi and increase the pressure by 1,000 psi every 60 seconds, or could start at a higher pressure and gradually reduce the pressure on the sample. There are an unlimited number of combinations the user can perform. The current Barocycler NEP2320 and NEP3229 instruments do not have this versatility.

Figure 15  
CUSTOM SOFTWARE FOR HUB UNITS



Source: Pressure BioSciences, Inc.

The HUB system was named after Dr. Wayne L. Hubbell, a distinguished professor of chemistry and biochemistry and the Jules Stein professor of ophthalmology at UCLA. Dr. Hubbell approached the Company in 2010 with a request for a device that would allow him to look at proteins in an EPR instrument under pressure, in order to study the structure of proteins as they change under pressure. This ability may help to improve researchers' understanding of the structure and function of proteins, potentially improving the discovery process and providing new insights into such important areas as biomarker discovery and rational drug design. Dr. Hubbell sponsored the development of this technology and worked closely with Pressure BioSciences throughout the development of the HUB model. The earliest version of the technology was introduced in 2011 to collaborators for the purpose of testing the machine in a laboratory environment and obtaining feedback from research scientists on the HUB's performance and capabilities.

#### *Status of HUB Development and Sales*

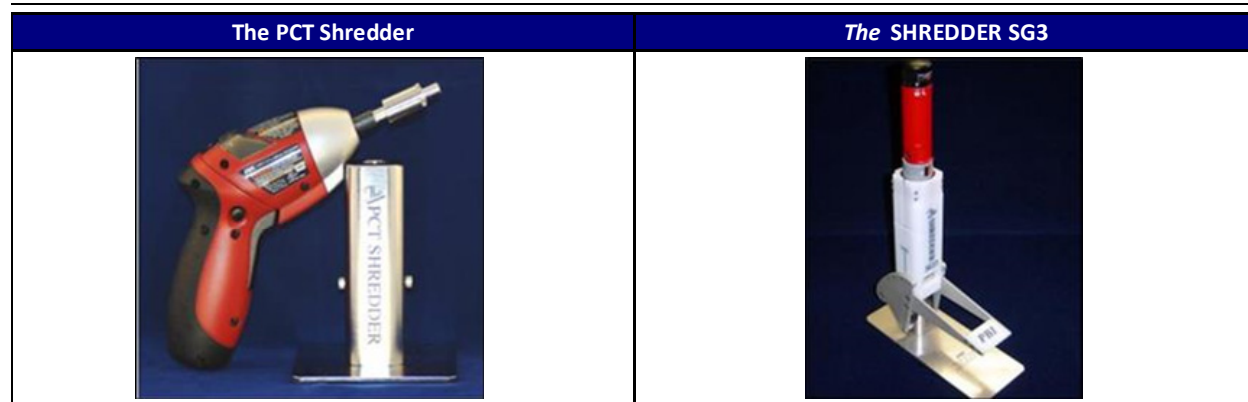
The HUB technology has remained largely in the research and development phase as the Company fine-tunes the technology and prepares for commercialization. To date, the Company has sold roughly 10 HUB440 models, which retail at approximately \$30,000 each (base unit; does not include the accessories). Under the guidance and funding of Dr. Hubbell, Pressure BioSciences is working to develop a HUB880 model that can reach 100,000 psi. To the Company's knowledge, the HUB880 system would be the first device to provide 100,000 psi pressure cycling in a convenient, table-top laboratory format.

## Shredders

Pressure BioSciences' Shredder systems encompass devices designed to rapidly shred, or grind, biological samples, such as tissues or organisms. The shredding process is fast, safe, and requires minimal energy. This low-shear mechanical disruption process, when combined with select buffers, can effectively extract proteins, DNA, RNA, lipids, and small molecules. Shredder systems provide superior efficiency and performance in extracting biomolecules from samples, versus many competing technologies and products. The shredding is performed entirely in the Company's Shredder Tubes, which double as PULSE Tubes, facilitating direct continuation in the Company's Barocyclers for further extraction requirements to the molecular level, while keeping the sample enclosed and avoiding handling and transfers between containers.

The Company's base system is The PCT Shredder, which is shown on the left side of Figure 16. Pressure BioSciences' second model, called *The SHREDDER SG3™* ("SG3"), is shown on the right side of Figure 16. The SG3 model is similar in function to the PCT Shredder but offers three settings (as described following the Figure). Both systems are powered by rechargeable lithium batteries and can be used in combination with various Shredder Tubes and buffers. Shredder PULSE Tubes are designed for single use, reducing the risk of sample contamination. The Shredder systems are also compatible with a number of sample preparation kits and reagents sold by major independent commercial suppliers, as well as with kits sold by Pressure BioSciences for specialized sample preparation applications.

Figure 16  
PRESSURE BIOSCIENCES' SHREDDER SYSTEMS



Source: Pressure BioSciences, Inc.

The driver mechanism differs on each Shredder model. The driver for the PCT Shredder is compact and lightweight and resembles a portable drill. The driver on the SG3 model (shown on the right side of Figure 16) is heavy duty and robust. The SG3 system also contains a Force Setting Lever that enables the user to select one of three force settings for the procedure: low ("1"), medium ("2"), and high force ("3") settings. An important advantage of the SG3 system is that the force is reproducible. As well, the varying degrees of force enable the homogenization of many types of tissues and organisms, with the higher settings used for tough, fibrous, and other difficult-to-disrupt samples. The lever also eliminates the need for the operator to exert force for long periods when processing one or more samples (compared to the PCT Shredder, which requires the user to place hand pressure on the driver). Figure 17 (page 30) illustrates the process of preparing and using the SG3 system. Some preparations can be further enhanced when shredding is followed by PCT, which can improve sample lysis and increase extraction of the desired analytes.

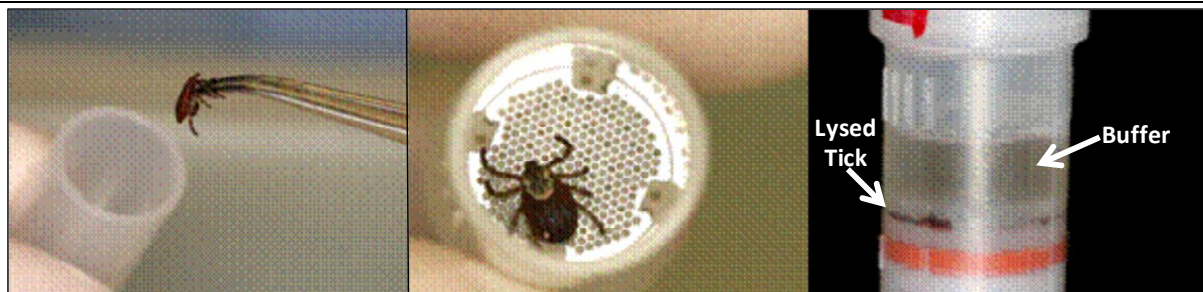
Figure 17  
GENERAL STEPS FOR USING THE SHREDDER SG3



Source: Pressure BioSciences, Inc.

Operators can shred the biological sample as long as necessary until most, or all, of the sample has passed through the lysis disc to the second chamber of the Shredder Tube. Most samples require only 20 to 30 seconds of shredding using Pressure BioSciences’ Shredder system. Figure 18 shows an example of a tick processed with the Shredder. While all shredding produces some heat (due to friction), the short processing time of the Company’s Shredder systems does not significantly heat the sample. While more heat may be generated in harder samples that require longer shredding times, such as tree bark and seeds, this can be minimized by pre-chilling the stainless steel SHREDDER Base in a refrigerator or freezer.

Figure 18  
SHREDDING PROCESS OF A TICK IN THE PCT SHREDDER OR THE SHREDDER SG3



Tick is placed in the PCT Shredder PULSE Tube (FT500-MS)

A metal insert is used to improve shredding of the tick.

The shredded tick and buffer are shown above after being processed in the Shredder system.

Source: Pressure BioSciences, Inc.

### *Performance of Shredder System versus Other Available Techniques*

Pressure BioSciences' Shredder systems have been compared to other commonly used techniques for homogenizing biological samples. The Company reports that its Shredder systems have produced comparable yields to bead beaters, various mortar and pestle tools, and certain homogenizers, while yielding other important benefits. Pressure BioSciences' systems are less labor intensive than mortar and pestle methods, and do not require transferring of the shredded material for further processing. As well, mortar and pestle techniques require cleaning between extractions, whereas Shredder Tubes are disposable and designed specifically for single use.

A comparison of the PCT Shredder system and bead beating methods with spinach samples showed that Pressure BioSciences' system achieved comparable yields, while better preserving longer DNA strands. When apple seeds were used, the Company's Shredder system with metal inserts yielded higher molecular weight genomic DNA than a bead beater, which causes significant shearing or "breaking up" of DNA.

Pressure BioSciences' systems also offer benefits over homogenizer techniques. When compared to **Dounce homogenization**, Pressure BioSciences' systems offered greater reproducibility and consistency. The Company's Shredder system also achieved similar yield and quality of mitochondria as a Teflon/glass homogenizer from a sample prepared from rat skeletal muscle, while also offering greater ease-of-use, lower costs, and improved safety. In all cases, where superior extraction via PCT was observed, the PCT compatible Shredder Tubes eliminated the need for further sample exposure, handling and transfer between containers. The Shredder system also generated far less heat compared to the other methods. This is an important benefit since the generation of heat often leads to a degradation in sample quality.

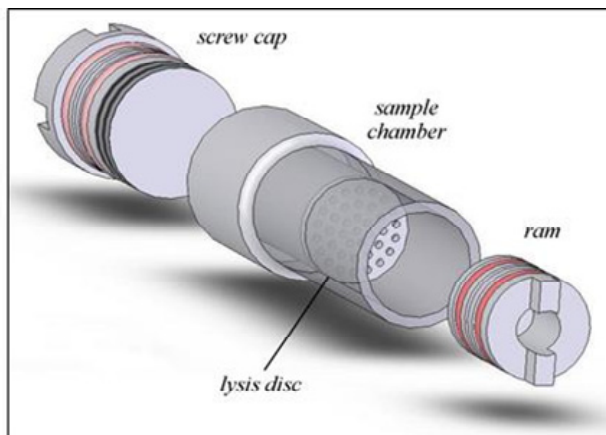
### **PULSE™ Tubes**

Pressure BioSciences sells specially designed processing containers, called PULSE Tubes (**P**ressure **U**sed to **L**yse **S**amples for **E**xtraction). PULSE Tubes are designed to withstand and transfer the pressure from a Barocycler instrument to the sample. The plastic tubes are designed for single use, thereby minimizing the risk of cross contamination between samples as well as the risk of exposure to the user. Currently, the Company offers two PULSE Tube varieties, detailed below.

#### *PULSE Tubes (FT500)*

The FT500 PULSE Tube (shown in Figure 19 [page 32]) is a consumable product designed for use with Pressure BioSciences' Barocycler and Shredder instruments. Each FT500 PULSE Tube has two compartments. The compartments are separated by a flat metal plate with small holes called a lysis disc, which helps to break up (or homogenize) samples. Each tube has a small movable piston, or ram, that serves to push a specimen from the sample chamber through the lysis disc and into the fluid retention chamber at the bottom of the tube as the pressure from a Barocycler increases. When pressure is released, the ram recedes and a portion of the sample (now partially homogenized) is pulled back through the lysis disc into the sample chamber.

Figure 19  
FT500 PULSE TUBE



Source: Pressure BioSciences, Inc.

Figure 20  
FT500-ND PULSE TUBE



Source: Pressure BioSciences, Inc.

#### Non-Disk PULSE Tubes (FT500-ND)

After commercially releasing the FT500, Pressure BioSciences received demand from consumers who sought a PCT consumable to process solutions and suspensions that do not require partial homogenization by passing through a lysis disc. Consumers also requested a consumable that could accept smaller sample volumes. In response to these market demands, Pressure BioSciences developed the FT500-ND PULSE Tube. The FT500-ND maintains the same function as the FT500, but does not have a lysis disc separating the tube into two chambers. The FT500-ND also supports variable sample volumes with a range that can be five times that of the FT500.

#### MicroTubes and MicroCaps

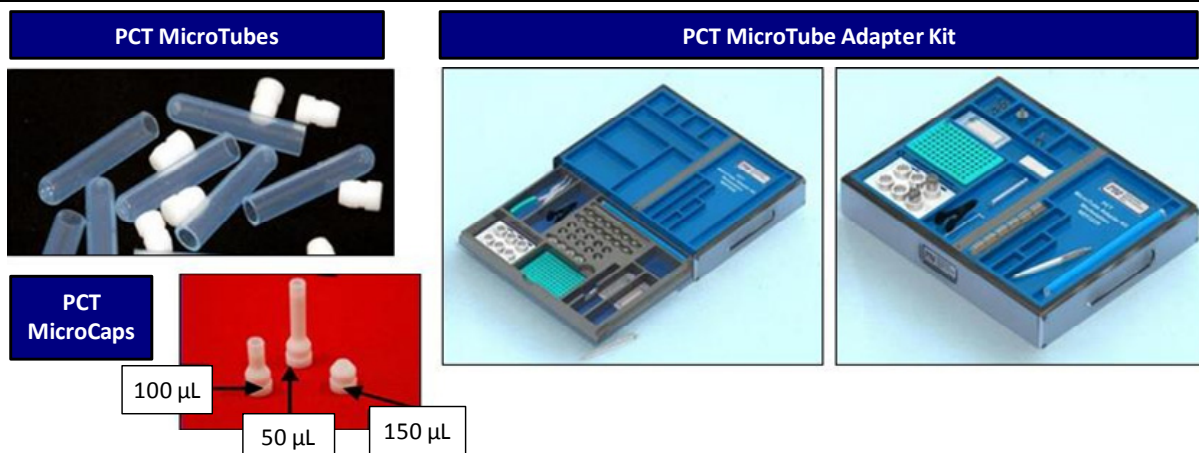
Pressure BioSciences also offers products to support higher-throughput PCT, including MicroTubes and MicroCaps. Whereas the Barocycler NEP3229 can run up to three PULSE Tubes at a time, users can place up to 48 MicroTubes in a specially designed holder to enable more samples to be tested simultaneously.

The PCT MicroTube, shown in Figure 21 (page 33), is a small plastic tube composed of a **fluoropolymer** called fluorinated ethylene propylene (FEP). The PCT MicroTube is designed to meet the critical requirements of modern proteomic analysis by **mass spectrometry**. It can withstand temperatures ranging from -200°C to 100°C. Other features that are important for high-pressure-enhanced enzymatic proteolysis include chemical resistance, a non-wetting surface, and negligible protein and nucleic acid adsorption. These characteristics facilitate nearly complete recovery of samples. Once a sample is placed in the PCT MicroTube, the tube is closed with a MicroCap. The PCT MicroCap is available in three sizes to support various sample volumes: 50, 100, and 150  $\mu$ L, and can also be used to transfer small protein spots from a gel to a PCT MicroTube.

The Company sells these products individually or as part of the PCT MicroTube Adapter Kit Workstation (illustrated on the right side of Figure 21). The kit includes PCT MicroTubes and MicroCaps, a space-saving workstation, and specialized tools to enable the user to process up to 48 samples simultaneously in the Barocycler NEP3229.



Figure 21  
PCT MICROTUBES AND PCT MICROCAPS



Source: Pressure BioSciences, Inc.

### Kits and Reagents

Pressure BioSciences also offers a number of kits and reagents that are designed specifically for use with PCT. A reagent is a substance or mixture for use in chemical analysis or other reactions. One widely used reagent in proteomic research is an antibody, which is used to capture and detect proteins. The Company's reagents and kits, as summarized in Figure 22, facilitate scientific work by reducing time, labor, and costs.

Figure 22  
PRESSURE BIOSCIENCES' KITS AND REAGENTS

ProteoSolve-SB (Systems Biology)	A PCT-dependent method for simultaneously extracting, isolating, and fractionating nucleic acids (DNA and RNA), proteins, and lipids from animal and plant samples routinely used in laboratory research. This patent-pending kit contains proprietary reagents and PULSE Tubes.
ProteoSolve-CE	A PCT-dependent kit for extracting proteins from various samples using an optimized detergent-based reagent system. The kit contains reagents to extract either denatured or non-denatured proteins, which can then be used for the analysis of protein structure and function.
ProteoSolve-TD	PCT-dependent methods for extracting intact membrane proteins. ProteoSolve-TD1 facilitates recovery of intact membrane proteins from cells harvested from culture and from soft tissues (e.g., blood cells). ProteoSolve-TD2 enables recovery of intact membrane proteins from solid tumors and other tissues when proteins are not efficiently extracted by ProteoSolve-TD1. Both kits are compatible with most subsequent proteomic analyses.
ProteoSolve-IEF Reagent	A PCT-based kit for solubilizing many types of proteins during sample preparation for a variety of analytical techniques, including isoelectric focusing (IEF) and gel- or solution-phase protein fractionation. This reagent contains powerful chaotropic agents and a detergent.
Mitochondria Isolation Kits	Kits containing the chemical ingredients necessary for a scientist to extract mitochondria from skeletal muscle and lung tissue for subsequent analysis.

Source: Pressure BioSciences, Inc.

## Sales and Margins

Pressure BioSciences has begun commercializing its PCT products and related consumables. To date, the Company has installed approximately 235 Barocyler units. Pressure BioSciences reported more than \$1.2 million in annual revenues in 2012, representing a 21% increase over revenues of \$988,000 in 2011. The Company's revenues are composed of sales of PCT-related products and services as well as grant funding. Most recently, in the second quarter 2013, the Company's total revenues were nearly \$358,000, up 10% from approximately \$324,900 in the corresponding timeframe in 2012.

Pressure BioSciences reports that its gross margins are approximately 55% to 60% on its current generation of Barocyler units. The next-generation HUB models are expected to have equal or even greater gross margins, as they have a higher retail price combined with a lower cost of production. The consumable products that are sold with Pressure BioSciences' Barocyler instruments, including the PULSE Tubes and the MicroTubes, have gross margins of roughly 65%. The Company expects Barocyler gross margins to decline as it begins to develop a more active sales and marketing program. On the other hand, the Company expects consumables gross margins to increase as sales of PULSE Tubes, Microtubes, and Kits increase concomitant with the expected increase in instrument sales.

## Applications for Pressure Cycling Technology

Pressure BioSciences' PCT platform is a highly versatile and adaptable technology. The Company has identified a range of applications and fields of use for PCT, a selection of which are highlighted in Figure 23. While Pressure BioSciences believes that its products have potential in each of these markets, the Company is currently focusing its product development efforts on biological sample preparation for life sciences research, which spans proteomics (the study of proteins), genomics (genes), transcriptomics (RNA), and lipidomics (fats), among other areas. In particular, Pressure BioSciences is focusing on two sectors: (1) biomarker discovery (primarily through mass spectrometric analysis) within the proteomics field of research; and (2) forensics, which is part of genomics.

Figure 23

### A SELECTION OF APPLICATIONS FOR PRESSURE CYCLING TECHNOLOGY

#### Proteomics (Proteins)

- **Biomarker Discovery:** Heart disease and cancer
- **Pressure-Enhanced Enzymology:** Trypsin digestion and trypsin and deglycosylation
- **Enhance/Enable Protein Analysis:** Mass spectrometry, electron paramagnetic resonance (EPR), electron spin resonance (ESR), and nuclear magnetic resonance (NMR)
- **Enhance Protein Extraction from FFPE:** Diagnostics and drug development
- **Biopharmaceuticals, Vaccines, and Biosimilars:** Discovery, synthesis, and quality control

#### Genomics (DNA, RNA)

- **Forensics:** Rape kit analysis, bone, and touch samples
- **Agriculture:** HLB disease (greening disease in citrus) and soil pathogens
- **Environmental Biology:** Oil-eating bacteria
- **Anti-bioterror:** Rapid identification and inactivation

**Transcriptomics (RNA):** Gene expression and drug discovery

**Lipidomics (Fats):** Lipid analysis of fecal material, metabolomic diseases (e.g., diabetes)

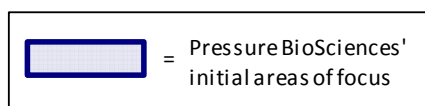
**Mitochondria:** Mitochondrial-related diseases (e.g., muscular dystrophy, autism, cerebral palsy)

**Food:** Pathogen testing, manufacturing

**Herbal Medicine:** Discovery, manufacturing

**Chemical:** High-pressure chemical syntheses and analysis

**Industrial Applications:** Automotive, oil well discovery and recovery



Source: Pressure BioSciences, Inc.

Pressure BioSciences' technology has also been used to study bio-therapeutics, vaccine development, stem cell research, soil and plant technology, and counter-bioterror applications. The Company expects this list to continue expanding as customers in other markets are exposed to and recognize the new capabilities delivered through PCT.

#### Strategic Collaboration Program

An important component of Pressure BioSciences' business strategy is its collaboration program. Third-party researchers who sign up for this program can install a Barocycler instrument in their laboratory under favorable lease terms for an agreed upon period of time (typically three to six months) in exchange for completing a predetermined work plan. The Company has identified several objectives for this program: (1) developing new applications for PCT in sample preparation; (2) advancing and validating the Company's understanding of PCT within an area of life sciences in which it already has products; (3) demonstrating the effectiveness of PCT to specific research scientists who the Company believes can have a positive impact on market acceptance of PCT; and (4) potential for peer-reviewed publications and presentations at scientific meetings by a third party on the merits of PCT. This program continues to provide Pressure BioSciences with independent and objective data about PCT from well-respected laboratories and key scientific and medical opinion leaders across the U.S., and in some cases, throughout the world.

## BIOMARKER DISCOVERY: PRESSURE BIOSCIENCES' FIRST TARGETED MARKET IN PROTEOMICS

Proteomics—a derivative of the words proteins and genomics—is the study of the structure and function of the proteome. The term “proteome” refers to the entire collection of proteins that are or can be expressed by a cell, tissue, or organism. Unlike the genome, which is relatively static, the proteome changes continually based on health or disease, the nature of each tissue, the stage of cell development, and effects of drug treatments. For this reason, the proteome is typically defined as the proteins present in one sample (tissue, organism, cell culture) at a certain point in time.

Proteomics covers a number of key aspects of protein function, as described below.

- *Structural Proteomics.* The analysis of protein structures. Protein structure comparisons can help to identify the functions of newly discovered genes. Structural analysis can also show where drugs bind to proteins and where proteins interact with each other.
- *Expression Proteomics.* The analysis of protein expression. This can help identify the main proteins found in a particular sample and proteins differentially expressed in related samples, such as diseased versus healthy tissue. A protein found only in a diseased sample may represent or lead the way to a useful drug target or diagnostic or prognostic marker.
- *Interaction Proteomics.* The analysis of protein interactions. The characterization of protein-protein interactions helps to determine protein functions and pathways, and can also show how proteins assemble in larger complexes.

Over the last decade, progress in the challenging field of proteomics has positioned it for resurgent importance in the fields of diagnosis, prognosis, and drug research. Proteins constitute most of the biochemically functional molecules in cells, and thus in entire organisms. When mutations occur in DNA (which contain coding sequences for production of different proteins), it is the proteins that are ultimately affected. Additionally, the majority of drugs interact with proteins to offer beneficial effects to patients. The global proteomics market is estimated to reach \$17.2 billion by 2017, expanding at a 14.2% CAGR (Source: MarketsandMarkets, *Proteomics Market-Instruments, Reagents and Services-Trends and Global Forecasts to 2017*, November 2012). Growth in this market is fueled by several factors, including new innovations and developments in proteomic instruments, greater availability of funds from various organizations, and higher R&D expenditures (Source: MarketsandMarkets).



"In my opinion, high pressure will play an important role in the discovery process that lies ahead in the exciting field of protein science."

-- Dr. Wayne L. Hubbell

*Distinguished Professor of Chemistry and Biochemistry and Jules Stein Professor of Ophthalmology at UCLA*

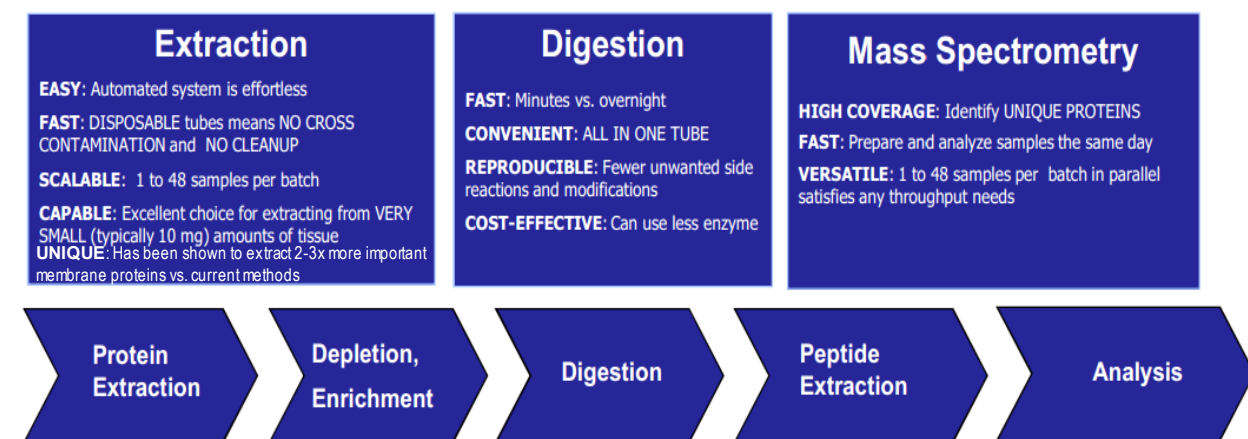
Researchers use a variety of techniques to study the proteome, including mass spectrometry, protein microarray, spectroscopy, chromatography, electrophoresis, surface plasmon resonance, and x-ray crystallography, among other technologies. Before a sample is analyzed with a mass spectrometer or other instrument, it is often subject to sample preparation procedures (introduced on pages 19-21). Sample preparation is an important part of proteomic research. While intended to improve the quality of results, it can negatively affect the outcome of the sample analysis if the sample is contaminated or if components are accidentally or unwittingly removed or damaged during the process. This is the motivation behind Pressure BioSciences' slogan: *Discovery Starts with Sample Preparation™*.

The use of high pressure in a proteomic workflow, such as occurs in Pressure BioSciences' PCT products, has significantly enhanced cell lysis and accelerated digestion of proteins (an important sample preparation phase that occurs prior to protein profiling analysis) as well as improved throughput. Additionally, the use of high pressure has facilitated the recovery of proteins that have been difficult to recover from samples previously, including membrane proteins, which have key biological roles in cancer, drug resistance, and viral infections. To this point, it is estimated that 70% or more of current biological drugs in development are directed against membrane proteins.

As highlighted in Figure 24, Pressure BioSciences' pressure cycling products and technologies are beneficial in several areas of the proteomic workflow. PCT has been shown to help extract proteins from a sample in an automated, rapid, and scalable manner. Additionally, pressure cycling has been shown to aid in protein digestion, in which enzymes are combined with protein samples to break down proteins into smaller components over time.

Figure 24

PRESSURE BIOSCIENCES' PRODUCTS ARE BENEFICIAL IN MULTIPLE STEPS OF A PROTEOMIC WORKFLOW



Source: Pressure BioSciences, Inc.

Research has shown that adding PCT to the workflow can reduce digestion time from overnight to several minutes, with no loss in sample quality. Reduced digestion time can mean faster time to discovery. PCT has also shown to be a useful tool in preparing samples for mass spectrometry, often resulting in improved identification of proteins, i.e., in improved chance for discovery. Mass spectrometers analyze biological samples by measuring the masses and relative concentrations of atoms and molecules. These instruments are widely used in the pharmaceutical industry for drug research and also have applications in the chemical, biotechnology, material sciences, and medical industries. While available mass spectrometry technologies allow scientists to detect and quantify proteins in a complex biological sample, they are not yet capable of separating complex protein mixtures from unprocessed human biospecimens, which is where sample preparation is essential.

Additionally, PCT may also have benefit in protein purification, a series of processes intended to isolate a single type of protein from a complex mixture. This process is especially important for vaccine and drug manufacturers, as these products frequently contain proteins. Current purification techniques often result in the loss of a significant amount of the protein. Pressure BioSciences reports that it has achieved proof-of-concept for protein purification in this context, and holds both U.S. and European patents in this area. The Company believes that a process incorporating PCT for protein purification has the potential to increase protein recovery, improve the quality of the product, and lower production costs.

Identifying unique patterns of protein expression, or biomarkers, associated with specific diseases is one of the most promising areas of proteomics. A biomarker is a measurable substance that can be used as an indicator of a process, event, or condition (e.g., infection or disease). Biomarkers can be measurements of mRNA expression, proteins, proteomic patterns, and lipids, among other biological elements. Biomarkers can be a useful measurement in the diagnosis, prognosis, therapy selection, response monitoring, prevention, surveillance, control, and cure of a number of diseases and medical conditions. Once such example is breast cancer markers, which can be useful in helping shape therapeutic decisions subsequent to a diagnosis of breast cancer.

## **PCT's Potential to Aid in Biomarker Discovery**

While PCT holds potential in a number of areas of proteomics, Pressure BioSciences has identified biomarker discovery as its first target market, with an emphasis on heart disease and cancer. Pressure BioSciences believes that PCT is becoming an important tool for biomarker discovery. The PCT platform offer significant advantages versus current techniques used to prepare samples for downstream testing, particularly mass spectrometry analysis. These advantages have been documented by researchers at established institutions, including the FDA's Center for Biologics Evaluation and Research (CBER), the Harvard School of Public Health, the Armed Forces Institute of Pathology, and Johns Hopkins School of Medicine, as well as by companies that could benefit from the technology, including Thermo Fisher Scientific and Target Discovery, Inc. These advantages have been presented at industry events and published in a number of scientific journals.

The use of PCT in a mass spectrometry workflow has shown to improve speed and quality of results in numerous studies, including in research completed by Dr. Andreas Huhmer at Thermo Fisher Scientific. Pressure BioSciences' has also shown an ability to improve protein identification. With the potential to recover greater numbers of proteins and with higher quality than other available technologies, the Company believes that PCT can become an essential sample preparation tool for pharmaceutical, biotechnology, and academic laboratories focused on biomarker discovery.

The study of proteins as biomarkers could support development of new and improved vaccines, therapeutics, and diagnostics. For example, PCT has demonstrated potential as a useful tool for vaccine research and manufacturing. Previously, a team of researchers at CBER sought to develop an improved method for influenza virus research and vaccine manufacturing. As presented at US HUPO 2010, PCT was used in a critical processing step of CBER's suggested influenza vaccine manufacturing protocol, and was important for improving influenza vaccine quality in a mass spectrometry workflow. This research was led by Dr. Michail Alterman, senior investigator at CBER.

Additionally, at the HUPO 11<sup>th</sup> Annual World Congress in late 2012, Dr. Alterman and colleagues from the FDA presented on the use of an improved method for sample preparation and protein identification, and identified PCT as an important part of this approach. The presented technology, using PCT, had enabled FDA researchers to better identify and analyze events likely to control the biology of multipotent stromal cells (MSCs) from human bone marrow. Current studies primarily focus on highly abundant proteins, which are generally unable to reveal important cellular events. Such information could be essential in the design and development of new, life-saving cell-based therapies for various diseases.

PCT has also been shown to improve protein extraction yields in archived human aorta samples through data collected by researchers at Johns Hopkins University. Aorta tissue is one of the most important tissues used to study heart disease, yet one of the most difficult tissues from which to extract proteins. To address this unmet need in cardiovascular disease research, Pressure BioSciences is developing a product called the Barocycler FFPE Protein Extraction Instrument System. Greater details on this development are provided on pages 52-53.

### *Unique Ability to Extract Greater Quantities of Whole Membrane Proteins*

Membrane proteins are protein molecules that are structurally attached to, or associated with, the membrane of a cell or an organelle. These proteins are involved in many important cell functions, including cell signaling, response, and transportation mechanisms, and also have key biological roles in cancer, drug resistance, and viral infections. As such, membrane proteins are also vital to the development of new vaccines, therapeutics, and diagnostics. Roughly 70% to 80% of new drugs being developed target membrane proteins (Source: *Cytotechnology*, 2009; 61[3]: 153-159).

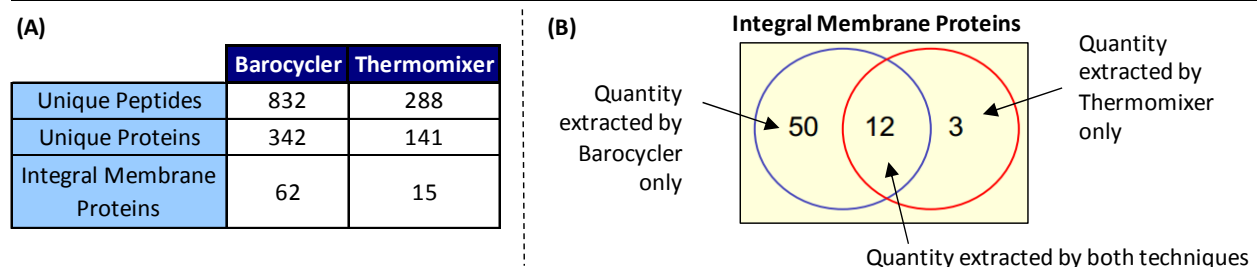
Despite their important roles, membrane proteins and their isoforms have historically been difficult to extract and study. In a cell, membrane proteins are tightly bound to lipids and other proteins within the membrane. Conventional methods for extracting membrane proteins, such as bead beaters and homogenizers, can destroy the structure and function of the proteins and produce useless protein fragments. As such, there is a need in life science research for an automated method to rapidly extract and recover intact, functional membrane proteins from tissue samples. The improvement in the recovery of membrane proteins may be critical to the understanding of diseases and disorders, such as cancer and Alzheimer's, and to the discovery of new drug therapies.

Pressure BioSciences' PCT platform has shown an ability to extract greater quantities of whole membrane proteins. Unlike homogenizers and bead beaters that physically break apart the membrane, Pressure BioSciences combines a tissue sample with select chemicals in a tube and subjects this combination to PCT, creating conditions that gently release the membrane proteins, often intact and functional. At the Harvard School of Public Health in Boston, Massachusetts, scientists demonstrated that PCT could achieve superior recovery of membrane-associated proteins with more than a 20-fold increase in throughput in addition to improved reproducibility (Source: *Journal of Proteome Research*, 2011, 10 [12]: 5536-5546).

Pressure BioSciences reports that its technology has been able to extract membrane proteins from samples that were not recovered in the membrane previously using other available methods. The ability for PCT to extract membrane proteins that have been missed by conventional sample preparation technologies has been confirmed in several publications by researchers at Target Discovery as well as the Institute for Research in Immunology and Cancer, Université de Montréal, Thermo Fisher Scientific, the Harvard School of Public Health, and the FDA. For example, when used on a mixture of nine standard proteins, the use of Pressure BioSciences' Barocycler led to the extraction of four times as many integral membrane proteins as a **Thermomixer**, as shown on the left side of Figure 25 (Part A). A Thermomixer is a compact shaker for mixing and incubating samples. Part B of Figure 25 illustrates the quantity of integral membrane proteins that were found by both techniques (12) as well as the unique number of integral membrane proteins extracted only by the Barocycler (50) versus unique proteins found only by the Thermomixer (3). With as many as 70% of new drugs targeting membrane proteins, Pressure BioSciences believes that this ability is a critical competitive advantage for its PCT system.

Figure 25

PRESSURE BIOSCIENCES' PCT EXTRACTS GREATER QUANTITIES OF INTEGRAL MEMBRANE PROTEINS THAN THERMOMIXER



Source: "The Effect of Pressure Cycling on Proteolytic Cleavage Efficiency, Reaction Time and Protein Sequence Coverage," a poster by researchers at the Institute for Research in Immunology and Cancer, Université de Montréal, and Thermo Fisher Scientific.

Partnership with Target Discovery

Target Discovery, Inc., a personalized medicine diagnostics company based in Palo Alto, California, has sought to develop next-generation clinical diagnostics by assessing protein modification states (the "missing link" in biomarkers) and using this information to create protein isoform diagnostics. One initiative Target Discovery has pursued under the direction of its chief scientific officer, Dr. Luke Schneider, is the development of a new method to extract membrane proteins from solid metastatic ovarian tumors. Target Discovery has used Pressure BioSciences' Barocyclers to extract ovarian cancer markers, and has found that PCT can improve membrane protein recovery in ovarian cancer studies when used as part of the mass spectrometry workflow, especially when used with the ProteoSolve-TD buffer system. The company has also found that the extraction buffers used are compatible with immunoaffinity methods (e.g., **ELISA** and **immunoaffinity chromatography**), as well as with conventional proteomic techniques (e.g., **2D gels**, **western blots**) (Source: *International Journal of Proteomics*, "Method for Recovery and Immunoaffinity Enrichment of Membrane Proteins Illustrated with Metastatic Ovarian Cancer Tissues," Volume 2012).

Pressure BioSciences has a strategic product licensing, manufacturing, co-marketing, and collaborative research and development agreement in place with Target Discovery. The agreement has been in place since 2010. As part of the original agreement, Pressure BioSciences agreed to combine its PCT platform with Target Discovery's proprietary reagents to facilitate the extraction and recovery of membrane protein biomarkers and related isoforms from human tissue. This resulted in a new line of products under the ProteoSolve™ brand for extracting intact membrane proteins from soft tissues, cultures, solid tumors, and other tissues.

When used together, Target Discovery's reagents and PCT are able to stabilize both the structure and function of membrane proteins as they are released from tissue samples, addressing unmet needs for more effective membrane protein extraction and recovery. Moreover, the companies believe that this method could assist with the discovery of important new biomarkers, and the subsequent development or improvement of vital therapeutic and diagnostic products. The companies may continue to develop products incorporating novel reagents and PCT for other tissue processing and extraction applications.

In April 2012, Pressure BioSciences and Target Discovery expanded their agreement, enabling Target Discovery to use the Company's PCT platform commercially in the clinical diagnostics testing market (previously the platform was available on a research-only basis). The PCT platform serves as the sample-processing foundation upon which many of Target Discovery's next-generation clinical laboratory testing services will likely be based. The technology is used to detect proteins that are thought to be biomarkers with utility for important treatment guidance decisions in ovarian and other cancers. Variations in these protein isoforms could translate into important commercial applications. In line with Target Discovery's mission to develop personalized medicine, the goal of this agreement is to use the technology to help guide patients when they must choose from various treatment options.

The first application for this technology is ovarian cancer. Approximately 22,000 women are diagnosed with ovarian cancer each year in the U.S. Target Discovery estimates that more than two-thirds of these patients could be helped by the introduction of reliable treatment selection guidance.

Under the agreement, Pressure BioSciences earns a minimum royalty, which is replaced by an annual royalty once Target Discovery achieves a specified minimum level of diagnostic testing sales. Additional terms under the contract are available at <http://bit.ly/1b44NW5>.

### **Using PCT with Spectroscopy to Improve Discovery and Drug Design**

Research has also been performed using PCT to enhance spectroscopy, which is a branch of science concerned with investigating and measuring spectra produced when matter interacts with or emits electromagnetic radiation. In particular, it may offer improved methods for preparing protein samples for spectroscopy, electron paramagnetic resonance (EPR), electron spin resonance (ESR), and nuclear magnetic resonance (NMR). Pressure BioSciences believes that these capabilities could lead to partnerships with larger manufacturers of EPR, ESR, and NMR equipment who have global sales forces that could enhance the Company's visibility.

UCLA researchers have developed a custom, pressure-based EPR system that uses rapid changes in pressure to monitor the rate of protein conformational changes likely related to a protein's function. This method has facilitated the study of dynamic events in proteins that are difficult or even impossible to study by traditional EPR technology, which could be beneficial for biomarker discovery and rational drug design. UCLA's research on this method was presented at the 27<sup>th</sup> Annual Symposium of the Protein Society in July 2013 and at the 8<sup>th</sup> Annual HUPO Conference in March 2012.

Based on positive data generated to date by researchers using pressure-based EPR and NMR systems, Pressure BioSciences believes that PCT can enhance the recovery, detection, and measurement of proteins from a wide variety of samples—information that is important in the design and manufacture of novel diagnostics, therapeutics, and vaccines. The Company is working with renowned scientists in this field—including Dr. Wayne Hubbell, a distinguished professor of chemistry and biochemistry at UCLA—to develop a PCT-based EPR system for commercial sale. Dr. Hubbell sponsored the development of Pressure BioSciences' HUB Barocycler product so that he could look at proteins in an EPR instrument under pressure, something that has rarely been done before. The Company has begun the development of specialized EPR pressure cells (consumables in which a sample is contained during the EPR process) as an important accessory to the modular HUB system.



### **Strategic Partnership to Build Next-Generation Sample Preparation/Data Analysis Platform**

On May 21, 2012, Pressure BioSciences announced an agreement with LEAP Technologies, Sage-N Research, and Valco Instruments to co-develop, market, and sell a next-generation sample preparation/data analysis platform for a specific form of mass spectrometry, called hydrogen-deuterium exchange (H/D exchange). H/D exchange is a chemical reaction carried out in research laboratories to examine changes in the structure of proteins as well as the effects of these changes on protein function. This information is important in the discovery, design, development, and manufacturing of new drugs and other therapeutics. The companies believe that demand for H/D exchange capabilities is growing.

The proposed next-generation H/D exchange sample preparation and data analysis platform is expected to use Pressure BioSciences' PCT systems, LEAP's proprietary robotics and automation equipment, Valco's precise high-pressure valves and fittings, and Sage-N Research's Integrated Data Appliances (iDA) with H/D exchange applications for data analysis. A schematic of the proposed platform was revealed at the 60<sup>th</sup> Annual American Society for Mass Spectrometry Conference in Vancouver, BC, Canada, in May 2012.

### **PCT-SWATH: A Novel PCT-based Method for Identifying Biomarkers**

Researchers from the Institute of Molecular Systems Biology at the ETH Zurich University in Switzerland have also sought to develop an improved method for the identification of biomarkers using PCT. Called PCT-SWATH, this technique combines a novel biomarker strategy with a new protein analysis method, and requires PCT to prepare samples before analysis. This technique was presented at the US HUPO 9<sup>th</sup> Annual Conference along with a case study on prostate cancer to demonstrate the concept.

### **FORENSICS: PRESSURE BIOSCIENCES' FIRST TARGET MARKET IN GENOMICS**

Pressure BioSciences' PCT system also has the potential to improve forensic DNA testing. Since 2009, the Company has collaborated with forensic scientists from leading institutions in the field. The Henry C. Lee Institute of Forensic Science (described on pages 47-48) has focused on evaluating PCT's ability to recover and detect DNA in difficult forensic samples under the direction of Dr. Henry C. Lee himself. At Florida International University (FIU), Dr. Bruce McCord and his team have sought to improve processing of forensic samples from rape kits using PCT. At the University of North Texas Health Science Center (UNTHSC), Dr. Bruce Budowle and his team are focused on using PCT to enhance DNA detection in forensic samples. These laboratories have collected data demonstrating PCT's ability to support faster time to result, automation, and processing of up to 48 samples simultaneously. As a result, PCT methods could lead to standardized processes and protocols that have the potential to relieve the backlog of rape kit samples and better identify DNA from **low copy number specimens**.

The development of PCT for forensic applications has been supported by several Small Business Innovation Research (SBIR) grants from the National Institutes of Health's (NIH) National Center for Research Resources (NCR). These funds were used to study PCT's ability to release DNA and RNA from tissues or difficult-to-disrupt cells, such as certain viruses and bacteria.

The Company has several patents protecting its technology in the forensics market, particularly as it relates to nucleic acids. Nucleic acids are complex compounds found in all living cells and viruses that allow organisms to transfer genetic information from one generation to the next. The two main types of nucleic acids are DNA and RNA. Pressure BioSciences' U.S. patent No. 6,111,096, entitled "Nucleic acid isolation and purification," covers methods and devices using PCT to isolate and purify nucleic acids from a broad range of sample types, including forensic samples, blood and other body fluids, and cultured cells. Additionally, U.S. patent No. 6,258,534, entitled "Pressure Controlled Nucleic Acid Hybridization," covers the use of PCT in nucleic acid hybridization and amplification, which are methods routinely used by forensic laboratories in DNA analysis.

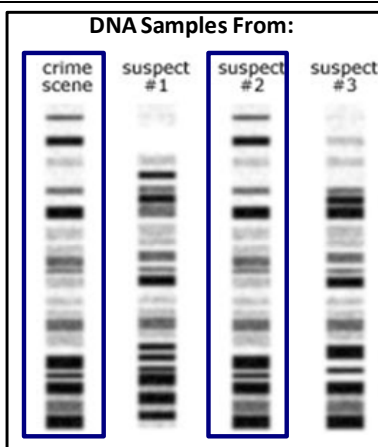
## DNA Forensics in Rape Investigations

Rape is a prevalent crime in the U.S. In 2010, an estimated 270,000 women reported rape or sexual assault incidents (Source: U.S. Department of Justice, *Female Victims of Sexual Violence, 1994-2010*, March 2013) and many more instances are thought to be left unreported. Victims who report being raped are directed to a hospital or other medical setting for treatment as well as to potentially collect evidence. Rape victims can elect to complete a sexual assault evidence kit (or “rape kit”) to collect DNA evidence from their body and clothing that may be helpful in identifying and prosecuting the perpetrator. It is used to collect blood, urine, semen, hairs, fibers, and fingernail scrapings, among other evidence. The process generally takes four to six hours to complete. If the victim chooses to report the crime to police, the rape kit is booked as part of the police evidence in the case. Law enforcement agents can then send the rape kit to a crime laboratory for testing. Once completed, its results can be compared to a national DNA database of known criminals and unsolved crimes.

Current methods used in forensics laboratories in rape kit testing include **differential organic extraction**, **laser microdissection**, and **filtration**. These methods can be time-consuming, cumbersome, and inefficient, as each kit can take up to two days to complete analysis. In a scenario where a female was attacked by a male, it is common for a swab sample to be collected from the rape victim. The swab can contain cells from both the victim and the assailant (sperm from the male and **epithelial** cells from the female, both of which contain DNA). The male and female cells must be purified and separated before a crime lab can identify the male DNA. If processed without proper preparation, these samples would result in a mixture of DNA from the perpetrator and from the victim, making the sample unusable.

Conventional methods for analyzing rape kits require the separation of sperm from female epithelial cells so that each can be tested individually. The analyst has to manually extract and separate male and female cells collected with swabs from the victim. In addition to being a complex and time-consuming process, this procedure is not aligned with modern efforts seeking to automate forensic technologies. Once separated, the forensic scientist must initiate a number of procedures designed to break the cell membrane and release its contents (e.g., DNA).

Figure 26 *How DNA Is Applied in Forensics*  
DNA SAMPLE COMPARISON



Source: <http://evolution.berkeley.edu>.

DNA is the biological blueprint of humans and most other living organisms. Most DNA is found in the cell nucleus but a small amount can be found in the mitochondria (called “mitochondrial DNA” or “mtDNA”). Nearly every cell in a human’s body—whether it is derived from the individual’s blood, saliva, semen, hair root, or other source—has the same DNA. With the exception of identical twins, it is highly unlikely that two individuals have the same DNA. Thus, DNA is a unique identifier used in many forensic investigations to identify the perpetrators of violent crimes and missing persons. The detection of DNA has become a part of the analysis of forensic samples by laboratories and criminal justice agencies worldwide in their efforts to identify the perpetrators of violent crimes and missing persons. As shown in Figure 26, forensic scientists can compare DNA from the scene to DNA from suspects (whether it is volunteered or collected after being convicted in a previous crime), in order to positively identify a criminal.

Samples from a crime scene or rape kit often contain suboptimal quantity and/or quality of DNA. Low level DNA samples do not yield reproducible and reliable results. Significant amounts of a sample of DNA are necessary for genetic analysis; however, this may not always be readily obtained from a rape kit, as only a small amount of the perpetrator’s DNA may have been collected. If needed, forensic scientists can use a technology called polymerase chain reaction (PCR) to amplify (or copy) small segments of DNA. For this reason, PCR is also referred to as “molecular photocopying.” This process is both fast and inexpensive, and can be used to create more than one billion exact copies of the original DNA segment within hours.

Some types of samples are even more challenging to analyze, such as bone and hair. Methods for preparing these particularly difficult forensic samples for analysis are often inadequate or even unavailable. Additionally, the presence of DNA contaminants or **inhibitors** in a sample can make DNA harder to detect and cause a poor or invalid result. Contaminants that copurify with DNA can affect the yield of PCR products and lead to interpretational difficulties. However, technologies designed to lessen the effects of these contaminants/inhibitors can improve the quality of the forensic DNA test. In particular, high-pressure technologies, such as Pressure BioSciences' PCT system, have been shown to minimize the effects of inhibitors and improve the yield of PCR products (Source: *International Journal of Legal Medicine*, March 2013; 127[2]:321-33).

Once the sample preparation process is complete, the DNA is compared to national and international DNA databases of known individuals to determine possible sources of DNA. In the U.S., the FBI maintains the Combined DNA Index System (CODIS), which is the largest DNA database in the world. CODIS contains DNA profiles from casework and offender samples from all 50 states. It is used to identify perpetrators, find links between cases, and assist in the identification of missing persons and unidentified human remains.

#### *Delays in Rape Kit/DNA Processing*

Despite their potential to aid in solving crimes, many rape kits are left unprocessed. Collectively, these untested kits are referred to as the rape kit backlog. Not every rape case progresses to the investigative stage where law enforcement requests the kit for testing. Only a handful of jurisdictions mandate that every rape kit booked into police evidence is sent to the crime laboratory and tested. Law enforcement agencies outside of these jurisdictions often cite a lack of resources or funding as the reason for their backlog. Individual rape kits can cost up to \$1,500 to collect and analyze (Source: *TIME*, "The Dark Side of Clearing America's Rape Kit Backlog," September 7, 2013). Additionally, even when a rape kit is sent to a crime lab, it can sit for months or years before being tested due to inadequate resources and personnel. Currently, there are an estimated 400,000 untested kits in police storage and crime lab facilities across the U.S. (Source: *TIME*, September 7, 2013).

Each untested kit may be allowing a rapist to remain free and potentially commit more violent crimes. In San Antonio, Texas, a backlog of 2,000 untested rape kits resulted in 207 positive CODIS hits, and five of the offenders were serial rapists (Source: *KSAT-San Antonio*, *Texas legislators approve \$11M to process untested rape kits*, September 9, 2013). In Detroit, testing of 569 untested rape kits (out of a backlog of 11,000 rape kits found abandoned in a Detroit Police Department storage facility in 2009) found 136 CODIS "hits" in 11 states and the District of Columbia, and identified 32 serial rapists (Source: *Michigan.gov*, June 5, 2013).

#### **How PCT Can Enhance DNA Testing in Forensic Investigations**

Through collaborations with leading researchers in the U.S., Pressure BioSciences has focused on the development of its patented PCT system for the forensic science markets, particularly as it relates to extracting DNA and other molecules from a wide variety of samples, including bone fragments, blood, hair, and skin. Studies to date have shown that incorporating PCT into the standard workflow of the forensic DNA testing laboratory could enhance DNA extraction and recovery, reproducibility, standardization, safety, productivity, and overall quality of rape kit testing while also decreasing labor time and cost versus currently available techniques. These improvements in the process could lead to more solved cases and arrests for sex crimes and help to reduce the rape kit backlog. Additionally, the PCT system may improve the workflow of crime labs, forensic companies, academic institutions, and research facilities. Pressure BioSciences estimates that there are over 1,000 such laboratories worldwide. Beyond rape kit and DNA testing, the PCT may also have application in anti-bioterrorism efforts and in detecting counterfeit foods.

#### **Collaboration with Leading Forensic Science Institutes**

Pressure BioSciences has focused on accelerating the development of its forensic-based instruments and consumables through key partnerships in the field. These relationships have validated PCT's potential as a new, effective technology for forensic DNA testing, particularly for analyzing samples that have proven to be challenging using today's techniques (e.g., as for bone and hair samples) as well as for re-evaluating aged biological evidence in cold cases. The Company believes that these collaborations could support its path to market for PCT-based

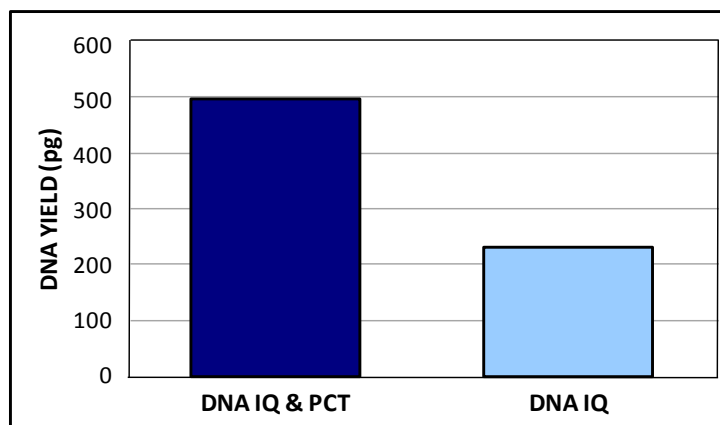
forensic products. To this extent, in April 2013, Pressure BioSciences announced plans to expand its collaborative efforts with key forensics groups focusing on rape kits to accelerate the development and launch of a PCT-based Rape Kit Processing System, augmenting previous research conducted at FIU.

*University of North Texas Health Science Center*

Pressure BioSciences has collaborated with scientists at the Institute of Applied Genetics, Department of Forensic and Investigative Genetics, University of North Texas Health Science Center (UNTHSC) for approximately four years. Over this timeframe, UNTHSC scientists have studied the potential of Pressure BioSciences' PCT system to improve DNA detection in forensic samples. In particular, research at UNTHSC has focused on using PCT in challenging forensic samples, including those that contain inhibitory compounds (e.g., some bone samples) or contaminants, as well as aged, degraded, or damaged samples. Dr. Bruce Budowle, executive director of the Institute of Applied Genetics at UNTHSC, serves as the principal investigator for these studies.

Results from studies to date have demonstrated PCT's potential to increase DNA yield, reduce processing time, decrease costs, and eliminate hazardous organic reagents used in certain extraction techniques. UNTHSC scientists have reported that PCT even improved DNA extraction from challenging forensic samples, such as human hair and bone samples (versus a process that did not include PCT). As shown in Figure 27, bone samples prepared using PCT and a DNA IQ™ sample preparation kit (produced by Promega Corporation) yielded at least a two-fold increase in DNA than DNA IQ™ alone (on average). Researchers at the Institute of Forensic Sciences (Beijing, China) have also identified PCT as an effective method for extracting DNA from bone in forensic applications.

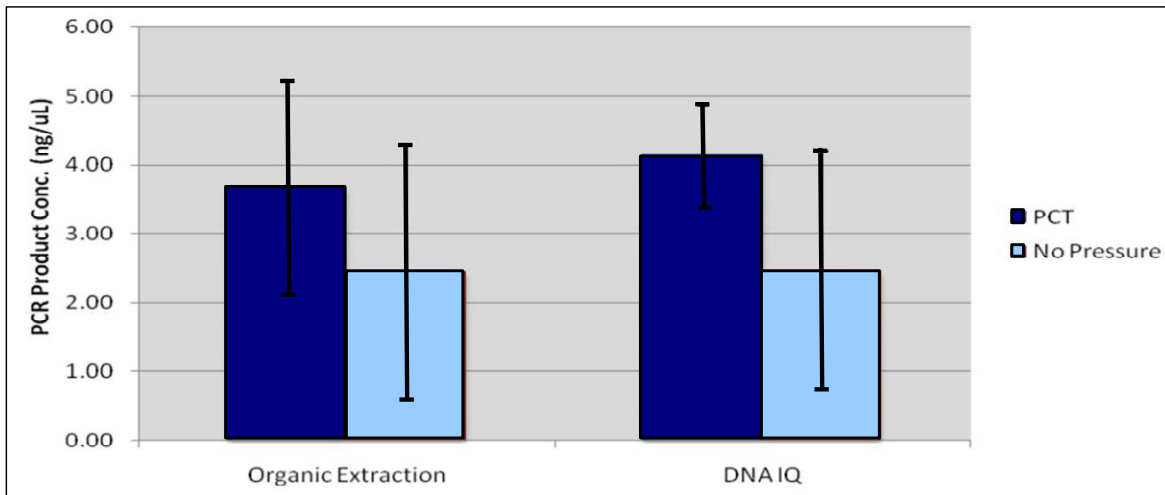
Figure 27  
AVERAGE DNA YIELDS FROM BONE



Source: the University of North Texas Health Science Center's "Pressure Cycling Technology (PCT) Applications for DNA Extractions from Challenging Forensic Samples."

Similar results were found using PCT in hair samples. Conventional methods for extracting DNA from hair shafts necessitate extensive sample preparation and processing, and sometimes require the use of hazardous chemicals. However, even after a post extraction clean-up or concentration step, PCR inhibitors can still interfere with downstream sample processing and analyses. In light of these challenges, UNTHSC researchers have investigated the use of PCT in increasing DNA recovery while preserving the quality of the DNA from hair samples. In one study, UNTHSC researchers used either organic extraction or the DNA IQ™ sample preparation kit, both with and without PCT. As shown in Figure 28 (page 45), samples extracted with organic method and processed with PCT resulted in a 67% increase in PCR product concentration (ng/uL) on average versus organic extraction alone. Similarly, hair samples prepared with the DNA IQ™ kit and PCT yielded 59% greater product yield on average versus DNA IQ™ alone.

Figure 28  
AVERAGE mtDNA YIELDS FROM HAIR



Source: the University of North Texas Health Science Center's "Pressure Cycling Technology (PCT) Applications for DNA Extractions from Challenging Forensic Samples."

Data from studies conducted by UNTHSC scientists have also shown that applying PCT to forensic samples containing inhibitory compounds can improve DNA analysis. In a study of bone samples, PCT appeared to reduce the effects of inhibitors resulting in a significant improvement in DNA testing. UNTHSC scientists presented these results at the 23<sup>rd</sup> Annual International Symposium on Human Identification (ISHI) held from October 15-18, 2012, in Nashville, Tennessee. In March 2013, UNTHSC researchers published their findings in *The International Journal of Legal Medicine* in an article entitled "Pressure cycling technology (PCT) reduces effects of inhibitors of the PCR." In this study, the scientists applied PCT to DNA exposed to various concentrations of two potent PCR inhibitors: (1) **hematin**, a substance formed as bloodstains age; and (2) **humic acid**, a residue of decaying organic matter. PCT was shown to reduce the effects of the inhibitors and increase the yield of DNA in contaminated samples (Source: *The International Journal of Legal Medicine* 2013; 127[2]:321-33).

These findings support the belief that PCT can improve the ability to analyze challenged or inhibited DNA samples, and may aid in the identification and prosecution of known sexual offenders by comparing the DNA to massive DNA databases. These results were also presented by Pamela Marshall, a Ph.D. candidate at UNTHSC, during the Mid America Forensic DNA Conference in April 2013. This study was supported by federal grant No. 2009-DNBX-K188, a nearly \$936,000 award by the U.S. Department of Justice to UNTHSC for studying "improved tools and interpretation guidelines for examining limited low copy number DNA obtained from degraded single source samples: bones, teeth, and hairs."

UNTHSC has also found that PCT can be used to extract and enrich DNA from **touch samples**, which typically yield too little DNA for accurate typing. Importantly, in an interview on *MoneyTV with Donald Baillargeon*, Dr. Budowle stated "With [Pressure BioSciences' PCT] technology, we've been able to get DNA from samples that yielded no results before." The full interview is available at <http://www.youtube.com/watch?v=x6zDnMnqzl>.

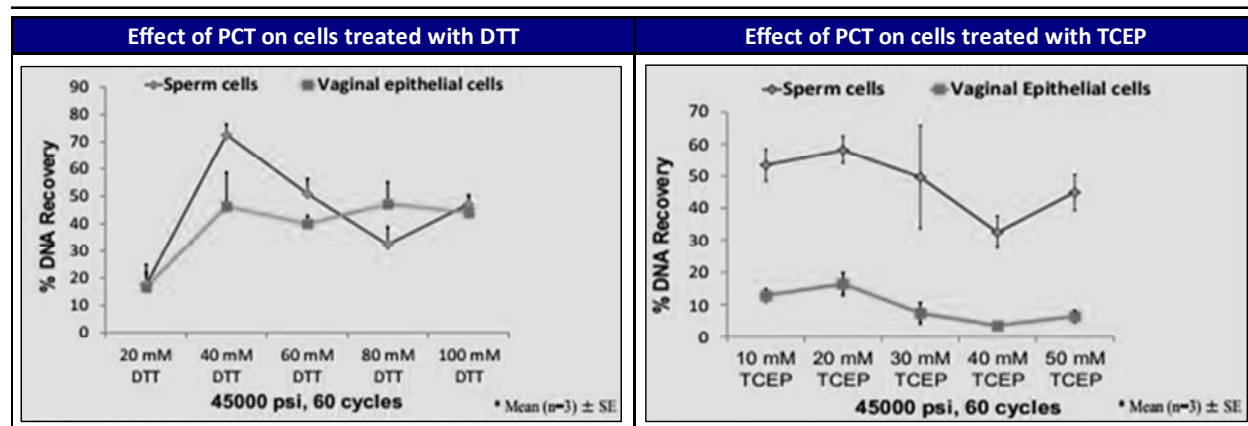
#### Florida International University (FIU)

Since 2010, FIU and Pressure BioSciences have collaborated to develop improved techniques for rape kit testing. Dr. Bruce McCord, a noted expert in forensic DNA research and the associate director of FIU's International Forensic Research Institute (IFRI), and his team at FIU are researching methods to selectively recover male and/or female DNA from rape kits, including high-pressure cycling.

Using Pressure BioSciences' PCT system, FIU sought to develop a method that eliminates the need to separate the male cells from larger quantities of female cells prior to analysis—an ability that could dramatically expedite rape kit testing. In rape kit swabs containing both male and female cells, the PCT system is able to break open and release DNA from the offender's sperm cells while leaving the victim's epithelial cells intact. The Company's system controls the breakage using a combination of chemicals and pressure. The chemicals attach only to the sperm head, making it weaker and more susceptible to breakage than the female epithelial cells. This advantage could help mitigate some of the key bottlenecks in rape kit processing and analysis by significantly decreasing rape kit processing time, increasing throughput, and decreasing costs. As well, it may also facilitate an improved DNA profile of the offender, which can be compared to the millions of profiles contained in various DNA databases globally in an attempt to identify the perpetrator.

FIU has explored the use of PCT in combination with various **reducing agents**. As shown in Figure 29, researchers found that reducing agents Dithiothreitol (DTT) and Tris (2-carboxyethyl) phosphine (TCEP) both improved selective isolation of sperm DNA. Independently, TCEP is more effective in the recovery of DNA from sperm cells, and offers higher selectivity between sperm cell and epithelial cell lysis. Further research at FIU has revealed that combining TCEP and DTT further improves sperm DNA yield in cases of mixed samples (Source: FIU's poster "Differential Extraction of Mixtures in Sexual Assault Casework Using Pressure Cycling Technology [PCT]," American Academy of Forensic Sciences [AAFS] 2012).

Figure 29  
EFFECTS OF PCT WITH DTT AND TCEP



Source: FIU's Application of Pressure Cycling Technology (PCT) in Differential Extraction poster, AAFS 2010.

Research at FIU has confirmed that PCT can selectively extract epithelial and sperm cells. In January 2012, FIU was awarded a \$349,000 grant from the U.S. Department of Justice to help develop and validate a new DNA extraction technique based on the PCT platform that can improve processing of DNA evidence in rape cases.

FIU has continued making progress on this initiative. In April 2013, FIU scientists reported encouraging results from studies using the PCT platform, which were presented at AAFS 2013 annual scientific meeting in a scientific session entitled, "Application of Pressure Cycling Technology (PCT) in Differential Extraction." Preliminary data supported the potential of PCT in analyzing samples from sexual assault cases by demonstrating improved extraction of sperm DNA at high pressures. Positive findings were also reported in a separate presentation, entitled "Application of the IPCRp Method for Genotyping of Male DNA Obtained by Pressure Cycling Differential Extraction." The presentation overviewed a novel technique for cell capture that, when used in combination with PCT, may make a greater quantity of sperm cells available in a rape kit sample for testing with less of the victim's epithelial cells. This ability could be beneficial in creating an improved DNA profile for identifying the rapist, which could aid in arrests and successful prosecutions.

FIU's research on PCT has been presented at a number of industry events beyond AAFS 2013, including the International Symposium on Human Identification (ISHI) 2011, the 17<sup>th</sup> Latin-American Symposium on Biotechnology, Biomedical, Biopharmaceutical, and Industrial Applications of Capillary Electrophoresis and Microchip Technology (LACE 2011), AAFS 2011, and the 21<sup>st</sup> International Symposium on Human Identification, among others.

#### Future Goals and Optimizations at FIU

Dr. McCord and his team are working on optimizing their process for separating male and female cells using the PCT system. Currently, the process entails adding two chemicals that attach to and weaken the sperm head before using PCT, which has resulted in excellent recovery of DNA from the sperm cells compared to the epithelial cells. Dr. McCord and his team are focused on optimizing this process to achieve an even higher level of male DNA recovery.

#### *Henry C. Lee Institute of Forensic Science*

In March 2012, Pressure BioSciences entered into a partnership with the internationally recognized Henry C. Lee Institute of Forensic Science (the "HCL Institute"), shown in Figure 30, at the University of New Haven in Connecticut. The HCL Institute sought to examine the potential of the PCT platform to improve the collection of forensic evidence, particularly DNA, in several important areas of forensics. In particular, the HCL Institute is evaluating the Company's PCT platform for extracting DNA and other biomolecules from bone, hair, plant tissue, pollen, finger nails, and other types of samples that are important during a criminal investigation.

Figure 30

THE HENRY C. LEE INSTITUTE OF FORENSIC SCIENCE



*Source: the Henry C. Lee Institute of Forensic Science.*

The HCL Institute is also testing the PCT platform's ability to detect counterfeit foods. According to a recent study by the U.S. Pharmacopeial Convention ([USP], a scientific nonprofit organization that helps set standards for foods and medicines), the food products that are counterfeited most often are olive oil, milk, saffron, honey, and coffee. Tea, fish, clouding agents (used in fruit juices, like lemon, to make products look freshly squeezed), maple syrup, and spices are other food products commonly counterfeited. Many sushi restaurants have been found to misrepresent the type of fish served, placing consumers at a greater risk of food poisoning. In October 2011, a study of Canadian seafood from 2008 to 2010 revealed that as many as 41% of 236 fish samples were mislabeled (Source: *Mitochondrial DNA*).

Through its relationship with the HCL Institute, Pressure BioSciences has had the opportunity to work side by side with Dr. Henry C. Lee and his team. Dr. Lee is a well-known forensic scientist who has received more than 20 honorary degrees, the Medal of Justice from the Justice Foundation, and who has authored or co-authored more than 40 books and hundreds of articles in professional journals. He has consulted on more than 8,000 criminal cases in 46 countries, including high-profile cases involving O.J. Simpson, John F. Kennedy, Jon Benet Ramsey, Laci Peterson, and many others.

### **Forensics Market Opportunity**

Pressure BioSciences believes that its products offer significant competitive advantages over traditional sample preparation methods used in forensics, which could result in increased rates of identification and arrests of perpetrators in sexual assaults and other crimes. Pressure BioSciences estimates that there are hundreds of forensic laboratories in the U.S. that could benefit from its technology. In 2009 (the latest year for which data is available), there were more than 400 federal, state, county, and municipal forensic science laboratories in operation that collectively received over four million requests for a range of forensic services (Source: U.S. Bureau of Justice Statistics, *Census of Publicly Funded Forensic Crime Laboratories*, 2009).

The forensic technologies market, which encompasses physical crime forensics, laboratory forensics, portable forensics, and other forensic tools and products, was estimated at \$8.3 billion in 2012. The market is expected to more than double to \$17.7 billion by 2019, with DNA profiling representing a high-growth market segment (Source: Transparency Market Research, *Forensic Technologies Market-Global Industry Analysis, Size, Share, Growth, Trends and Forecast, 2013-2019*, August 19, 2013).



## Product Pipeline

Pressure BioSciences emphasizes continued innovation both internally and with the help of strategic partners. Currently, the Company has three products in its R&D pipeline, as noted below.

- (1) *Barocycler HT Multiwell (24-384)*. This product candidate is designed for high-throughput, PCT-enhanced biomolecule extraction/accelerated enzymatic digestion with the capability of processing 24 to 384 samples.
- (2) *Barocycler FFPE Protein Extraction Instrument System*. This is a PCT-based system offering the enhanced extraction of proteins from formalin-fixed, paraffin-embedded (FFPE) samples using a modified Barocycler instrument that combines the advantages of pressure cycling, high temperature, and certain reagents.
- (3) *XstreamPCT™ HPLC Digestion Module*. The XstreamPCT™ is the first module in Pressure BioSciences PCT-based high-performance liquid chromatography (HPLC) platform. XstreamPCT™ is designed for automated, in-line, on-demand PCT-enhanced protein digestion.

### A Core Technology Breakthrough: High-Throughput PCT with the Barocycler HT Multiwell

Numerous studies have confirmed the benefits of using the PCT platform in preparing biomolecules (e.g., DNA, proteins, lipids) for analysis. However, the manual processing required to use the PCT platform has limited the technology's use primarily to small research studies. The Company's current PCT products enable scientists to evaluate up to 48 samples simultaneously using individual tubes; however, each tube must be manually processed. The scientist must physically unscrew the cap, place a sample in each tube, and close the cap before processing. Pressure BioSciences believes that this limitation has prevented the PCT platform from being more widely accepted by the research community.

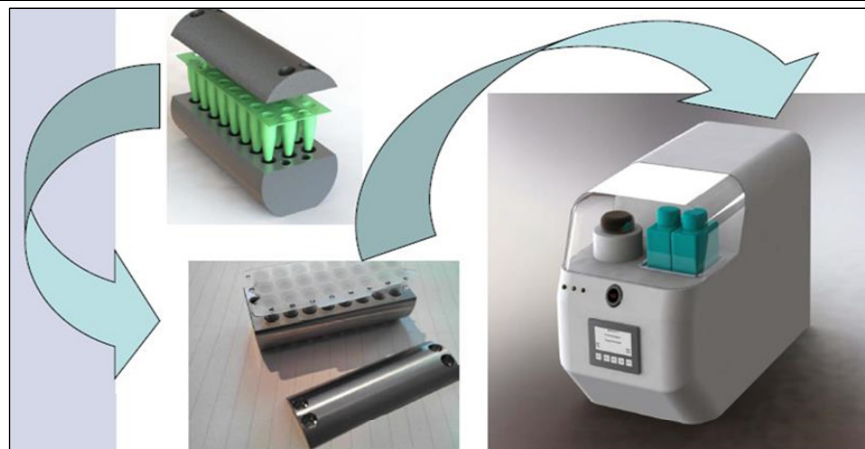
While manual processing works well for small laboratories and individual scientists, larger research and clinical diagnostics laboratories and companies are accustomed to using a more streamlined, automated method for processing large number of samples. Many laboratories turn to automated sample preparation methods to increase the precision and quality of laboratory testing, thus increasing the probability of discovery as well as for developing new and improved vaccines, therapeutics, and diagnostics. These entities use multiwell test plates in standardized formats (e.g., 24, 96, or 384 test wells per plate) to process and test large quantities of samples simultaneously. A machine automatically dispenses liquid into each well before processing. The automation of this process enables scientists to perform other important tasks while samples are being processed. As well, it offers better accuracy and reproducibility versus manual sample preparation. Pressure BioSciences estimates that as many as 80,000 research laboratories work with biological samples worldwide, with a number of these entities using automated, high-throughput sample preparation and analytical systems.

Pressure BioSciences has identified high throughput as a significant growth opportunity for PCT. The Company has invested over four years and several million dollars to develop this technology. On May 30, 2013, Pressure BioSciences announced that it achieved proof-of-principle for a high-throughput PCT system at the Marcum SmallCap Conference. The High Throughput PCT ("HTP PCT") platform combines a modified pressure system with new accessories and modules as well as liquid handling automation technologies from its strategic partner, LEAP Technologies.

For the pressure system, the Company may use its new Barocycler HUB model or a modified instrument. A concept drawing of the Company's first generation high-throughput model, to be called the Barocycler HT Multiwell (24-384), is shown in Figure 31 (page 50). It utilizes a container that holds 24 samples and generates pressure up to 20,000 psi, which is the optimum working pressure for many PCT-enhanced enzymatic applications. Importantly, Pressure BioSciences is developing the system to integrate with the automated, high-throughput sample preparation and analytical systems that are installed in tens of thousands of biological research laboratories worldwide.

Figure 31

PRESSURE BIOSCIENCES' FIRST-GENERATION HIGH-THROUGHPUT PCT SYSTEM CONCEPT



Source: Pressure BioSciences, Inc.

Traditional multiwell plates cannot withstand the high pressures and temperatures demanded by the PCT process. As such, Pressure BioSciences needed to design and develop a custom microwell test container that could withstand the pressure (up to 20,000 psi), temperature, and chemicals in the PCT process. Additionally, the Company had to redesign its pressure system under the guidance of Dr. Edmund Ting and Dr. Alexander Lazarev, who lead Pressure BioSciences' development team. The Company is currently developing a second-generation model, but details have yet to be released publicly.

Pressure BioSciences believes that a high-throughput multiwell product has the potential to fuel growth and increase revenues for existing and new PCT-based applications and products, as well as to facilitate new strategic partnerships. The Company expects beta units to be built and tested by the first quarter 2014, with market-ready units to be available for sale as early as the second quarter 2014.

Pressure BioSciences believes that this product could help increase its penetration in the mass spectrometry area and in other areas of the \$5.4 billion sample preparation market (Source: BCC Research, September 2013). The Company is initially developing its high-throughput PCT platform for three applications:

- (1) rapid protein digestion (multiple labs);
- (2) deglycosylation (biopharmaceutical manufacturing and quality control); and
- (3) lipid extraction (multiple diseases and disorders).

*First Major Market: Protein Digestion*

Pressure BioSciences initially plans to target the protein digestion market. After protein is extracted from a sample, it must be digested before it can be studied. This is achieved by combining the protein with enzymes, such as trypsin, which breaks down protein into peptides and amino acids over time. Once complete, the researcher can place the sample into a mass spectrometer to identify the proteins. Using protein digestion to study proteomics has resulted in a better understanding of disease diagnosis, prevention, control, prognosis, and treatment. Whereas traditional methods can require up to 18 hours to completely digest a protein, the Company reports that the same process can be accelerated dramatically by subjecting the protein and enzyme mixture to high-pressure cycling. Based on published data from multiple customers (as outlined on page 51), Pressure BioSciences believes that its technology can complete digestion in as few as several minutes. In addition, the use of PCT can facilitate the use of much lower concentrations of enzymes, which can reduce costs while simplifying and improving downstream applications.

Pressure BioSciences' High Throughput PCT platform has the potential to accelerate the protein digestion process, while providing the desired automation features. PCT has been shown to enhance and accelerate reactions for a number of enzymes, including Trypsin, Chymotrypsin, Pepsin, Lys-C, Glu-C, Asp-N, Proteinase K, and PNGase F. This effect has been confirmed in both solution- and gel-based digestion protocols. A selection of recent data supporting these findings is summarized below.

- In a presentation at the 8<sup>th</sup> Annual U.S. Human Proteomic Organization (US HUPO) Conference in San Francisco, Pressure BioSciences presented a poster confirming that data to date has shown that PCT can significantly improve quantitative recovery of peptides for those proteins that are otherwise resistant to trypsin digestion, an ability that could help researchers discover proteins that have been traditionally difficult to detect.
- Research from Janssen Research & Development (a Johnson & Johnson Company) has supported PCT's ability to accelerate enzymatic digestion of monoclonal antibodies, which are blood proteins produced in response to a specific antigen. Monoclonal antibodies are widely used in scientific research, in diagnostics, and in therapeutics; however, current digestion methods can take up to 24 hours to complete. The authors reported that PCT significantly reduced the total time required to digest monoclonal proteins and to expedite their complete characterization.
- A study performed by researchers from the Institute for Research in Immunology and Cancer, Université de Montréal and Thermo Fisher Scientific studied the effect of PCT on the protein digestion process. PCT was found to improve reproducibility and provide more efficient proteolysis compared to conventional overnight digestion, and enable the identification of a higher number of integral membrane proteins.
- Research at the University of Missouri School of Medicine has confirmed that PCT with trypsin digestion reduces digest time from hours to minutes and may also facilitate a reduced analyte to trypsin ratio versus when no pressure is used. Additionally, PCT facilitates digestion at ambient temperatures and does not cause tissue maceration (disintegration as a result of excessive soaking).

Pressure BioSciences estimates that thousands of labs in the U.S. perform protein digestion procedures. The Company estimates that the majority of protein digestion is performed with trypsin. The recommended pressure for protein digestion with trypsin is 20,000 psi, which is the level planned for the first-generation High Throughput PCT system.

#### *Deglycosylation*

One of the quality controls for testing protein molecules is testing for sugar molecules, as **glycosylation** often leads to problems in subsequent protein analysis procedures. Thus, researchers use a process called deglycosylation to chemically remove sugars from glycoproteins. This is typically achieved by combining the glycoprotein with enzymes, but as with protein digestion, this process can be time intensive. Similar to protein digestion, deglycosylation may take several hours or require overnight treatment.

PCT has been shown to enhance enzyme reactions in deglycosylation and significantly decrease the time required for these procedures. In one study, pressure cycling at 20,000 psi was shown to accelerate the rate of deglycosylation with enzymes (Source: *Analytical Chemistry*, 2010; 82[6]: 2588–2593). Moreover, the study's authors stated that PCT could support large-scale glycan analysis of biopharmaceuticals with rapid deglycosylation times.

#### *Lipid Extraction*

Lipids, which are a group of naturally occurring molecules that include fats and cholesterol, have been associated with a number of diseases and disorders, such as heart disease, cancer, and obesity. As such, scientists have focused on lipid research to gain better understanding of how to control, treat, and prevent these diseases. It is believed that new and improved laboratory tools that enable a better understanding of gut function could have a significant impact on diagnosing, treating, curing, and preventing such diseases and disorders. One technique researchers use to study lipids are lipid extraction procedures. Lipid extraction procedures are designed to separate cellular or fluid lipids from the other constituents, proteins, polysaccharides, and small molecules (e.g., amino acids, sugars), while preserving the lipids for further analysis.

Importantly, Pressure BioSciences' PCT platform has been found to enhance results of lipid extraction at 20,000 psi. The Company may develop this program in house or may seek to spin out this component of the business.

#### Novel Method for Lipid Analysis in Fecal Material

Understanding the GI system has shown to be an important factor for many diseases, from colon cancer, inflammatory bowel disease, and chronic diarrhea, to disorders such as autism, allergies, and obesity. Thus, new tools and techniques to help researchers better understand the GI system—including through the analysis of fecal material—could help improve the diagnosis, treatment, and prevention of such diseases.

A team of scientists at Harvard Medical School have developed what is believed to be a breakthrough method for lipid analysis in fecal material. In a recent study, lipid analysis of fecal material was used to better understand the GI system and its role in health and disease in premature infants. The Harvard Medical School researchers combined Pressure BioSciences' PCT platform and certain chemicals, and found that in many cases this improved the extraction of lipids from fecal material versus using the chemicals alone. The study was led by Dr. Bruce Kristal, Associate Professor of Surgery at Harvard Medical School and the Department of Neurosurgery at Brigham and Women's Hospital, and the findings from the study were published in *Analytical Chemistry*.

Following the announcement of the Harvard Medical School study on May 16, 2013, an article on Investing.com likened Pressure BioSciences to Exact Sciences (EXAS-NASDAQ), which is developing a noninvasive stool-based DNA technology for detecting colorectal cancer. Despite recommendations for regular screening, many individuals refuse the more invasive colonoscopies, which help diagnose colorectal cancer at an earlier, less deadly stage. In June 2013, Exact Sciences completed its premarket approval (PMA) application to the FDA. Exact Sciences had an approximately \$850 million market cap as of September 2013. Similar to Exact Sciences' test, the fecal lipid profiling technique incorporating PCT is a noninvasive technique for establishing gut function in patients, especially for vulnerable and difficult-to-study populations, such as premature infants and the elderly.

The PCT-based fecal lipid profiling method has market opportunities in both research and clinical applications. On the research side, it could help researchers better understand GI diseases. On the clinical side, it could have both diagnostic and prognostic functions, with potential to help determine and assess intestinal function, response to nutrition, inflammation, and even the occurrence and/or progression of a wide range of gut conditions.

#### **Barocyler FFPE Protein Extraction Instrument System**

Researchers use various techniques to preserve cancer and other tissues for subsequent pathology evaluation. The most commonly used method is formalin-fixation followed by paraffin-embedding ("FFPE"). FFPE tissues are the most widely available specimens for retrospective clinical studies of disease mechanisms. It is estimated that hundreds of millions of FFPE specimens have been archived globally (Source: Frost and Sullivan, June 21, 2012). Data from this research can be used to develop novel diagnostics and therapeutics. However, the fixation process makes extraction of marker proteins from FFPE tissues difficult. As a result, a need exists for novel methods that can accurately, rapidly, and efficiently fix or extract proteins from these samples.

Recently, there has been increased demand for FFPE sample preparation solutions, particularly by pharmaceutical companies and research institutes. Archived tissues, including FFPE samples, represent a feasible alternative to validating biomarkers versus through costly clinical trials. An additional benefit of FFPE archive specimens is that many are associated with a known clinical outcome. This provides an economical and readily available source to validate biomarkers for developing clinical molecular cancer tests (Source: Frost & Sullivan, June 21, 2012).

Positive data using PCT to enhance protein recovery from FFPE samples has led Pressure BioSciences to begin to develop a PCT-based FFPE extraction system. The Company has collaborated with the Armed Forces Institute of Pathology (AFIP) in the development of pressure-based methods aimed at improving the quality and speed preparation for biomolecular extraction of archived FFPE tissue samples, which could help improve research and treatments in the study of various diseases.

Pressure BioSciences has placed a prototype of its PCT-based FFPE Extraction System in the laboratory of Dr. Jennifer Van Eyk, Professor of Medicine at Johns Hopkins University and director of the Hopkins NHLBI Innovation Proteomic Group on Heart Failure. Research at Johns Hopkins University School of Medicine using archived FFPE human aorta samples demonstrated that the combination of cycled pressure and heat were key for strong and steady extraction. As well, PCT combined with high heat improved protein extraction yields by more than six-fold and resulted in a greater number of protein identifications versus heat alone. Dr. Zongming Fu, a member of Professor Van Eyk's proteomics lab at the Johns Hopkins University School of Medicine, presented this data at the US HUPO 9<sup>th</sup> Annual Conference in March 2013. Importantly, the study's authors concluded that the PCT and heat method makes it feasible to test archives of FFPE arterial and aorta samples for biomarker discovery. Pressure BioSciences notes that this is an important achievement, as aorta tissue is one of the most important tissues used to study heart disease, yet one of the most difficult tissues from which to extract proteins.

Additional research studies, including those listed below, have confirmed the benefits of using PCT on FFPE tissue samples.

- Researchers at AFIP and the Department of Veterans Health Affairs (VHA) found that the addition of elevated pressure to all steps in FFPE tissue preparation could improve quality, reduce processing time, and expedite pathology results (Source: *Journal of Cancer*, October 2010).
- Dr. Carol Fowler (AFIP, VHA) presented data at the National Cancer Institute (NCI) showing that the use of high pressure plus heat improved the number of total and unique proteins recovered (4-fold and 1.7-fold, respectively) from FFPE tissues versus heat alone (Source: *Innovative Molecular Analysis Technologies*, October 2010).
- Dr. Timothy O'Leary (VHA) presented data on the use of high pressure and heat to improve protein extraction from FFPE tissues (as presented at the Association for Molecular Pathology, November 2010).

Collectively, Pressure BioSciences believes that these data indicate that its PCT system can efficiently and reproducibly extract significantly more total and unique proteins from FFPE samples than existing non-pressure-based methods. Pressure BioSciences has spent several years developing its PCT-based Barocycler FFPE Protein Extraction Instrument System, which encompasses a modified Barocycler instrument that combines the advantages of pressure cycling, high temperature, and certain reagents to improve extraction of proteins from FFPE samples. The device is designed to support standardization as well as improve speed and safety.

### **XstreamPCT™ HPLC Digestion Module**

In 2008, the Company licensed rights from the Battelle Memorial Institute relating to a method for improving the analysis of protein samples. Battelle Memorial Institute's intellectual property covered an automated system using pressure and a pre-selected agent to obtain a digested sample in less time than current methods while maintaining the sample's integrity.

Pressure BioSciences is using this technology to develop a PCT-based sample preparation method for high-performance liquid chromatography (HPLC) platforms. The Company's goal is to develop instruments that can be directly integrated with HPLC and mass spectrometry for the complete processing of proteins, from sample preparation to final result. Pressure BioSciences is developing its first product under this platform, called the XstreamPCT™ HPLC Digestion Module. XstreamPCT™ is designed for automated, in-line, on-demand PCT-enhanced protein digestion.

## Competition

Pressure BioSciences competes with companies that have technologies for the extraction of nucleic acids, proteins, and small molecules from cells and tissues. Such technologies include techniques like mortar and pestle grinding, sonication, rotor-stator homogenization, French Press, bead beating, freezer milling, enzymatic digestion, and chemical dissolution, as well as new technologies under development that may not yet be commercialized. A number of issues related to the use of these methods may include: complexity, sample containment, cross-contamination, shearing of biomolecules of interest, limited applicability to different sample types, ease-of-use, reproducibility, and cost.

The Company maintains that its PCT Sample Preparation System is a novel enabling system for genomic, proteomic, transcriptomic, lipidomic, and small molecule sample preparation. Many users of current manual techniques would need to be willing to change their existing methods of sample preparation and invest time to evaluate a new method that could change their overall workflow in the sample preparation process. To be competitive in the industry, Pressure BioSciences must clearly demonstrate to its potential customers that its PCT Sample Preparation System provides improved performance capabilities over competing methods, including providing for the following:

- a reduction in labor;
- versatility;
- temperature control;
- efficiency;
- precision;
- simplicity;
- reproducibility; and
- safety.

Since costs for the Pressure BioSciences PCT Sample Preparation System are greater than that of most other techniques currently employed, the Company is focused on emphasizing its product attributes, which it believes will be most important and appealing to potential customers—specifically versatility, reproducibility, quality, and safety.

The market leaders in the sample preparation space include several major firms, such as Agilent Technologies, GE Healthcare, Life Technologies Corp., Qiagen, Sigma-Aldrich Corp., Thermo Fisher Scientific Inc., and Illumina, Inc. In some ways, these firms may be competitive to Pressure BioSciences as the Company seeks to increase its market share for sales of its Barocycler products, but for the most part, the large-cap entities sell instrumentation that can be used in addition to Pressure BioSciences' products, such as EPR or NMR equipment, liquid chromatography devices, or mass spectrometers. As such, these companies could become customers for Pressure BioSciences as well as competitors, by purchasing Pressure BioSciences' sample preparation products to use as part of the sample preparation process or before the sample is analyzed in their own large-scale equipment.

Accordingly, Pressure BioSciences' competition is more likely to be the small and mid-sized companies that participate in this space, such as, but not limited to, those profiled on the accompanying pages. This is a cross-section of companies and is not intended to be an exhaustive collection of potential competitors; however, it is believed to be representative of the type of competition the Company may encounter as it seeks to further commercialize its products/technologies.

**BEE International ([www.beei.com](http://www.beei.com))**

Massachusetts-based BEE International markets high-pressure homogenizer products worldwide. The company started in Israel in 1994 in cooperation with the Technion-Israel Institute of Technology, a major science and technology university. The company's U.S. arm opened in 1998. Today, BEE International reports that it supplies high-pressure homogenizers to global pharmaceutical, biotechnology, chemical, cosmetic, and food industries. The company's products have application in laboratories, pilot plants, and industrial settings, as well as for custom work. Homogenizers are a type of fluid processing equipment that can be used to reduce particle size and for cell disruption. BEE International reports that its high-pressure homogenizers can uniformly reduce particle size to nanoparticles as well as accomplish high-yield cell disruption, and are scalable technologies up to pilot and production quantities. The company's product lines include Nano DeBEE, Micro DeBEE, and Mini DeBEE. BEE International uses a proprietary homogenization technology in each of its product lines, which allows independent control of pressure, flow, cavitation, impact, shear, and process duration. This technology uses pressure of up to 45,000 psi to force the product through a small nozzle of less than 0.2 mm, which causes cavitation and creates a high-velocity jet stream that is flowed through an absorption cell. Within the absorption cell, there are various mechanisms for fluid-on-fluid impact, though the company prides itself on not using grinding or mixing action.

**Bertin Technologies, a company of CNIM Group ([www.bertin.fr/en/](http://www.bertin.fr/en/))**

Bertin Technologies is a French company with U.S. offices in Rockville, Maryland. The company has supplied highly technological laboratory and industrial equipment since 1956. Its customers are primarily in the fields of energy, the environment, pharmaceuticals and life sciences, defense, aerospace, industry, and services. Bertin's product line includes Hovereye-EX, a brand of tactical, vertical takeoff and landing, unmanned aerial vehicle (UAV); Vigisight® for real-time image processing; Suzon®, a wireless network of unattended sensors; Coriolis Micro, a biological air sampler; and Precellys, a biological sample grinder. The Precellys product is where Bertin may compete with Pressure BioSciences. Precellys ([www.precellys.com](http://www.precellys.com)) is a laboratory apparatus for preparing high-performance biological samples that has already sold over 1,000 units (Source: Bertin Technologies). The company reports that Precellys can release and extract DNA or RNA molecules or proteins in less than 30 seconds, and offers grinding, homogenization, and lysis for a wide range of sample types. It also includes Cryolys, which is Bertin's patented cooling option, to keep stable temperatures during homogenization of approximately 4°C.

**BioSpec Products, Inc. ([www.biospec.com](http://www.biospec.com))**

BioSpec Products, located in Bartlesville, Oklahoma, has been designing and selling scientific equipment for over 30 years. The company has built a business supplying basic laboratory instruments online through its website as well as through global distributors. BioSpec Products states on its website that its product sales are driven by word-of-mouth and methods referenced in scientific publications, rather than by advertising investments. The company's distributors reach across the U.S., and many major distributors include BioSpec products in their catalogs. As well, through a global network of distributors, BioSpec products are available in over 20 countries, including the United Kingdom, China, Brazil, and India. BioSpec's product categories consist of the following: homogenizers and cell disrupters, pulverizers, tissue grinders, beads (guide-lines), tissue dispersers, blenders, mixers and stirrers, vials, rotators, coldblocks, and microcentrifuges.

**GEA Niro Soavi | GEA Group ([www.niro-soavi.com](http://www.niro-soavi.com))**

Italian company GEA Niro Soavi was founded in 1947 to produce butter making machines, which led the company to later make high-pressure homogenizers for the dairy industry. Today, GEA Niro makes over 600 machines per year, specializing in high-pressure homogenizers and plunger pumps that are sold worldwide. GEA Niro is part of the GEA Group's (Global Engineering Alliance [G1A-Frankfurt]) GEA Mechanical Equipment division, which focuses on engineering, manufacturing, and sales of specialized process components for several industrial applications. The company offers a wide range of homogenizer designs providing throughputs from 100 liters per hour (l/h) to 50,000 l/h at continuous working pressures from 100 bar up to 1,500 bar (or over 21,700 psi). Custom engineered solutions are also available. GEA Niro's products today are no longer focused on just the dairy industry. The company offers a number of application-specific technologies and instruments, such as a High Efficiency NanoVALVE™, an aseptic homogenizer to maintain sterility in pharma/biotech samples, and the OpenXFLO™

compression block design designed to comply with specialized pumping requirements for latex and polymers nanodispersions, slurries, polymerizing and shear sensitive fluids, and products with high fibers and solids content. As a result, GEA Niro's equipment is used in the food and beverage, pharma/biotech, chemical, cosmetics, and many other industries.

**IKA Group ([www.ikaprocess.com](http://www.ikaprocess.com))**

IKA offers an extensive range of innovative equipment for applications in research and development. Leading entities employ the company's technology for their mixing, heating, distilling, and crushing applications. IKA has gained a prominent position in the world market with its magnetic stirrers, mixers, overhead stirrers, shakers, homogenizers, mills, rotary evaporators, calorimeters, laboratory reactors, and specially developed software for laboratory and analysis applications. The company's Process Technology division offers solutions and state-of-the-art manufacturing options, which include: dispersing machines, homogenizers, stirrers, jet flow agitators, kneading machines, vacuum dryers, and ready-for-use process plants. Certain applications in the food, pharmaceutical, cosmetic, biotechnology, and environmental industries require particles to reach sizes in a nano range, which can be accomplished with high-pressure homogenizers. IKA's high-pressure homogenizer, HPH, is used for the homogenizing of dairy products, disintegrating cellulose in vegetables, producing gel systems in the pharmaceutical industry, and disintegrating cells in the biotechnology. The company has headquarters in Wilmington, North Carolina and employs roughly 800 employees at eight locations on four continents, with customers such as BASF, Bayer, and Procter & Gamble.

**Microfluidics | a unit of IDEX Corp. ([www.microfluidicscorp.com](http://www.microfluidicscorp.com))**

Microfluidics has headquarters in Massachusetts and the United Kingdom. The business is part of specialty engineering company, IDEX Corp. (IEX-NYSE). Microfluidics supplies a brand of high-shear fluid processors known as Microfluidizer®. Versus conventional homogenizers, the company believes that is Microfluidizer® high-shear fluid processors offer benefits in terms of generating a uniform shear field for particle size reduction and cell disruption, and being able to do so with repeatable and scalable results—which Microfluidics does not view as possible with today's homogenizers. Microfluidics' technology platform has given rise to a number of high-pressure (up to 30,000 psi) homogenizer machines, from plug and play to low volume to pneumatic, as well as pilot and production scale instruments designed to reduce time to market and enhance process efficiency. Microfluidics also emphasizes its bottom-up nanoparticle creation possible with its PureNano Continuous Crystallizer. The company reports that it has installed more than 3,000 processors worldwide, which includes at 17 major pharmaceutical companies and across the chemical, cosmetic, nutraceutical, food, and energy industries.

**MO BIO Laboratories, Inc. ([www.mobio.com](http://www.mobio.com))**

California-headquartered MO BIO Laboratories has been supplying molecular biology tools since 1993. The company is an international producer, researcher, and distributor of life science laboratory instruments. Among other products, MO BIO sells PowerMAX® DNA Isolation kits and RNA PowerSoil® kits, which perform soil and microbial isolation for environmental and microbiology researchers, as well as a PowerFecal™ DNA Isolation Kit, which isolates DNA from stool, gut material, and biosolids. The company believes that its DNA isolation and purification kits are among the fastest available, which include its UltraClean® 6 Minute Plasmid Mini Prep, the UltraClean® PCR Clean-up Kit (only 3 minutes per reaction), and UltraClean® GelSpin® Kit (only 5 minutes per reaction). MO BIO's PowerLyzer™ 24 Bench Top Bead-Based Homogenizer provides lysis and homogenization of any biological sample using high-speed mechanical action, horizontal tube positioning, and optimized bead-beating technology to process up to 24 samples in 2 ml tubes in as little as 30 seconds. It accommodates all sample types, including difficult samples such as pine needles, seeds, spores, fungal mats, bone, and skin. In addition to homogenization and DNA/RNA isolation equipment, MO BIO's product categories further include protein extraction, high-throughput purification, dye dots, DNA clean-up, plasmid DNA isolation, DNA-free reagents, growth media, plastics and spin filters, lab supplies, certified water, enzymes, and samples.



**Omni International ([www.omni-inc.com](http://www.omni-inc.com))**

Omni International, which markets itself as “the homogenizer company,” designs and produces a range of laboratory homogenizer technology. The company maintains an “Applications” database designed to help customers find the best solution, quickly. The database consists of customer published applications, sorted into useful categories, complete with product recommendations to identify a preferred laboratory homogenizer for any sample type. The company’s products include high-shear and multi-sample laboratory homogenizers, ultrasonic cell disruptors, bead mills, and tissue grinders, as well as a variety of disposable, accessory, and replacement parts.

**Silverson Machines Inc. ([www.silverson.com](http://www.silverson.com))**

Silverson Machines is a global company with sales offices, distributors, and agents in over 50 countries around the world. For more than 60 years, the company has specialized in the design and construction of high-shear mixers for processing and manufacturing industries, including food, pharmaceutical, chemical, and cosmetics and toiletries markets. Silverson offers free on-site trials of its products and has test facilities in the U.S. and UK that offer laboratory and production-scale machines for clients to test in confidence and in collaboration with Silverson’s engineers. The company’s customers have included DuPont, Johnson and Johnson, Novartis, Coca-Cola, Proctor and Gamble, and Heinz, among many others. Silverson’s industrial mixing products comprise laboratory mixers, laboratory in-line mixers, batch mixers, Ultramix mixers, in-line mixers, in-line ultra-sanitary mixers, Silverson mixer homogenizers, Flashblend powder/liquid mixers, Flashmix powder/liquid mixers, bottom entry mixers, sanitary mixers, and disintegrator/dissolvers.

**Stansted Fluid Power Ltd. ([www.homogenizersystems.com](http://www.homogenizersystems.com))**

British company, Stansted Fluid Power, has been producing ultra-high-pressure industrial fluid processing equipment for over 40 years. The company states that its line of laboratory, pilot, and production scale homogenizers each offer the following attributes: process pressure of 60,000 psi or 410 MPa to yield among the smallest particle sizes in emulsions and suspensions, constant pressure through pneumatically controlled valves to yield among the narrowest particle size distributions, ease of use and safety, durability, scalability, and serviceability. Typical applications for Stansted’s homogenizers include particle size reduction for emulsions and dispersions, cell disruption/lysis, encapsulation, solid lipid nanoparticles, liposome formulations, structural modification, and preservation.

## Key Points

- Pressure BioSciences, Inc. is focused on the development and commercialization of proprietary laboratory instruments and consumable products.
- The Company's product portfolio is based on its patented pressure cycling technology (PCT), which is able to create and release high levels of hydrostatic pressure in a safe and controlled manner. Pressure BioSciences has 24 issued and 6 pending patents globally that protect various aspects and applications of PCT.
- The PCT platform is a highly versatile and adaptable technology that can be used across many different industries, including the life sciences, pharmaceutical, biotechnology, forensics, food, anti-bioterror, agriculture, automotive, and oil and gas industries, among others. While Pressure BioSciences believes that its products have potential in each of these markets, the Company is currently focusing its product development efforts on biological sample preparation for life sciences research.
  - The sample preparation market for life sciences research could reach \$5.4 billion in 2013, with potential to more than double to \$11.5 billion by 2018, driven by new technologies and growing demand.
- Pressure BioSciences' line of bench-top Barocycler® laboratory instruments employ high pressure to prepare samples for downstream analysis and testing. These PCT-based systems have been shown to extract higher quality and quantities of proteins, nucleic acids (DNA/RNA), and small molecules from samples versus competitive products, which could have positive implications in biomarker discovery, forensics, and many other areas of life science research.
  - The Company has installed 235 Barocycler units to date. Customers include large pharmaceutical and biotechnology companies (e.g., Bristol-Myers, Amgen, and Merck), U.S. government agencies (FBI, FDA, NIH, USDA), and universities (Harvard Medical School, Stanford, and UCLA).
  - Pressure BioSciences' next-generation PCT platform is the HUB family of Barocyclers. The goal of the HUB is to create a modular system that can grow and adapt to a laboratory's needs, with users purchasing accessories as needed to expand the functionality of their HUB unit.
- Pressure BioSciences has gained exposure to corporate, government, and university laboratories through its strategic collaboration program, which allows third-party researchers to temporarily install a Barocycler instrument in their laboratory in exchange for collecting independent, third-party data on a certain subject.
  - Data collected through this program has helped validate existing applications for PCT and establish potential in new applications. As well, much of this research has resulted in peer-reviewed publications and presentations at scientific meetings. To date, over 100 papers have been published on PCT.
  - The program also exposes the technology to potential customers as well as specific research scientists who the Company believes could have a positive impact on market acceptance of PCT.
- Pressure BioSciences' portfolio also includes Shredder instruments (for rapidly shredding or grinding samples), sample preparation kits and reagents, and related consumable products (e.g., sample tubes).
- Pressure BioSciences has multiple novel products in its R&D pipeline, including a high-throughput PCT system designed to meet the needs of larger research and clinical diagnostics laboratories and companies. The Company has achieved proof-of-principle for this system and expects to complete beta testing by early 2014.
- At June 30, 2013, the Company's cash position was \$113,000.

## Historical Financial Results

Figures 32, 33, and 34 summarize Pressure BioSciences' key historical financial statements: its condensed consolidated Statements of Operations, Balance Sheets, and Statements of Cash Flows, as presented in the Company's Form 10-Q filed with the U.S. Securities and Exchange Commission (SEC) on August 15, 2013.

Figure 32  
Pressure BioSciences, Inc. and Subsidiary  
CONDENSED CONSOLIDATED STATEMENTS OF OPERATIONS (UNAUDITED)

	For the Three Months Ended		For the Six Months Ended	
	June 30,		June 30,	
	2013	2012	2013	2012
Revenue:				
PCT products, services, other	\$ 196,522	\$ 224,384	\$ 418,092	\$ 389,156
Grant revenue	161,214	100,524	310,382	241,413
Total revenue	<u>357,736</u>	<u>324,908</u>	<u>728,474</u>	<u>630,569</u>
Costs and expenses:				
Cost of PCT products and services	83,829	109,203	188,373	187,397
Research and development	260,408	256,307	506,866	527,918
Selling and marketing	188,392	168,173	382,292	406,265
General and administrative	683,133	474,015	1,233,992	1,156,361
Total operating costs and expenses	<u>1,215,762</u>	<u>1,007,698</u>	<u>2,311,523</u>	<u>2,277,941</u>
Operating loss	(858,026)	(682,790)	(1,583,049)	(1,647,372)
Other (expense) income:				
Interest (expense) income	(63,110)	(4,214)	(72,013)	(60,527)
Other (expense)	(208,709)	—	(208,709)	—
Change in fair value of derivative liability	63,804	177,312	18,359	135,300
Total other (expense) income	<u>(208,015)</u>	<u>173,098</u>	<u>(262,363)</u>	<u>74,773</u>
Net loss	<u>(1,066,041)</u>	<u>(509,692)</u>	<u>(1,845,410)</u>	<u>(1,572,599)</u>
Accrued and deemed dividends on convertible preferred stock	(107,871)	(579,974)	(722,592)	(604,388)
Net loss applicable to common shareholders	<u>\$ (1,173,912)</u>	<u>\$ (1,089,666)</u>	<u>\$ (2,568,002)</u>	<u>\$ (2,176,987)</u>
Net loss per share attributable to common stockholders - basic and diluted	\$ (0.10)	\$ (0.11)	\$ (0.22)	\$ (0.21)
Weighted average common stock shares outstanding used in the basic and diluted net loss per share calculation	11,521,794	10,197,850	11,883,797	10,352,461

Source: Pressure BioSciences, Inc.

Figure 33  
Pressure BioSciences, Inc. and Subsidiary  
CONDENSED CONSOLIDATED BALANCE SHEETS

	June 30, 2013 (Unaudited)	December 31, 2012 (Audited)
<b>ASSETS</b>		
<b>CURRENT ASSETS</b>		
Cash and cash equivalents	\$ 113,663	\$ 1,461
Accounts receivable	314,157	216,265
Inventories, net of \$50,000 reserve at June 30, 2013 and December 31, 2012	866,639	923,362
Prepaid income taxes	7,381	7,381
Prepaid expenses and other current assets	53,380	83,435
Total current assets	<u>1,355,220</u>	<u>1,231,904</u>
<b>PROPERTY AND EQUIPMENT, NET</b>		
Deposits	—	6,472
Intangible assets, net	60,814	85,130
<b>TOTAL ASSETS</b>	<u>\$ 1,431,744</u>	<u>\$ 1,353,788</u>
<b>LIABILITIES AND STOCKHOLDERS' DEFICIT</b>		
<b>CURRENT LIABILITIES</b>		
Accounts payable		
Accrued employee compensation	\$ 1,083,249	\$ 1,199,846
Accrued professional fees and other	128,506	119,338
Deferred revenue	429,018	267,936
Promissory note	19,352	46,466
Dividend liability	75,000	75,000
Related party debt	—	60,000
Convertible debt, net of debt discount of \$436,171 and \$0, respectively	26,697	98,675
Conversion option liability	93,949	863,004
Warrant derivative liability	497,501	—
Total current liabilities	<u>284,876</u>	<u>160,812</u>
<b>LONG-TERM LIABILITIES</b>		
Convertible debt – long term	<u>2,638,148</u>	<u>2,891,077</u>
Deferred revenue	12,384	—
<b>TOTAL LIABILITIES</b>	<u>—</u>	<u>2,487</u>
<b>COMMITMENTS AND CONTINGENCIES (Note 4*)</b>		
	<u>2,650,532</u>	<u>2,893,564</u>
<b>STOCKHOLDERS' DEFICIT</b>		
Series D convertible preferred stock, \$.01 par value; 850 shares authorized; 300 shares issued and outstanding on June 30, 2013 and on December 31, 2012	3	3
Series G convertible preferred stock, \$.01 par value; 240,000 shares authorized; 145,320 shares issued and outstanding on June 30, 2013 and on December 31, 2012	1,453	1,453
Series H convertible preferred stock, \$.01 par value; 10,000 shares authorized; 10,000 shares and 0 shares issued and outstanding on June 30, 2013 and December 31, 2012, respectively	100	—
Series J convertible preferred stock, \$.01 par value; 6,250 shares authorized; 5,087.5 shares and 0 shares issued and outstanding on June 30, 2013 and on December 31, 2012	51	—
Common stock, \$.01 par value; 50,000,000 shares authorized; 11,449,267 and 12,149,267 shares issued and outstanding on June 30, 2013 and on December 31, 2012	114,493	121,493
Warrants to acquire preferred stock and common stock	3,901,306	3,015,996
Additional paid-in capital	17,951,348	15,940,818
Accumulated deficit	(23,187,542)	(20,619,539)
Total stockholders' deficit	<u>(1,218,788)</u>	<u>(1,539,776)</u>
<b>TOTAL LIABILITIES AND STOCKHOLDERS' DEFICIT</b>	<u>\$ 1,431,744</u>	<u>\$ 1,353,788</u>

\* Notes are included in the Company's Form 10-Q filed on August 15, 2013, with the SEC.

Source: Pressure BioSciences, Inc.

Figure 34  
Pressure BioSciences, Inc. and Subsidiary  
CONDENSED CONSOLIDATED STATEMENTS OF CASH FLOWS (UNAUDITED)

	For the Six Months Ended	
	June 30,	
	2013	2012
<b>CASH FLOWS FROM OPERATING ACTIVITIES:</b>		
Net loss	\$ (1,845,410)	\$ (1,572,599)
Adjustments to reconcile net loss to net cash used in operating activities:		
Depreciation and amortization	40,421	57,168
Accretion of interest and amortization of debt issuance costs	260,753	46,159
Stock-based compensation expense	28,017	20,090
Amortization of third-party fees paid in restricted common stock	256,000	79,575
Change in fair value of derivative liability	(18,359)	(135,300)
Changes in operating assets and liabilities:		
Accounts receivable	(97,892)	27,511
Inventories	56,722	93,116
Accounts payable	(116,597)	304,891
Accrued employee compensation	9,168	2,260
Deferred revenue	(29,601)	—
Other accrued expenses and liabilities	160,167	50,520
Prepaid expenses and other current assets	(19,973)	(157,639)
Net cash used in operating activities	<u>(1,316,584)</u>	<u>(1,184,248)</u>
<b>CASH FLOWS FROM INVESTING ACTIVITIES:</b>		
Purchases of property, plant, and equipment	(1,531)	—
Net cash used in investing activities	<u>(1,531)</u>	<u>—</u>
<b>CASH FLOWS FROM FINANCING ACTIVITIES:</b>		
Payment of related party debt	(46,778)	—
Net Proceeds from convertible debt	640,500	—
Payments on convertible debt	—	(43,000)
Payment of dividends	(60,000)	—
Funds held for private placement	—	295,000
Net proceeds from the issuance of equity securities	896,595	730,389
Net cash provided by financing activities	<u>1,430,317</u>	<u>982,389</u>
Change in cash and cash equivalents	112,202	(201,859)
Cash and cash equivalents, beginning of period	1,461	222,775
Cash and cash equivalents, end of period	<u>\$ 113,663</u>	<u>\$ 20,916</u>
<b>SUPPLEMENTAL INFORMATION:</b>		
Non-investing and financing activities		
Income taxes paid in cash	—	1,900
Issuance of common stock dividends on preferred stock	—	161,557
Convertible debt exchanged for Series J convertible preferred stock	1,018,000	—
Issuance of common stock for services	120,000	155,575
Convertible debt exchanged for common stock	—	387,547
Deemed dividend on Series J convertible preferred stock	651,182	—

Source: Pressure BioSciences, Inc.

## Risks and Disclosures

This Executive Informational Overview® (EIO) has been prepared by Pressure BioSciences, Inc. (“Pressure BioSciences” or “the Company”) with the assistance of Crystal Research Associates, LLC (“CRA”) based upon information provided by the Company. CRA has not independently verified such information. Some of the information in this EIO relates to future events or future business and financial performance. Such statements constitute forward-looking information within the meaning of the Private Securities Litigation Act of 1995. Such statements can only be predictions and the actual events or results may differ from those discussed due to the risks described in Pressure BioSciences’ statements on Forms 10-K, 10-Q, and 8-K, as well as other forms filed from time to time.

The content of this report with respect to Pressure BioSciences has been compiled primarily from information available to the public released by the Company through news releases, Annual Reports, and U.S. Securities and Exchange Commission (SEC) filings. Pressure BioSciences is solely responsible for the accuracy of this information. Information as to other companies has been prepared from publicly available information and has not been independently verified by Pressure BioSciences or CRA. Certain summaries of activities and outcomes have been condensed to aid the reader in gaining a general understanding. CRA assumes no responsibility to update the information contained in this report. In addition, CRA has been compensated by the Company in cash of thirty-nine thousand, five hundred U.S. dollars and one hundred thousand warrants for its services in creating this report and for updates. For more complete information about the risks involved in an investment in the Company, please see Pressure BioSciences’ most recently filed Annual Report on Form 10-K for the year ended December 31, 2012.

Investors should carefully consider risks and information about Pressure BioSciences’ business. Investors should not interpret the order in which considerations are presented in this or other filings as an indication of their relative importance. The risks and uncertainties overviewed in Pressure BioSciences’ Form 10-K are not the only risks that the Company faces. Additional risks and uncertainties not presently known to Pressure BioSciences or that it currently believes to be immaterial may also adversely affect its business. If any of such risks and uncertainties develops into an actual event, Pressure BioSciences’ business, financial condition, and results of operations could be materially adversely affected, and the trading price of the Company’s shares could decline.

This report is published solely for information purposes and is not to be construed as an offer to sell or the solicitation of an offer to buy any security in any state. Past performance does not guarantee future performance. Additional information about Pressure BioSciences and its public filings, as well as copies of this report, can be obtained by calling (508) 230-1828.

### Going Concern

Pressure BioSciences has experienced negative cash flows from operations with respect to its PCT business since inception. As of December 31, 2012, the Company did not have adequate working capital resources to satisfy its current liabilities. Based on its current projections, Pressure BioSciences will need to raise capital before the end of 2013.

As a result, the audit report issued by Pressure BioSciences’ independent registered public accounting firm on its audited consolidated financial statements for the fiscal year ended December 31, 2012, contains an explanatory paragraph regarding the Company’s ability to continue as a going concern. The audit report issued by Pressure BioSciences’ independent registered public accounting firm for the Company’s financial statements for the fiscal year ended December 31, 2012, states that Pressure BioSciences’ auditing firm has substantial doubt in its ability to continue as a going concern due to the risk that the Company may not have sufficient cash and liquid assets to cover its operating and capital requirements for the next 12-month period; and, if sufficient cash cannot be obtained, Pressure BioSciences would have to substantially alter, or possibly even discontinue, operations.

The conditions described above could adversely affect Pressure BioSciences’ ability to obtain additional financing on favorable terms, if at all, and may cause investors to have reservations about its long-term prospects, and may adversely affect the Company’s relationships with its customers. There can be no assurance that Pressure

BioSciences' auditing firm will not qualify its opinion in the future. If the Company cannot successfully continue as a going concern, Pressure BioSciences' stockholders may lose their entire investment in the Company.

Pressure BioSciences' management has developed a plan to continue operations. This plan includes reducing expenses, streamlining operations, and obtaining capital through an equity and/or debt financing. Although the Company has successfully completed equity financings and reduced expenses in the past, it cannot assure its investors that plans to address these matters in the future will be successful. Additional financing may not be available to the Company on a timely basis or on terms acceptable to it, if at all. In the event Pressure BioSciences is unable to raise sufficient funds on terms acceptable to it, the Company may be required to:

- severely limit or cease operations or otherwise reduce planned expenditures and forego other business opportunities, which could harm its business;
- obtain financing with terms that may have the effect of diluting or adversely affecting the holdings or the rights of the holders of the Company's capital stock; or
- obtain funds through arrangements with future collaboration partners or others that may require Pressure BioSciences to relinquish rights to some or all of its technologies or products.

Pressure BioSciences has experienced negative cash flows from operations from its PCT business since commencing its PCT operations. As of June 30, 2013, Pressure BioSciences' cash position was \$113,000 which, based on current projections, will not be sufficient to fund operations until the end of 2013. The Company therefore requires substantial additional capital to fund operations beyond 2013.

**The Company's business could be adversely affected if it fails to implement and maintain effective disclosure controls and procedures and internal control over financial reporting.**

Pressure BioSciences concluded that, as of December 31, 2012, its disclosure controls and procedures and internal control over financial reporting was not effective. The Company determined that it has limited resources for adequate personnel to prepare and file reports under the Securities Exchange Act of 1934 within the required time periods and that material weaknesses in its internal control over financial reporting exist relating to accounting for complex equity transactions. If the Company is unable to implement and maintain effective disclosure controls and procedures and remediate the material weaknesses in a timely manner, or if Pressure BioSciences identifies other material weaknesses in the future, its ability to produce accurate and timely financial statements and public reports could be impaired, which could adversely affect its business and financial condition. The Company has identified a lack of sufficient segregation of duties. Specifically, this material weakness is such that the design over these areas relies primarily on detective controls and could be strengthened by adding preventive controls to properly safeguard assets. In addition, investors may lose confidence in the Company's reported information and the market price of the Company's common stock may decline.

The Company will need a greater amount of additional capital than it currently expects if it experiences unforeseen costs or expenses, unanticipated liabilities or delays in implementing its business plan, developing its products, and achieving commercial sales. Pressure BioSciences needs substantial capital to implement its sales distribution strategy for its current products and to develop and commercialize future products using its PCT products and services in the sample preparation area, as well as for applications in other areas of life sciences. The Company's capital requirements will depend on many factors, including but not limited to the following:

- the problems, delays, expenses, and complications frequently encountered by early-stage companies;
- market acceptance of the Company's PCT products and services for sample preparation;
- the success of its sales and marketing programs; and
- changes in economic, regulatory, or competitive conditions in the markets the Company intends to serve.

To satisfy the Company's potential capital requirements to cover the cost of implementing its sales distribution strategy for Pressure BioSciences' current products and services and to develop and commercialize future products and services using the Company's PCT for sample preparation and other life science applications, Pressure BioSciences need to raise additional funds in the public or private capital markets. The Company may seek to raise any necessary additional funds through the issuance of warrants, equity or debt financings, or executing collaborative arrangements with corporate partners or other sources, which may be dilutive to existing stockholders or otherwise have a material effect on the Company's current or future business prospects. Additional financing may not be available to Pressure BioSciences on a timely basis, if at all, or on terms acceptable to it. If adequate funds are not available or if the Company fails to obtain acceptable additional financing, it may be required to do the following:

- severely limit or cease operations or otherwise reduce planned expenditures and forego other business opportunities, which could harm the Company's business;
- obtain financing with terms that may have the effect of substantially diluting or adversely affecting the holdings or the rights of the holders of the Company's capital stock; or
- obtain funds through arrangements with future collaboration partners or others that may require the Company to relinquish rights to some or all of its technologies or products.

Actual results and performance could be adversely affected by the current economic conditions in the global economy, which continue to pose a risk to the overall demand for the Company's products from its customers who may elect to defer or cancel purchases of, or decide not to purchase products in response to continuing tightness in the credit markets, negative financial news, and general uncertainty in the economy. In addition, Pressure BioSciences' ability to obtain additional financing, on acceptable terms, if at all, may be adversely affected by the uncertainty in the current economic climate.

**The Company has a history of operating losses, anticipates future losses, and may never be profitable.**

Pressure BioSciences has experienced significant operating losses in the area of PCT in each period since the Company began investing resources in PCT. These losses have resulted principally from research and development, sales and marketing, and general and administrative expenses associated with the development of the PCT business. During the year ended December 31, 2012, the Company recorded a net loss applicable to common shareholders of \$4,400,215, or (\$0.44) per share, as compared with \$5,107,661, or (\$0.77) per share, of the corresponding period in 2011. Pressure BioSciences expects to continue to incur operating losses until sales of its PCT products increase substantially. The Company cannot be certain when, if ever, it will become profitable. Even if it were to become profitable, it might not be able to sustain such profitability on a quarterly or annual basis.

**The Company's financial results depend on revenues from its PCT products and services, and from government grants.**

Pressure BioSciences currently relies on revenues from its PCT products and services in the sample preparation area and from revenues derived from grants awarded to it by governmental agencies, such as the National Institutes of Health. The Company has been unable to achieve market acceptance of its product offerings to the extent necessary to achieve significant revenue. Competition for government grants is very intense, and the Company can provide no assurance that it will continue to be awarded grants in the future. If Pressure BioSciences is unable to increase revenues from sales of its PCT products and services and government grants, its business will likely fail.

**The Company may be unable to obtain market acceptance of its PCT products and services.**

Many of the Company's initial sales of its PCT products and services have been to its collaborators, following their use of its products in studies undertaken in sample preparation for genomics, proteomics, and small molecules studies. The Company's technology requires scientists and researchers to adopt a method of sample extraction that is different than existing techniques. The PCT sample preparation system is also more costly than existing



techniques. Pressure BioSciences' ability to obtain market acceptance will depend, in part, on its ability to demonstrate to its potential customers that the benefits and advantages of its technology outweigh the increased cost of its technology compared with existing methods of sample extraction. If the Company is unable to demonstrate the benefits and advantages of its products and technology as compared with existing technologies, it will not gain market acceptance and its business will likely fail.

**Pressure BioSciences' business may be harmed if it encounters problems, delays, expenses, and complications that often affect companies that have not achieved significant market acceptance.**

The Company's PCT business continues to face challenges in achieving market acceptance, including the following:

- availability of adequate financing;
- unanticipated problems and costs relating to the development, testing, production, marketing, and sale of its products;
- delays and costs associated with its ability to attract and retain key personnel; and
- competition.

The sales cycle of the Company's PCT products is lengthy. Pressure BioSciences has incurred and may continue to incur significant expenses and the Company may not generate any significant revenue related to those products. Many of the Company's current and potential customers have required between three and six months or more to test and evaluate its PCT products. This increases the possibility that a customer may decide to cancel its order or otherwise change its plans, which could reduce or eliminate the sales to that potential customer. As a result of this lengthy sales cycle, Pressure BioSciences have incurred and may continue to incur significant research and development, selling and marketing, and general and administrative expenses related to customers from whom the Company has not yet generated any revenue from its products, and from whom the Company may never generate the anticipated revenue if a customer is not satisfied with the results of the evaluation of its products or if a customer cancels or changes its plans.

**The Company's business could be harmed if its products contain undetected errors or defects.**

Pressure BioSciences is continuously developing new and improving its existing PCT products in sample preparation and expects to do so in other areas of life sciences depending upon the availability of resources. Newly introduced products can contain undetected errors or defects. In addition, these products may not meet their performance specifications under all conditions or for all applications. If, despite internal testing and testing by the Company's collaborators, any of its products contain errors or defects, or fails to meet customer specifications, then the Company may be required to enhance or improve those products or technologies. Pressure BioSciences may not be able to do so on a timely basis, if at all, and may only be able to do so at considerable expense. In addition, any significant reliability problems could result in adverse customer reaction, negative publicity, or legal claims and could harm its business and prospects.

**Pressure BioSciences' success may depend on its ability to manage growth effectively.**

Failure to manage growth effectively could harm its business and prospects. Given the Company's limited resources and personnel, growth of its business could place significant strain on Company management, information technology systems, sources of manufacturing capacity, and other resources. To properly manage its growth, Pressure BioSciences may need to hire additional employees and identify new sources of manufacturing capabilities. Failure to effectively manage this growth could make it difficult to manufacture products and fill orders, as well as lead to declines in product quality or increased costs—any of which would adversely impact its business and results of operations.

**The Company's success is substantially dependent on the continued service of its senior management.**

Pressure BioSciences' success is substantially dependent on the continued service of its senior management. The Company does not have long-term employment agreements with its key employees. In September 2013, the Company's chief financial officer left to pursue other opportunities. While he has been replaced with an acting chief executive officer, the loss of the services of any of its senior management has made and could make it more difficult to successfully operate the Company's business and achieve its business goals. In addition, failure to retain existing engineering, research and development, and sales personnel could harm its product development capabilities and customer and employee relationships, delay the growth of sales of its products, and could result in the loss of key information, expertise, or know-how.

The Company may not be able to hire or retain the number of qualified personnel, particularly engineering and sales personnel, required for its business, which would harm the development and sales of its products and limit its ability to grow.

Competition in this industry for senior management, technical, sales, marketing, finance, and other key personnel is intense. If Pressure BioSciences is unable to retain its existing personnel, or attract and train additional qualified personnel, either because of competition in its industry for such personnel or because of insufficient financial resources, the Company's growth may be limited. Success also depends, in particular, on Pressure BioSciences' ability to identify, hire, train, and retain qualified engineering and sales personnel with experience in design, development, and sales of laboratory equipment.

**The Company's reliance on a single third party for all of its manufacturing, and certain of its engineering and other related services could harm its business.**

Pressure BioSciences currently relies on Source Scientific, LLC, a third-party contract manufacturer, to manufacture its PCT instrumentation, provide engineering expertise, and manage the majority of its sub-contractor supplier relationships. Because of its dependence on one manufacturer, the Company's success will depend, in part, on the ability of Source Scientific to manufacture its products cost effectively and in sufficient quantities to meet the Company's customer demands—if and when such demand occurs—and meeting its quality requirements. If Source Scientific experiences manufacturing problems or delays, or if Source Scientific decides not to continue to provide Pressure BioSciences with these services, its business may be harmed. While the Company believes other contract manufacturers are available to address its manufacturing and engineering needs, if the Company find it necessary to replace Source Scientific, there will be a disruption in its business and Pressure BioSciences would incur additional costs and delays that would harm its business.

**Failure to manage current or future alliances or joint ventures effectively may harm the Company's business.**

Pressure BioSciences has entered into business relationships with 11 distribution partners and one co-marketing partner, and may enter into additional alliances, joint ventures, or other business relationships to further develop, market, and sell its PCT product line. The Company may not be able to accomplish the following:

- identify appropriate candidates for alliances, joint ventures, or other business relationships;
- assure that any candidate for an alliance, joint venture, or business relationship will provide it with the support anticipated;
- successfully negotiate an alliance, joint venture, or business relationship on terms that are beneficial to it; or
- successfully manage any alliance or joint venture.

Furthermore, any alliance, joint venture, or other business relationship may divert management's time and resources. Entering into a disadvantageous alliance, joint venture, or business relationship, failing to manage an alliance, joint venture, or business relationship effectively, or failing to comply with any obligations in connection therewith, could harm the Company's business and prospects.

**Pressure BioSciences may not be successful in growing its international sales.**

The Company cannot guarantee that it will successfully develop its international sales channels to enable it to generate significant revenue from international sales. Pressure BioSciences currently has international distribution agreements that cover 24 countries in Europe, Asia, and Australia. It has generated limited sales to date from international sales and cannot guarantee that it will be able to increase sales. As the Company expands, its international operations may be subject to numerous risks and challenges, including the following:

- multiple conflicting and changing governmental laws and regulations, including those that regulate high-pressure equipment;
- reduced protection for intellectual property (IP) rights in some countries;
- protectionist laws and business practices that favor local companies;
- political and economic changes and disruptions;
- export and import controls;
- tariff regulations; and
- currency fluctuations.

**The Company's operating results are subject to quarterly variation. Its operating results may fluctuate significantly from period to period depending on a variety of factors, including but not limited to the following:**

- its ability to increase sales of its PCT products for sample preparation on a consistent quarterly or annual basis;
- the lengthy sales cycle for its products;
- the product mix of the Barocyler instruments that are installed in a given period, and whether the installations are completed pursuant to sales, rental, or lease arrangements, and the average selling prices that the Company is able to command for its products;
- its ability to manage costs and expenses;
- its ability to continue research and development activities without incurring unexpected costs and expenses; and
- its ability to comply with state and federal regulations without incurring unexpected costs and expenses.

The Company's instrumentation operates at high pressures and may therefore become subject to certain regulation in the European Community. Regulation of high-pressure equipment may limit or hinder the Company's development and sale of future instrumentation.

Pressure BioSciences' Barocyler instruments operate at high pressures. If its Barocyler instruments exceed certain pressure levels, they may become subject to the European Pressure Equipment Directive, which requires certain pressure equipment meet certain quality and safety standards. The Company does not believe that it is subject to this directive because its Barocyler instruments are currently below the threshold documented in the text of the directive. If the Company's interpretation were to be challenged, it could incur significant costs defending the challenge, and could face production and selling delays—all of which could harm the business.

**The Company expects that it will be subject to regulation in the U.S., such as by the FDA, and overseas if and when it begins to invest more resources in the development and commercialization of PCT in applications outside of sample preparation for the research field.**

The Company's current PCT products in the area of sample preparation for the research field are not regulated by the FDA. Applications in which Pressure BioSciences intends to develop and commercialize PCT, such as protein purification, pathogen inactivation, and immunodiagnostics, are expected to require regulatory approvals or clearances from regulatory agencies, such as the FDA, prior to commercialization, when the Company expands its commercialization activities outside of the research field. The Company expects that obtaining these approvals or clearances will require a significant investment of time and capital resources and there can be no assurance that such investments will receive approvals or clearances that would allow it to commercialize the technology for these applications.

**If unable to protect its patents and other proprietary technology relating to the Company's PCT products, its business will be harmed.**

The ability to further develop and successfully commercialize the Company's products will depend, in part, on its ability to enforce patents, preserve trade secrets, and operate without infringing the proprietary rights of third parties. The Company currently has 14 U.S. and 10 foreign patents, expiring between 2015 and 2027. There can be no assurance that (a) any patent applications filed by the Company will result in issued patents; (b) patent protection will be secured for any particular technology; (c) any patents that have been or may be issued to the Company will be valid or enforceable; (d) any patents will provide meaningful protection to Pressure BioSciences; (e) others will not be able to design around its patents; and (f) patents will provide a competitive advantage or have commercial value. The failure to obtain adequate patent protection would have a material adverse effect on the Company and may adversely affect its ability to enter into, or affect the terms of, any arrangement for the marketing or sale of any product.

**The Company's patents may be challenged by others.**

Pressure BioSciences could incur substantial costs in patent proceedings, including interference proceedings before the U.S. Patent and Trademark Office, and comparable proceedings before similar agencies in other countries, in connection with any claims that may arise in the future. These proceedings could result in adverse decisions about the patentability of the Company's inventions and products, as well as about the enforceability, validity, or scope of protection afforded by the patents.

**If unable to maintain the confidentiality of the Company's trade secrets and proprietary knowledge, others may develop technology and products that could prevent the successful commercialization of Pressure BioSciences products.**

The Company relies on trade secrets and other unpatented proprietary information in its product development activities. To the extent it relies on trade secrets and unpatented know-how to maintain its competitive technological position, there can be no assurance that others may not independently develop the same or similar technologies. Pressure BioSciences seeks to protect its trade secrets and proprietary knowledge, in part, through confidentiality agreements with its employees, consultants, advisors, and contractors. These agreements may not be sufficient to effectively prevent disclosure of the Company's confidential information and may not provide it with an adequate remedy in the event of unauthorized disclosure of such information.

If the Company's employees, consultants, advisors, or contractors develop inventions or processes independently that may be applicable to its products, disputes may arise about ownership of proprietary rights to those inventions and processes. Such inventions and processes will not necessarily become the Company's property, but may remain the property of those persons or their employers. Protracted and costly litigation could be necessary to enforce and determine the scope of Pressure BioSciences' proprietary rights. Failure to obtain or maintain trade secret protection, for any reason, could harm the Company's business.

**If the Company infringes on the intellectual property rights of others, its business may be harmed.**

It is possible that the manufacture, use, or sale of the Company's PCT products or services may infringe patent or other IP rights of others. Pressure BioSciences may be unable to avoid infringement of the patent or other IP rights of others and may be required to seek a license, defend an infringement action, or challenge the validity of the patents or other IP rights in court. The Company may be unable to secure a license on terms and conditions acceptable to it, if at all. Also, the Company may not prevail in any patent or other IP rights litigation.

Patent or other IP rights litigation is costly and time-consuming, and there can be no assurance that the Company will have sufficient resources to bring any possible litigation related to such infringement to a successful conclusion. If Pressure BioSciences does not obtain a license under such patents or other IP rights, or if it is found liable for infringement, or if it is unsuccessful in having such patents declared invalid, the Company may be liable for significant monetary damages, may encounter significant delays in successfully commercializing and developing its PCT products, or may be precluded from participating in the manufacture, use, or sale of its PCT products or services requiring such licenses.

**The Company may be unable to adequately respond to rapid changes in technology and the development of new industry standards.**

The introduction of products and services embodying new technology and the emergence of new industry standards may render the Company's existing PCT products and related services obsolete and unmarketable if the Company is unable to adapt to change. Pressure BioSciences may be unable to allocate the funds necessary to improve its current products or introduce new products to address its customers' needs and respond to technological change. In the event that other companies develop more technologically advanced products, the Company's competitive position relative to such companies would be harmed.

**The Company may not be able to compete successfully with others that are developing or have developed competitive technologies and products.**

A number of companies have developed, or are expected to develop, products that compete or will compete with Pressure BioSciences' products. The Company competes with entities that have existing technologies for the extraction of nucleic acids, proteins, and small molecules from cells and tissues, including methods such as mortar and pestle, sonication, rotor-stator homogenization, French press, bead beating, freezer milling, enzymatic digestion, and chemical dissolution. There are additional companies pursuing new technologies with similar goals to the products developed or being developed by Pressure BioSciences. Some of the firms with which the Company competes, or may compete in the future, have or may have more extensive research, marketing, and manufacturing capabilities, more experience in genomics and proteomics sample preparation, protein purification, pathogen inactivation, immunodiagnostics, and DNA sequencing, and significantly greater technical, personnel, and financial resources than Pressure BioSciences, and may be better positioned to improve their technology to compete in an evolving industry. To compete, the Company must be able to demonstrate to potential customers that its products provide improved performance and capabilities. Failure to compete successfully could harm Pressure BioSciences' business and prospects.

**Provisions in the Company's articles of organization and bylaws may discourage or frustrate stockholders' attempts to remove or replace its current management.**

The Company's articles of organization and bylaws contain provisions that may make it more difficult or discourage changes in its management that the Company's stockholders may consider to be favorable. These provisions include the following items:

- a classified Board of Directors;
- advance notice for stockholder nominations to the Board of Directors;
- limitations on the ability of stockholders to remove directors; and
- a provision that allows a majority of the directors to fill vacancies on the Board of Directors.

These provisions could prevent or frustrate attempts to make changes in the Company's management that its stockholders consider to be beneficial and could limit the price that its stockholders might receive in the future for shares of the Company's common stock.

**The costs of compliance with the reporting obligations of the Exchange Act, the Sarbanes-Oxley Act of 2002, and the Dodd-Frank Wall Street Reform and Consumer Protection Act may place a strain on the Company's limited resources and its management's attention may be diverted from other business concerns.**

As a result of the regulatory requirements applicable to public companies, the Company incurs legal, accounting, and other expenses that are significant in relation to the size of the Company. In addition, the Sarbanes-Oxley Act of 2002 and the Dodd-Frank Wall Street Reform and Consumer Protection Act, as well as rules subsequently implemented by the SEC and OTC Markets Group, Inc., have required changes in corporate governance and financial disclosure practices of public companies, some of which are currently applicable to Pressure BioSciences and others will or may become applicable to it in the future. These rules and regulations have increased and will continue to increase the Company's legal and financial compliance costs and may make some activities more time-consuming. These requirements have placed and will continue to place a strain on the Company's systems and on its management and financial resources.

**Certain of the Company's net deferred tax assets could be substantially limited if Pressure BioSciences experiences an ownership change as defined in the Internal Revenue Code.**

Certain of the Company's net operating losses (NOLs) give rise to net deferred tax assets. Pressure BioSciences' ability to utilize NOLs and to offset its future taxable income and/or to recover previously paid taxes would be limited if it were to undergo an "ownership change" within the meaning of Section 382 of the Internal Revenue Code. In general, an ownership change occurs whenever the percentage of the stock of a corporation owned by 5% shareholders, within the meaning of Section 382 of the Code, increases by more than 50 percentage points over the lowest percentage of the stock of such corporation owned by such 5% shareholders at any time over the preceding three years.

An ownership change under Section 382 of the Code would establish an annual limitation on the amount of NOLs the Company could utilize to offset its taxable income in any single taxable year to an amount equal to (i) the product of a specified rate, which is published by the U.S. Treasury, and the aggregate value of the Company's outstanding stock plus; and (ii) the amount of unutilized limitation from prior years. The application of these limitations might prevent full utilization of the deferred tax assets attributable to its NOLs. Pressure BioSciences may have or will have experienced an ownership change as defined by Section 382 through the sale of equity and, therefore, will consider whether the sale of equity units will result in limitations of its net operating losses under Section 382 when the Company starts to generate taxable income. However, whether a change in ownership occurs in the future is largely outside of Pressure BioSciences' control, and there can be no assurance that such a change will not occur.

**RISKS RELATED TO SHARE OWNERSHIP**

**The holders of the Company's common stock could suffer substantial dilution due to its corporate financing practices.**

The holders of Pressure BioSciences' common stock could suffer substantial dilution due to its corporate financing practices, which, in the past few years, have included private placements and a registered direct offering. As of December 31, 2012, the Company has issued shares of Series A convertible preferred stock, Series B convertible preferred stock, Series C convertible preferred stock, Series D convertible preferred stock, Series E convertible preferred stock, and Series G convertible preferred stock. The Company has also authorized shares of Series H convertible preferred stock.

As of December 31, 2012, all of the shares of Series A convertible preferred stock, Series B convertible preferred stock, Series C convertible preferred stock, and Series E convertible preferred stock had been converted into shares of common stock. As of December 31, 2012, only shares of Series D convertible preferred stock and Series G convertible preferred stock were outstanding. As of March 31, 2013, there were also shares of Series J convertible

preferred stock outstanding. Further, in connection with those private placements and the Series D registered direct offering, the Company issued warrants to purchase common stock.

If all of the outstanding shares of Series D convertible preferred stock, Series G convertible preferred stock, and Series J convertible preferred stock were converted into shares of common stock and all outstanding warrants to purchase shares of common stock were exercised, each as of March 31, 2013, an additional 18,190,299 shares of common stock would be issued and outstanding. This additional issuance of shares of common stock would cause immediate and substantial dilution to the Company's existing stockholders and could cause a significant reduction in the market price of its common stock.

**Sales of a significant number of shares of the Company's common stock in the public market or the perception of such possible sales, could depress the market price of the common stock.**

Sales of a substantial number of shares of the Company's common stock in the public markets, which include an offering of its preferred stock or common stock, could depress the market price of the common stock and impair the Company's ability to raise capital through the sale of additional equity or equity-related securities. Pressure BioSciences cannot predict the effect that future sales of its common stock or other equity-related securities would have on the market price of its common stock.

**The Company's share price could be volatile and its trading volume may fluctuate substantially.**

The price of the Company's common stock has been and may in the future continue to be volatile. Many factors could have a significant impact on the future price of Pressure BioSciences' shares of common stock, including the following:

- the inability to raise additional capital to fund operations, whether through the issuance of equity securities or debt;
- the failure to successfully implement the Company's business objectives;
- compliance with ongoing regulatory requirements;
- market acceptance of the Company's products;
- technological innovations and new commercial products by the Company's competitors;
- changes in government regulations;
- general economic conditions and other external factors;
- actual or anticipated fluctuations in the Company's quarterly financial and operating results; and
- the degree of trading liquidity in shares of the Company's common stock.

**A decline in the price of Pressure BioSciences' shares of common stock could affect its ability to raise further working capital and adversely impact its ability to continue operations.**

The relatively low price of Pressure BioSciences shares of common stock, and a decline in the price of its shares of common stock, could result in a reduction in the liquidity of the Company's common stock and a reduction in its ability to raise capital. Because a significant portion of operations has been and will continue to be financed through the sale of equity securities, a decline in the price of the Company's shares of common stock could be especially detrimental to its liquidity and its operations. Such reductions and declines may force the Company to reallocate funds from other planned uses and may have a significant negative effect on its business plans and operations, including its ability to continue current operations.

If the price for Pressure BioSciences shares of common stock declines, it may be more difficult to raise additional capital. If unable to raise sufficient capital and unable to generate funds from operations sufficient to meet the Company's obligations, Pressure BioSciences will not have the resources to continue its operations. The market price for shares of the Company's common stock may also be affected by its ability to meet or exceed expectations of analysts or investors. Any failure to meet these expectations, even if minor, may have a material adverse effect on the market price of the Company's shares of common stock.

**If the Company issues additional securities in the future, it will likely result in the dilution of shares for existing stockholders.**

The Company's restated articles of organization, as amended, currently authorize the issuance of up to 50,000,000 shares of common stock and 1,000,000 shares of preferred stock. As of March 31, 2013, Pressure BioSciences had 12,149,267 shares of common stock issued and outstanding; 300 units of Series D issued and outstanding (convertible into 750,000 shares of common stock); 145,320 shares of Series G convertible preferred stock (convertible into 1,453,200 shares of common stock); 4,650 shares of Series J convertible preferred stock (convertible into 4,650,000 shares of common stock); outstanding options and warrants to purchase an aggregate of 8,418,974 shares of common stock; and 359,250 shares of common stock reserved for future awards, which the Company may grant under its equity compensation plan.

In September 2012, the Company raised the number of its authorized shares of common stock from 20,000,000 to 50,000,000. From time to time, Pressure BioSciences also may increase the number of shares available for issuance in connection with its equity compensation plan, adopt new equity compensation plans, and issue awards to its employees and others who provide services to it outside the terms of its equity compensation plans. The Board of Directors may fix and determine the designations, rights, preferences, or other variations of each class or series of preferred stock and may choose to issue some or all of such shares to provide additional financing in the future.

The issuance of any securities for acquisition, licensing, or financing efforts, upon conversion of any preferred stock or exercise of warrants, pursuant to the Company's equity compensation plans or otherwise, may result in a reduction of the book value and market price of the outstanding shares of the Company's common stock. If Pressure BioSciences issues any such additional securities, such issuance will cause a reduction in the proportionate ownership and voting power of all current stockholders. Furthermore, such issuance may result in a change in control of the Company.

**Financial Industry Regulatory Authority (FINRA) sales practice requirements may also limit a stockholder's ability to buy and sell the Company's common stock.**

FINRA has adopted rules that require that in recommending an investment to a customer, a broker-dealer must have reasonable grounds for believing that the investment is suitable for that customer. Prior to recommending speculative low-priced securities to their non-institutional customers, broker-dealers must make reasonable efforts to obtain information about the customer's financial status, tax status, investment objectives, and other information. Under interpretations of these rules, FINRA believes that there is a high probability that speculative low-priced securities will not be suitable for at least some customers. FINRA requirements make it more difficult for broker-dealers to recommend that their customers buy the Company's common stock, which may limit the ability to buy and sell the Company's common stock and have an adverse effect on the market for its shares.

**The Company has never paid dividends on its common stock and does not anticipate paying any in the foreseeable future.**

Pressure BioSciences has never declared or paid a cash dividend on its common stock and does not expect to pay cash dividends on its common stock in the foreseeable future.

**The shares of Series D convertible preferred stock are entitled to certain rights, privileges, and preferences over the Company's common stock, including a preference upon a liquidation of the company, which will reduce amounts available for distribution to the holders of the Company's common stock.**

The holders of shares of Series D are entitled to payment, prior to payment to the holders of common stock in the event of liquidation of the Company.



## Glossary

**2D Gels**—Used in two-dimensional (2D) gel electrophoresis, an established technique for high-resolution profiling of low-abundance proteins.

**Analytes**—A substance for which the chemical constituents are being identified and measured.

**Bead Beaters**—Instruments that break apart cells by vibrating them against glass beads. This is a widely used and inexpensive method to lyse cells.

**Biological Samples**—Cell and tissue specimens from humans, animals, plants, and other living organisms. Biological samples from humans can include tissues, bodily fluids (e.g., blood, saliva), and excreta (stool and urine).

**Biomarker**—A measurable substance in an organism whose presence is indicative of some phenomenon such as disease, infection, or environmental exposure.

**Buffers**—In biochemistry, a buffer, specifically a buffer solution, is essential for many biochemical processes. It serves to maintain the correct pH necessary as many enzymes work only under precise pH conditions.

**Chemical Dissolution**—Breaking down, disrupting, or dispersing a sample by applying chemicals.

**Chromatography**—The separation of a mixture by passing it in solution or suspension or as a vapor through a medium in which the components move at different rates.

**Differential Organic Extraction**—A process by which the DNA from two different types of cells can be extracted without mixing their contents.

**Dounce Homogenization**—A glass mortar and pestle with a very small clearance between the mortar and the pestle.

**Electron Paramagnetic Resonance (EPR)**—A spectroscopy technique for studying materials with unpaired electrons. Also called electron spin resonance (ESR).

**ELISA (Enzyme-linked Immunosorbent Assay)**—An immunological assay technique making use of an enzyme bonded to a particular antibody or antigen.

**Enzymatic Digestion**—The use of enzymes to break down proteins into amino acids.

**Epithelial**—Relating to the epithelium, the membranous tissue covering internal organs and other internal surfaces of the body.

**Filtration**—Passage through a filter or other material that prevents passage of certain molecules, particles, or substances.

**Fluoropolymer**—An organic polymer containing fluorine atoms, such as polytetrafluoroethylene.

**Fractionating**—Breaking up a sample into smaller parts.

**Freezer Mills**—Cryogenic laboratory impact mills that process and grind samples.

**French Press**—An apparatus used in biological experimentation to disrupt the plasma membrane of cells by passing them through a narrow valve under high pressure.

**Genomics**—The branch of molecular biology concerned with the structure, function, evolution, and mapping of genes.

**Glycosylation**—The process by which sugars are chemically attached to proteins to form glycoproteins.

**Hematin**—The hydroxide of heme; it stimulates the synthesis of globin, inhibits the synthesis of porphyrin, and is a component of cytochromes and peroxidases. It is also used as a reagent.

**Homogenizer**—A machine that forces a substance through fine openings against a hard surface for the purpose of blending or emulsification.

**Humic Acid**—One of two classes of natural acidic organic polymer that can be extracted from humus found in soil, sediment, or aquatic environments.

**Hydrostatic**—Of or relating to the pressures fluids exert or transmit.

**Immunoaffinity Chromatography**—A method of separating a mixture of proteins or nucleic acids (molecules) by specific interactions of those molecules with a component known as a ligand, which is immobilized on a support.

**Inhibitors**—Substances that reduce or suppress the activity of another substance.

**Laser Microdissection**—A contact- and contamination-free method for isolating specific single cells or entire areas of tissue from a wide variety of tissue samples.

**Lipidomic**—An emerging field of biomedical research that studies and analyzes the set of lipid species present in an organism (the “lipidome”).

**Low Copy Number Specimens**—A sample with only small amounts of DNA recovered for analysis.

**Mass Spectrometry**—An analytical laboratory technique to separate the components of a sample by their mass.

**Mortar and Pestle Grinder**—A tool used to grind and mix substances in a laboratory. The pestle is a club-shaped, hand-held tool and the mortar is a bowl.

**Nuclear Magnetic Resonance (NMR)**—The selective absorption of electromagnetic radiation by an atomic nucleus in the presence of a strong, static, magnetic field: used in research and in medicine to monitor tissue metabolism and to distinguish between normal and abnormal cells.

**Proteomics**—The branch of biotechnology concerned with applying the techniques of molecular biology, biochemistry, and genetics to analyze the structure, function, and interactions of the proteins produced by the genes of a particular cell, tissue, or organism.

**Reducing Agents**—A substance that tends to bring about reduction by being oxidized and losing electrons.

**Sample Preparation**—The series of steps needed to convert a representative bulk sample into a form suitable for chemical analysis.

**Sensitivity**—Measures the proportion of actual positives which are correctly identified as such (e.g., the percentage of sick people who are correctly identified as having the condition).

**Small Molecules**—Organic molecules of small molecular weight. They possess a biological activity determined by the fact that they bind to the active site of a protein responsible for the disease, called “molecular target.”

**Sonicators**—A laboratory instrument used to disperse, disrupt, or inactivate biological material (e.g., viruses) by sound waves.

**Supernatant**—The liquid lying above a solid residue after crystallization, precipitation, centrifugation, or other process.

**Thermomixer**—A laboratory instrument produced by Eppendorf Group for mixing and incubating samples.

**Touch Samples**—A forensic method for analyzing DNA left at the scene of a crime. It is called “touch sample DNA” because it requires very small samples, such as from skin cells left on an object after it has been touched or casually handled.

**Western Blots**—A technique for identifying specific antibodies or proteins in which proteins are separated by electrophoresis, transferred to nitrocellulose, and reacted with antibody.



## CRYSTALRESEARCH ASSOCIATES

### EXECUTIVE INFORMATIONAL OVERVIEW®

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Crystal Research Associates is led by veteran Wall Street sell-side analyst Jeffrey Kraws, who is well known by the international financial media for his years of work on Wall Street and for providing consistent award-winning analyses and developing long-term relationships on both the buy-side and sell-side. He has been consistently ranked on Wall Street among the Top Ten Analysts for pharmaceutical stock performance in the world for almost two decades as well as ranked as the Number One Stock Picker in the world for pharmaceuticals by Starmine and for estimates from Zacks. Additionally, Mr. Kraws has been 5-Star ranked for top biotechnology stock performance by Starmine.

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