

A Market Review Of DNA-encoded Libraries Technology In Drug Discovery

Dec. 7, 2019 by Irina Bilous , Andrii Buvailo

The interest of pharma organizations in DNA-encoded chemical libraries (DEL) technology has been growing over the years, with numerous pharma organizations now having their own screening programs using DELs, or outsourcing capabilities from specialized DEL providers.

This report provides a bird's view on the DEL market, including a brief summary of DEL technology benefits and limitations, key players, major research deals, and several examples of successful hit discovery programs using DELs.

Historical context

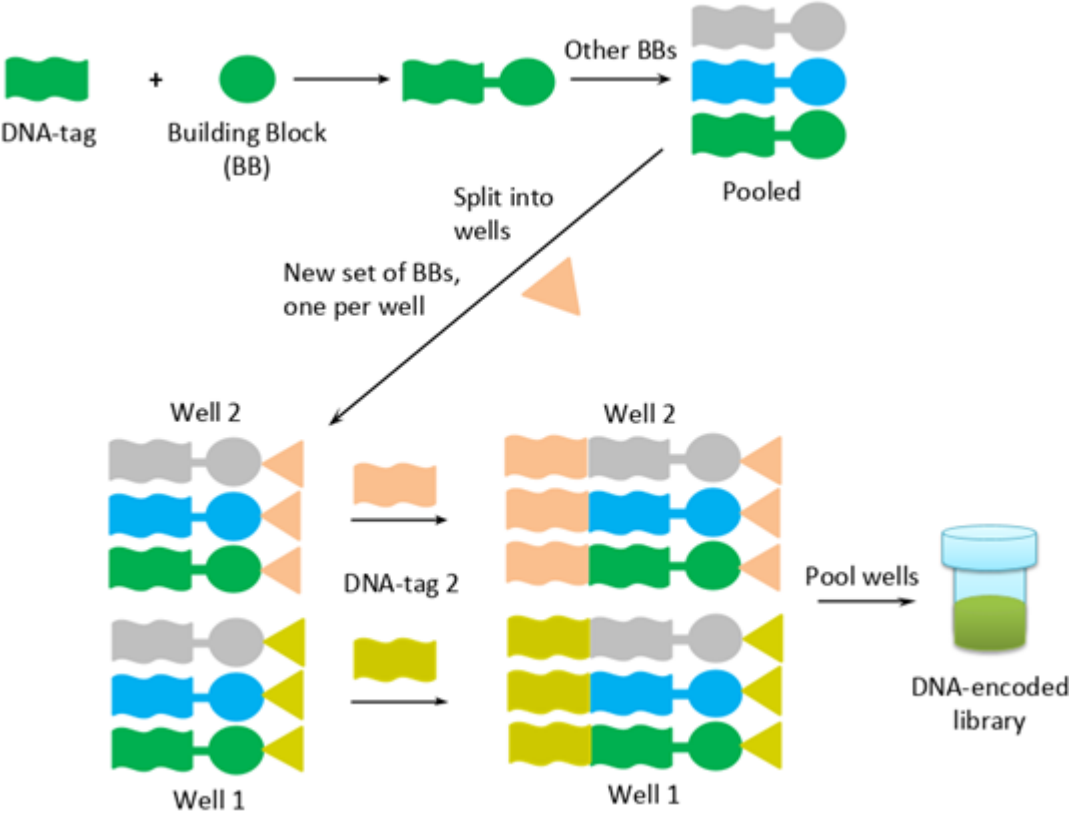
The concept of DNA-encoded libraries (DEL) appeared in the early 1990th when scientists from Scripps Research Institute published a paper on "Encoded combinatorial chemistry". The main principle of DEL technology is to encode each compound in the library with unique DNA barcode. Thereby library of billion compounds can be screened as a mix in a single test tube and the active compounds are identified by amplification and sequencing of the DNA barcodes.

At the initial stage, companies pioneering in DEL development were faced with a lack of relevant methods for DNA sequencing. Scientists from Praecis Pharmaceuticals then founded a company which managed to sequence DEL libraries. Praecis and Nuevolution were the first to have taken a chance on DEL in the 2000s. In 2005 and subsequent years the second generation sequencing has emerged making DEL a much more attractive and economically feasible approach in drug discovery.

An overview of DEL technology

DEL technology comes from the merge of DNA encoding and combinatorial chemistry. The most popular method to build DEL is "split and pool" approach. At the first step of the synthesis, chemical building blocks (BBs) are tagged with DNA barcodes. In the second step, they are mixed together and then split into different portions. Next, a new set of building blocks is added to the chemical reaction, and then, corresponding DNA barcodes are attached and linked with previous DNA molecules. Thus, DNA encodes

each step of chemical synthesis for each compound.



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Size and costs of DELs

“Split and pool” method enables to build libraries containing up to 10^{10} compounds. For example, HitGen’s libraries contain 400 billion molecules, X-chem provides 200 million compounds for screening, and Nuevolution has assembled a collection of 40 trillions of compounds. To compare, the size of a common HTS library is restricted to several million compounds due to the prohibitive cost of synthesizing and managing larger collections. Overall, the cost of creating and screening HTS library of 1 million compounds would cost millions of dollars (approximately \$1,100 per compound), while screening a DEL library of 800 thousand compounds would cost on the order of \$150,000.

Hit discovery using DELs

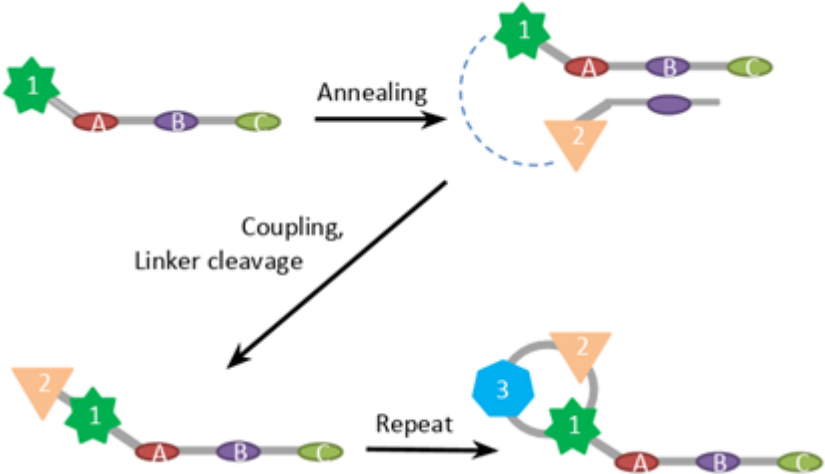
Screening of the DELs is conducted in single test tubes, where the target protein is incubated with a library. The target proteins can be anchored on a solid support, commonly magnetic beads. After thorough washing, only high-affinity ligands remain associated with a target. After DNA sequencing and decoding of the structure of active compounds, the last should be resynthesized to repeat the affinity binding assay. A lot of different selection strategies can be applied, even phenotypic screening is compatible with DELs.

DEL is suitable for the synthesis and screening of small molecules, macrocyclic compounds, and peptides. Scientists are harnessing chemical properties of DNA, especially the principle of complementarity, for developing new strategies for library design.

DNA templated strategy

One of the more sophisticated methods to build libraries is to use DNA code to determine the sequence of chemical reactions. Templates are prepared before synthesis and contain regions complementary to barcodes of BBs. When barcodes couple to the template, the relative sterical arrangement of building blocks facilitates a chemical reaction between them. This method is particularly useful when assembling libraries of macrocycles and peptides.

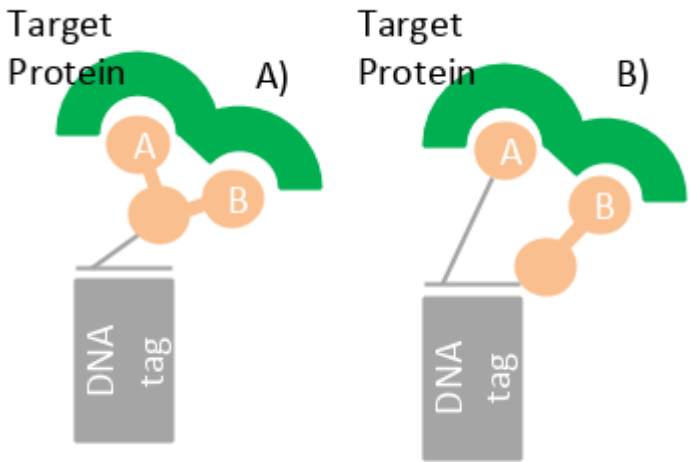
The alteration of the approach is used in Viperger’s YoctoReactor.



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Dual-pharmacophore libraries

The advantage of dual-pharmacophore libraries is that two chemical moieties have the flexibility to reach adjacent non-overlapping binding sites of a target. Dual-pharmacophore libraries are assembled from two sub-libraries, when mixing together they form heteroduplexes representing two compounds. ESAC (Encoded Self Assembling Chemical) is the most popular method.



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DEL-compatible chemistry

The chemistry applicable to DEL technology is restricted to reactions compatible with DNA. DNA is a water-soluble molecule, thereby traditional organic reactions should be translated to corresponding conditions. In the beginning, only a few reactions were available in the toolbox of chemists, but their number is extended, harnessing various synthesis approaches. One of the recent ideas was to employ photoredox catalysis for DNA-Encoded Chemistry.

Pros and Cons of DNA-encoded Libraries Technology

Benefits	Limitations/challenges
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Large size (up to 10 ¹⁰ molecules available for screening)	Limitations in types of BBs and reactions that can be used (has to be compatible with DNA chemistry)
Might be applied toward “undruggable” targets	Oligonucleotide might affect binding affinity
Low costs and time of screening compared to HTS	Targeting of DNA/RNA-binding proteins is challenging
Less target amount is needed when compared to HTS, relatively simple assay methods	Need to resynthesize active molecules to determine their affinity
Library refinement capabilities, flexibility.	Challenges with signal-to-noise ratio when selecting active compounds

Market and trends

HitGen and X-Chem are two major providers of DEL capabilities. Both companies leverage the “split and pool” approach to construct libraries. They have announced the bigger part of DEL collaborations with pharmaceutical companies. These two companies offer their partners to screen ready-to-use DELs, or to construct libraries for their needs. HitGens libraries contain approximately 400 bln novel drug-like molecules.

Several pharmaceutical giants as GSK, Novartis and, from recent times, Amgen, are developing DEL primarily for their own targets of interest. GlaxoSmithKnille was the first company adopting technology. In 2007 GSK acquired Praecis Pharmaceuticals (pioneering company) after the successful collaboration in 2006, which eventually led to 2 compounds getting their ways to clinical trials. More about GSK’s DEL you can find here. A similar story occurred with Amgen and Nuevolution which started collaboration in 2016. Three years of collaboration have led to an acceleration of the progress of two cancer programs. This year, 2019, Amgen acquired Nuevolution for \$167 million to advance its drug discovery efforts.

The number of partnerships announced for DEL technology keeps on growing (Fig.1). The year 2014 was particularly fruitful for the DEL market, due to the activity of Vipergen, X-chem and Nuevolution. HitGet, founded in 2012, went on a deal-sprees starting in 2017 with more than a dozen deals with various partners.

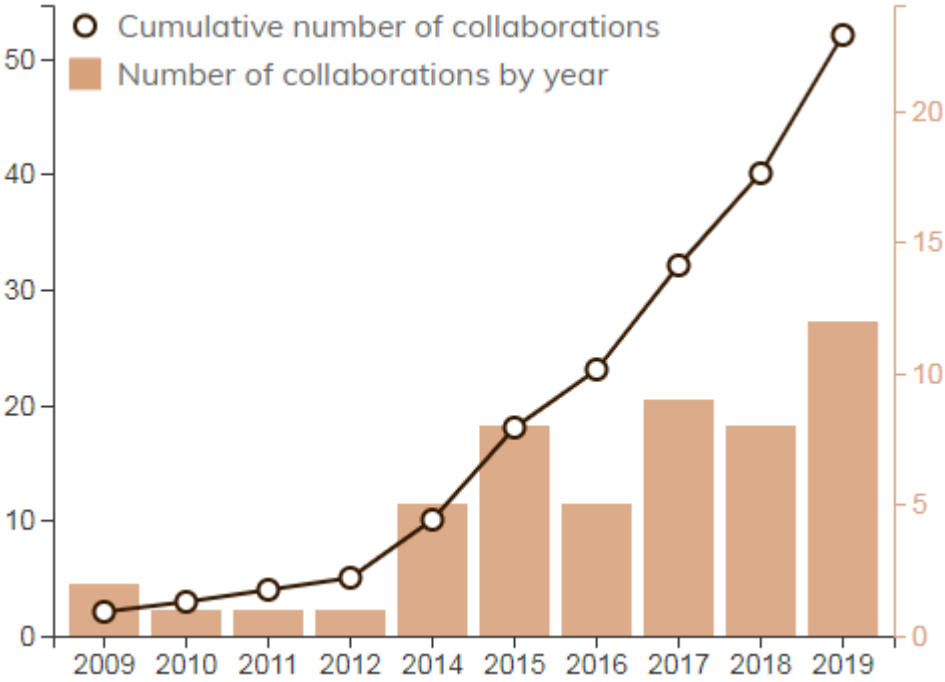


Fig.1 Number of collaborations on DEL per years*

* Source: BPT Analytics

Behind acknowledging the activity of large brands in the DEL space, a number of startups have been founded over the last few years, including Haystack Sciences (received \$4.2M series A financing in October 2019), DyNAbind GmbH (raised seed round in 2019), Plexium (received \$28M in series A financing in October 2019), etc.

The technology seems to be developed to such a level that several companies are providing DEL kits for the library screening. In September 2019 WuXi AppTech announced the launch of DELlight platform which would supply customers with DEL kits containing 8.4 billion compounds together with operational protocols for performing the procedure. DyNAbind's kits are available for around 9 thousand EUR at Sigma-Aldrich.

WuXiAppTech also established the open-source library DELopen to provide free access to more than 2.8 billion DEL compounds for academic users. With the use of blockchain technology, DELopen offers maximum IP protection for users who utilize the online platform.

Smaller companies and startups

Small companies and startups harnessing DELT are worth to be considered in more detail. They exploit DEL approach for a number of notable use cases, including ubiquitin-proteasome system (UPS), allosteric regulatory sites, PPI and multi-drug resistant bacterial infections. Below is a brief reference to several advanced combinations of DELs with other technologies.

DEL in phenotypic screening -- protein degradation

Plexium's DELPhe platform enables cell-based phenotypic screening of DNA-encoded libraries. Their technology miniaturizes screening to the scale of picoliter volumes. Plexium is focused on developing modulators of E3 ligases.

DEL for studying allosteric sites

HotSpot is focused on developing drugs that interact with allosteric sites - "regulatory hotspots". In 2014, they acquired Macroceutics, Inc, a provider of DEL screening technologies to advance their HotSpot's SpotFinder™ platform which can be valuable in predicting and evaluating different allosteric regulatory sites.

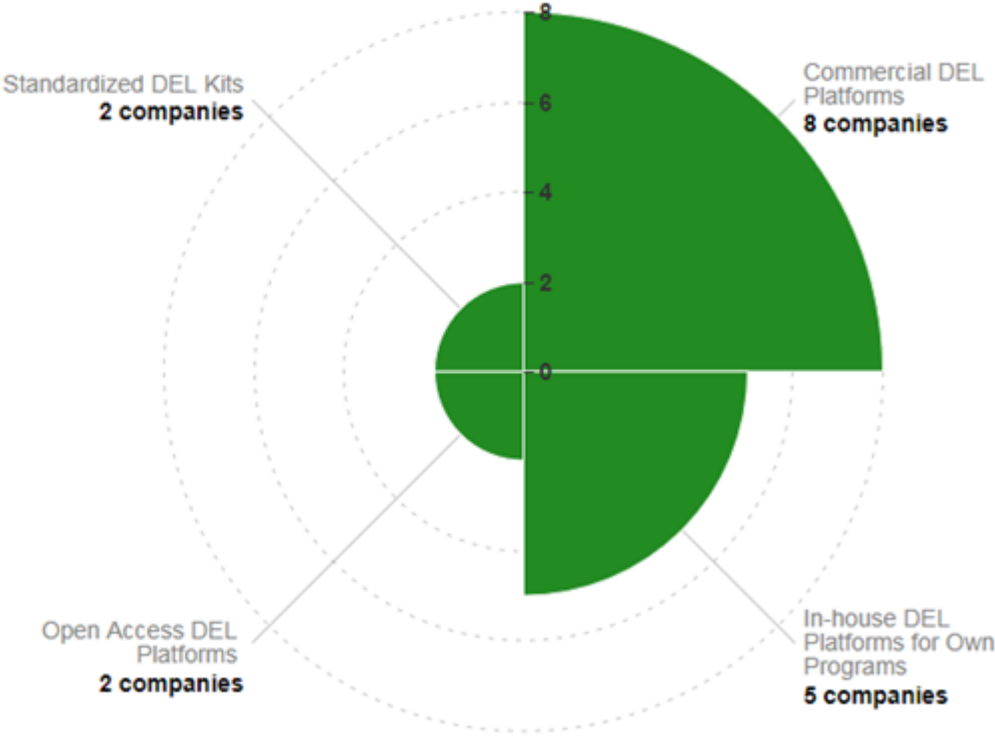
Applying machine learning (ML) for optimizing DELs

Haystack uses the power of machine learning to estimate structure-activity relationships of compounds and provide a rapid refinement of libraries

In summary, most of the DEL platforms can roughly be classified into four categories:

1. commercial DEL platforms

- 2. in-house DEL platforms for own programs
- 3. open-access DEL platforms
- 4. standardized DEL kits



* Source: BPT Analytics

Some of the notable DELT successes to date

Besides the number of deals and emerged companies, the pipeline derived from DELs is no less encouraging. The first example is Nuevolution’s BET-BD1 program. BD1 is the first bromodomain of the BET family which is responsible for epigenetic regulation of the immune system. A candidate molecule NUE20798 has started clinical trial this year against atopic dermatitis. Previously, inhibitors of this class have not been approved. Another successful example came from a partnership with Almiral in 2016: ROR γ t inverse agonist enters the clinic for treatment psoriasis (PsO) and psoriatic arthritis in 2019.

X-165 has been accepted by FDA for Phase 1 clinical trial. This is a highly potent inhibitor of autotoxin owned by X-Rx. This company spun out from X-chem, and was using their DEXtm (based on DEL technology) platform for drug discovery. Since 2012, X-chem has licensed 50 drug discovery programs.

HitGen got approval from FDA for the clinical trial of a drug candidate against multiple myeloma. The company also developed other molecules against different targets in their pipeline.

Finally, GSK specific inhibitor of the RIP1 kinase GSK2982772 completed a Phase II clinical trial for psoriasis. Another inhibitor of the soluble epoxide hydrolase (sEH) GSK2256294 is currently undergoing Phase II testing.

- DyNABind GmbH
- Haystack Sciences
- HitGen
- HotSpot Therapeutics
- Plexium
- X-Chem Pharmaceuticals