The Future of Peptide Production and Automated Peptide Synthesizers

Protein Technologies, Inc.

Recent years have seen interesting advances in the automated peptide synthesizer field. While initial innovations typically involved retrofitting existing technology platforms to peptide synthesis, the next generation of peptide synthesizers use technologies designed specifically for the task of peptide synthesis. This issue contains the latest developments in peptide synthesizer technology including improved UV-monitoring technologies and improved robotic peptide library synthesis platforms.

When Bruce Merrifield invented solid phase peptide synthesis in 1963, he opened up the doors to automation. Since then, automated synthesizers have grown in their capabilities as the chemistry has grown. Some of the major innovations in peptide synthesizers over the years include the introduction of online-monitoring systems using conductivity or ultraviolet light to measure the extent of the deprotection reaction, robotic peptide library synthesizers to meet the throughput needs of combinatorial chemistry applications, and microwave technology to speed up synthesis reactions.

When microwave peptide synthesizers were first introduced a few years ago, they were constructed by hooking up an existing microwave digester to a new fluidics system. As with any new technology, microwaves have eventually settled into their own niche. They promised faster synthesis times, higher purities and higher yields. Heat has been shown to increase reaction speed and efficiency in some cases, and microwaves are a convenient way to add heat. However, heating can also accelerate some side reactions including racemization, gamma-lactam formation, phosphate-group cleavage, and diketopiperazine formation, requiring synthetic workarounds.

Symphony 12-channel peptide synthesizer

The main weaknesses of microwave synthesizers are that they are only able to process one sample at a time, and they cannot be scaled up beyond 1 mmol due the inability of microwaves to penetrate solutions by more than a few centimeters. Therefore, despite the promises of higher throughput through faster synthesis speeds, they have not been able to compete with existing parallel conventional synthesizers like the 12-channel Symphony®. With the availability of low-cost, highly-efficient activators such as HCTU, conventional synthesizers can easily match the reaction times of microwave synthesizers and produce higher numbers of high purity, high yield peptides in comparison to serial microwave units. In addition, microwave methods developed at small scales cannot be used in large scale production. Thus, currently, microwave systems are not used for general peptide production, but are usually used as one alternative option for the occasional difficult coupling at small scales. As a result, newer microwave synthesizer offerings have gone to lower throughput models, or include a microwave on the side for occasional use, while the majority of the synthesis is performed conventionally.

The ability to monitor the extent of peptide synthesis reactions to ensure they have gone to completion before moving on to the next step would result in the highest purities and yields for a given peptide. Although the ability to automatically monitor the coupling reaction has not been achieved, on-line monitoring systems for measuring the extent of the deprotection reaction have been around since the 1990's, starting with conductivity monitoring in 1993, which was eventually replaced by UV-monitoring first at 365 nm and finally at 301 nm. These systems were made by connecting existing external UV flow-cell monitors to a peptide synthesizer. Since then, no major breakthroughs

in this technology have taken place until 2009, with the introduction of the IntelliSynth UV-Monitoring and Feedback Control System for the Tribute® peptide synthesizer from Protein Technologies, Inc. This wholly new technology was developed to be incorporated inside a peptide synthesizer, giving it the never before seen ability to monitor *during* the deprotection reaction. It can then use that data to control the *length* of the reaction, rather than just monitoring the waste stream at the end of the reaction. In order to achieve this breakthrough, a whole new UV platform was developed specifically for peptide synthesis.

Tribute peptide synthesizer with UV-Monitoring

Robotic peptide library synthesizers were also introduced in the 1990's to meet the demands of the growing combinatorial chemistry market spurred on by improvements in high throughput screening methods. These synthesizers typically use generic, off-the-shelf robotic platforms and adapt them to peptide synthesis. Although robotic units have been developed to handle increasingly higher numbers of samples in one run, very little innovation has taken place since then in the actual robotic platforms. The Overture™ Robotic Peptide Library Synthesizer, introduced this year by Protein Technologies, Inc., was designed to overcome a lot of the shortfalls in the typical off-the-shelf robotic platforms now available for use. New features include innovative fluid delivery methods, single-point calibration, and integrated automated cleavage without user intervention. By offering significant savings in solvent and time, better consistency, and efficiency, the Overture™ represents the next leap forward in robotic peptide library synthesizer technology.

Overture robotic peptide library synthesizer

Many companies have chosen to adapt existing technologies and platforms to the task of peptide synthesis resulting in platforms that perform the task, but often leave room for improvement. Recent breakthroughs in UV-monitoring and robotic peptide library synthesizer technologies represent a different approach where a technology is specifically designed for the task of synthesizing peptides. It is this second approach that has supplied the best technologies for the task, and will be needed in the future to design the best possible instruments for peptide synthesis going forward.

Protein Technologies, Inc. (PTI) is a private company located in Tucson, AZ that specializes in the manufacture and distribution of high quality peptide synthesizers and reagents. PTI peptide synthesizers have a reputation for their high performance, flexibility and reliability, and are based on a unique, patented matrix block technology. PTI has placed hundreds of peptide synthesizers in academic institutions, biotechnology companies and pharmaceutical companies worldwide, and strives to provide the best service to its customers through both technical and chemistry support. For more information about Protein Technologies, visit: www.ptipep.com.