PATENT REVIEW

The section on patent review will be focused in the areas of interest to the readers of CCHTS. The search was conducted using the following key words: *combinatorial chemistry, high throughput screening, drug repurposing, chemical library, high content screening, drug discovery and natural products*. All patents highlighted here are identified by the patent number issued either by the World Intellectual Property Organization or by a regional patent office.

DRUG REPOSITIONING

The paradigm for finding new uses for known or failed drugs has generated new investments and interest in pharmaceutical sector. Drug repositioning or repurposing includes finding different therapeutic indications for drugs with established or even unknown pharmacological mechanism. Both marketed drugs as well as compounds with history of failure during clinical development are candidates for repositioning. Recent patent activity has focused on the experimental methods or theoretical mathematical modeling for selecting drugs for drug repurposing.

WO2010141592A2: *Chemical fragment screening and assembly utilizing common chemistry for NMR probe introduction and fragment linkage*, (Sem, Daniel, S., Marquette University, USA) patent describes a potentially powerful tool to overcome the bottleneck in fragment-based drug design and extends the approach to tailoring known marked drugs for repurposing or increasing potency or reducing side-effects of targeted treatments with existing drugs. The fragment based approach requires identification of short molecular fragments that bind to adjacent sites of a target. One of the fragments, an active scaffold is labeled with 13C-methyl group. The challenges associated with linking two short fragments for drug design is approached using NMR-based fragment assembly. NMR is used at an early stage to detect any transfer of magnetization (NOE) between the labeled scaffold and test fragments, which is detectable only if the fragments bind to a target within 5 Angstrom distance, followed by chemical tethering of the fragments to sites where NOEs were detected. Any substantial increase in the binding affinity of tethered compounds selects the fragments and also precludes the time and cost associated with chemical synthesis of linked fragments. In a related set of patent applications, the uptake of isotope-labeled substrates/precursors in a biological system is accompanied with administration of known or unknown drug (s) or test compound(s). In the patent WO05051434A1: *Method for high-throughput screening of compounds and combinations of compounds for discovery and quantification of actions, particularly unanticipated therapeutic or toxic actions, in biological systems* (Hellerstein, Marc, K, University of California), the changes in patterns or content of isotopically labeled target molecules is determined in cells, tissues or whole animals through expected or unanticipated metabolic pathways and the rates are compared between labeled systems left untreated with drugs/compounds. An extension of the same principle in related patents WO06017812A1 and WO2007041611A2 focus on quantifying the effect of drugs/compounds on the dynamics of assembly or disassembly of isotopically labeled subunits of cytoskeletal system like the microtubules, amyloid plaques or plasma membrane disruptions.

In the following set of patents, methods involving data mining information on diseases, pathways and known drugs forms the basis for the development of various approaches and models to drug repositioning. The patent WO2009068659A2: *Novel disease treatment by predicting drug association* (Cohen, Daniel; Chumkov, Ilya, Pharnet, France), describes a methodology based on mining of public database to select a disease and build a dynamic model of the disease and the molecular pathways. This is followed by in silico screening of drugs approved for other diseases that encompass all target pathways directly or indirectly implicated in the model. The *in silico* selected drugs either alone or in combination(s), are then tested in available biological model of the disease, to identify candidates for the treatment of the selected disease. Using this in silico approach, some compounds were proven to have the required biological activity against the targets selected in the patent. The strategy was shown to be effective The patent WO2009027843A2: *Techniques for purposing a new compound and for repurposing a drug*, (Zoref, Tali, Eilam and Agur, Zvia, Optimata ltd , Israel) describes a computer modeling based approach for repurposing and is based on information and disease models available for an approved drug or for compound(s) that failed in clinical development. Based on information on the drug's pre-clinical and clinical trials, modified pharmacokinetic and pharmacodynamic mathematical models are reconstructed and the model is adjusted based upon information about new patient populations or new indications. A new treatment protocol is suggested to salvage the failed drug or a new way to use an approved drug.

HIGH THROUGHPUT SCREENING

In patent WO2009078876A1: *Assay method for group transfer reactions* (Lowery, Robert *et al*., Bellbrook Labs, LLC, USA), a generic methodology is described for high throughput screening of catalytic activities generating the donor-products in group-transfer reactions of enzymes like methyltransferases, sulfotransferases, kinases, glycosyltransferases, uridine glucuronide transferase, UDP- glucuronosyltransferases, acetyl transferases, glutathione transferases, and ADP-ribosyltransferases. The methodology is based on generating an antibody which binds with high specificity to the cleaved donor product in a reaction, donor-X + acceptor -» donor-product + acceptor –X. In a methyltransferase reaction, the antibody that specifically binds to S-adenosylhomocysteine with high affinity is used at the detection step in which the enzymatically generated donor product displaces the tracer-labeled donor-product from its complex with the specific antibody, as in fluorescence polarization immunoassay. Because the donor product is the same for all enzymes that catalyze a given type of group transfer reaction, the same detection reagents can be used for all the members within a family of group transfer enzymes

and with any acceptor substrate. The assay products can be detected using homogenous fluorescence or chemiluminescence methods.

Patent WO2010137017: *Proteasome inhibitors and uses thereof* (Lavlin, I *et al*., Yeda Research and Development Co. Ltd. Weizmann Institute of Science), describes a new class of proteasome inhibitors which bind to proteasome of a cell. The compounds consist of a copper bound to a ligand, the ligand being configured such that upon binding to the proteasome, the copper interacts with two cysteines comprising the active site of proteasome thereby treating the disease. The patent also describes the stable cell line expressing a mutant p53 peptide fused to a Yellow Fluorescent protein. The mutant p53 is cytoplasmic in absence of proteasome inhibitor and is localized in the nucleus when proteasome inhibitors were present. In the proof of concept patent US20100330599A1: *Inhibitors of USP1 deubiquitinating enzyme complex*, (D'Andrea, Alan, Dana-Farber Cancer Institute, Inc., Boston), the inventors claim that inhibition of USP1 deubiquitinase resulted in increased levels of monoubiquitinated PCNA in eukaryotic cells, leading to enhanced DNA repair activity in the cell nucleus. USP1, a protease (deubiquitinase) that normally deubiquitinates the monoubiquitinated form of PCNA (PCNA-Ub). The inhibition of USP1 or its heterodimeric partner, USF1 (USP1 Accessory Factor 1) improves cell survival by increasing translesion DNA synthesis activity in the nucleus and such inhibitors are radioprotective and chemoprotective to cells. Identification of USP1 inhibitors will be useful in treatment of radiation exposure from industrial, research or medical equipment, a nuclear reactor, or from an explosive device, radioactive fallout from a nuclear accident or explosion, and radiation exposure from mines, mineral refineries, or space travel. The invention also provides a composition consisting of a fragment of USP1 having deubiquitinase activity, PCNA, and an inhibitor of the USP1 fragment or of UAF1 that result in increased levels of ubiquitinated PCNA, which increases DNA transcription through increased translesion DNA synthesis.

The success of Velcade, a proteasome inhibitor, in the treatment of multiple myeloma has resulted in several patents that target more specific players in the ubiqitination and proteasome pathways. Patent WO2010003908: *Screening assay for compounds targeting the p97 AAA-ATPase complex in the ubiquitin proteasome system* (Young, Patrick *et al*.) describes a novel system based on p97 complex (AAA-ATPase) which targets many misfolded proteins to the endoplasmic reticulum targeted degradation. The method is based on using a cell-line expressing substrate reporter comprising of a degron (targets reporter for degradation) upstream of fluorescent reporter protein fused to a short unfolded extended peptide, which gets ubiquitinated and result in reporter degradation in absence of a proteasome inhibitor. The controls are cell lines expressing the reporter with degron-reporter fusion lacking the extended peptide which retains activity at all times. WO2010000372: *New drug for inhibiting aggregation of proteins involved in diseases linked to protein aggregation and/or neurodegenerative diseases* (Giese, Armin *et al*., Ludwig-Maximilians-Universität München) patent describes the use of substituted heterocyclic derivatives for treating disease linked to protein aggregation and neurodegenerative diseases, e.g. Parkinson's disease, Prion disease, Alzheimer's disease and amyotrophic lateral sclerosis.

WO2010017478A2: *PAK1 agonists and methods of use* (Ke, Yunbo & Solaro, Ross, John; The Board of Trustees of the University of Illinois). The p21 activated kinase-1 (Pakl), a serine/threonine protein kinase, is abundant in the heart and has a role in cytoskeletal function and reorganization in cardiomyocytes. Pak1 activators promoter cardiac cell survival and have anti-adrenergic effects similar to that of known beta-blockers but without any side-effects. The patent reports identification of c2 and c6 ceramides, safingol, D-sphingosine and sphingosine 1-phosphate which modulate Pak1 phosphorylation and reduce heart rate *in vitro* and may serve as building blocks for development of drugs targeting heart patients especially ones with asthma in combination with heart failure.

Patent US20100250218: System and method for prediction of drug metabolism, toxicity, mode of action, and side effects of novel small molecule compounds (Ekins, Sean et al.) describes a system for the prediction of human drug metabolism and toxicity of novel compounds. The system enables the visualization of preclinical and clinical high-throughput data in the context of a complete biological organism. Substructure and similarity structure searches can be performed using the underlying databases of xenobiotics, active ligands, and endobiotics. The system also has an analytical component for the parsing, integration, and network analysis of genomics, proteomics, and metabolomics high-throughput data. From this information, the system further generates networks around proteins, genes and compounds to assess toxicity and drug-drug interactions.

CHEMICAL LIBRARY

US20080255001: Screening *of chemical compounds purified from biological source* (Eldridge, Gary *et al*.), describes a method for creating a natural product library which removes highly abundant components that may mask the activity of less expressed molecules. The method aims at producing chemical compound library from plants, containing normalized chromatographic fractions by removing non-drug like compounds expressed in high abundance from the plant extracts and enriching low abundance biologically active chemical compounds for drug discovery. The method requires chromatographically separating the processed extract into several chromatographic fractions and determining the amount of chemical compounds in at least one of the chromatographic fraction. The characterized fraction is normalized into multiple chromatographic fractions, each such fraction comprising from about 1 microgram to about 500 micrograms from one to seven chemical compounds that were present in lower concentrations in the extract and that each have a log P of from about -1 to about 5 and a molecular weight less than about 1000 Daltons ; this will help produce a chemical compound library of much greater number of purified, relevant drug-like chemical compounds per plant for biological screening than currently used by those skilled in the art. In patent WO2009015059: *Organo-cascade catalysis: one-pot production of chemical libraries*

(Macmillan, David, The Trustees of Princeton University), a method for large-scale synthesis of a chemical library is described. The method comprises of reacting unsaturated aldehydes and/or ketones, nucleophiles and/or electrophiles, in presence of organo-cascade catalyst like iminium-enamine, for the production of chemical libraries. The single operation method described in the patent results in synthesis of complex products in a reaction that avoids the introduction of metallic based reagents and reagent byproducts that otherwise need to be removed since they have an adverse impact on the biological screen to be performed. The cascade products can be used as intermediates in other chemical transformations to introduce chemical diversity and extend the range of chemical functionalities present in the final products.

COMBINATORIAL CHEMISTRY

Patent WO2009037642: Heterocyclic *scaffolds useful for preparation of combinatorial libraries, libraries and methods of preparation thereof* (Gellerman, Gary, Ariel - the university company for research and development, Ltd) describes methods using new orthogonally-protected, heterocyclic, chiral scaffolds for the synthesis of drug candidates and for solid-phase organic synthesis of combinatorial libraries. Patent WO2010114762 describes a new reagent for incorporating colorimetric-oxycarbonyl protecting group, and its use in solid supported organic syntheses of oligonucleotides, polypeptides, polysaccharides, and combinatorial libraries. Patent US20090197772: *Compartmentalised combinatorial chemistry by microfluidic control* (Griffiths, Andrew *et al*.) describes methods compartmentalizing two or more sets of primary compounds into microcapsules and forming secondary compounds in the microcapsules by chemical reactions between primary compounds from different sets; wherein one or both of steps are performed under microfluidic control. The invention further allows for the identification of compounds which bind to a target component of a biochemical system or modulate the activity of the target, and which is co-compartmentalized into the microcapsules.

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