A. DRUG DISCOVERY STRATEGY

Modern drug discovery and development play a vital role in transforming a molecule from laboratory into a drug candidate and is a high priority contribution from a scientific community. In this context, the drug discovery process can be broadly segregated into two segments.

1. Identification and optimization of lead molecules to enhance their selectivity towards the target including their toxicity profile.

2. Development of a relevant biological system to test the compounds in vitro and in vivo models to speed up the drug discovery process and to improve the screening efficiency and success rate.

Drug discovery and development is a complex, time consuming and an expensive process since the safety, efficacy and other issues are mandatory. Generally, it takes about more than 10 years to bring a new drug developed from its initial screening stage to final FDA approval and has a huge failure rate at each stage of the developmental process. To identify this issue, there are several new techniques available such as, combinatorial chemistry, green organic synthesis, high-throughput purification, solid phase synthesis, liquid phase synthesis, logic centered molecular synthesis, molecular docking and QSAR analysis. Even with such modernization and advancement in research and development, the number of new chemical entities reaching the market has decreased considerably. New drug candidates meant to treat a disease might theoretically range from 5,000 to 10,000 chemical compounds. On an average, about 250 test compounds might reach the
In vivo evaluation stage. In all probability, may be ten of these shall qualify for experiments on humans. A study conducted by the Tufts center for the study of drug development covering the period 1980 to 1990 alluded that, only 1/5th of the drugs that begin with phase I clinical trials get eventually approved for marketing, giving an impression that, selection of the appropriate molecules for synthesis becomes one of the most challenging tasks.

New methodologies which pave way for faster development of promising biologically active molecules such as, high-throughput screening of commercially available compound libraries against the target of interest are readily available nowadays. The evaluation of ADMET profiles of drug candidates during preclinical development represents one of the crucial parts of the drug discovery process. Efficient profiling operations are now run in parallel to potency screening during lead optimization. In vitro assays are used during early drug development and high-throughput ADMET screens are available. The advantage of using in silico approaches over in vitro assays is that less investment is needed in resources, time and technology.1, 2

B. SYNTHETIC STRATEGY OF POTENT HETEROCYCLICS

Heterocyclic compounds constitute an important area in organic chemistry. Novel heterocycles designed and produced synthetically by organic chemicals are used as agrochemicals and pharmaceuticals. Nitrogen and oxygen containing heterocycles are by far the most widely occurring. For example.

1. The indole template is generally recognized as a privileged structure in medicinal chemistry,3 and in particular, oxindoles are important
constituents of natural indole alkaloids as well as drugs under development and also in the clinic. The oxindole motif is present in the anti-parkinson’s drug ropinirole, in non-opioid nociceptin receptor ligands and in the growth hormone secretagogues. In addition, the oxindole moiety constitutes a key structural element in several natural products including the antibiotic speradine and the cytostatin welwistatin. Compound 3,3-diaryloxindoles have been shown to possess mechanism-specific antiproliferative, antibacterial, antiprotozoal and anti-inflammatory activities. These compounds have also been used as laxatives and lead compounds for Ca\(^{2+}\)-depletion-mediated inhibition of translation initiation.

2. The **pyrazole** unit is the core structure in a number of natural products. Many pyrazole derivatives are known to exhibit a wide range of biological properties such as anti-hyperglycemic, antipyretic, sedative - hypnotic, anti-coagulant. Particularly arylpyrazoles were widely used in medicinal, pesticidal chemistry and recently reported to display non-nucleoside HIV-I reverse transcriptase inhibitory activity. Pyrazoles also possess anti-cancer and anti-inflammatory activities.

3. Considerable attention has been focused on spiro compounds in particular to **spiropyrolidines**, due to their interesting biological activities. They have been found to possess antimicrobial, antitumor, antibiotic, anticonvulsant, potential antileukaemic, local anaesthetic and antiviral activities. Furthermore, they also act as inhibitors of human NK-I receptor activity.

The structural diversity and biological importance of oxygen and nitrogen containing heterocycles like **spiro-oxindoles, pyrazoles and pyrrolidines** as anti-
cancer agents have made them attractive targets for synthesis in recent years through, Multicomponent reactions (MCRs) which are convergent reaction, in which three or more starting materials react to form a product, where basically all or most of the atoms contribute to the newly formed product. The result is clearly dependent on the reaction conditions solvent, temperature, catalyst, concentration, nature of the starting materials and functional groups. Such considerations are of particular importance in connection with the design and discovery of novel MCRs.12

**C. PRINCIPLES OF GREEN CHEMISTRY**

It is a philosophy of chemical research and engineering that encourages the design of products and processes that minimize the use and generation of hazardous substances.13 Attempts are being made not only to quantify the *greenness* of a chemical process but also to factor in other variables such as chemical yield, the price of reaction components, safety in handling chemicals, hardware demands, energy profile and ease of product workup and purification.

Paul Anastas, then of the United States Environmental Protection Agency, and John C. Warner, President of the Warner Babcock Institute of Green chemistry, Wilmington, Mass, developed 12 principles of green chemistry, which help to explain what the definition means in practice. The principles cover concepts such as:

- the design of processes to maximize the amount of raw material that ends up in the product
• the use of safe, environment - benign substances, including solvents, whenever possible
• the design of energy efficient processes
• the best form of waste disposal: not to create it in the first place.

The 12 principles are

1. It is better to prevent waste than to treat or clean up waste after it is formed.
2. Synthetic methods should be designed to maximize the incorporation of all materials used in the process into the final product.
3. Wherever practicable, synthetic methodologies should be designed to use and generate substances that possess little or no toxicity to human health and the environment.
4. Chemical products should be designed to preserve efficacy of function while reducing toxicity.
5. The use of auxiliary substances (e.g. solvents, separation agents, etc.) should be made unnecessary wherever possible and innocuous when used.
6. Energy requirements should be recognized for their environmental and economic impacts and should be minimized. Synthetic methods should be conducted at ambient temperature and pressure.
7. A raw material or feedstock should be renewable rather than depleting wherever technically and economically practicable.
8. Reduce derivatives – Unnecessary derivatization (blocking group, protection/deprotection and temporary modification) should be avoided whenever possible.
9. Catalytic reagents (as selective as possible) are superior to stoichiometric reagents.

10. Chemical products should be designed so that at the end of their function they do not persist in the environment and break down into innocuous degradation products.

11. Analytical methodologies need to be further developed to allow for real-time, in-process monitoring and control prior to the formation of hazardous substances.

12. Substances and the form of a substance used in a chemical process should be chosen to minimize potential for chemical accidents, including releases, explosions, and fires.

D. IN SILICO SCREENING APPROACH

Over the past decade a new field of research in silico biology is the use of computerized models to predict outcomes in biological studies has emerged. The concept of using in silico biology for the modeling of human disease is still in its infancy, but many eventually facilitate a better understanding and prediction of chronic human disease pathogenesis and ultimately help to design better and more rational approaches for developing and testing new drug candidates.

In general docking programs such as Auto Dock Vina generate multiple protein-ligand conformations, by sampling the ligand’s probable conformations in the binding pocket of the target protein using flexible ligand-ligand receptor docking. Scoring functions are used for docking calculations by their programs in an attempt to approximate the standard chemical potential of the system. Auto
Dock uses a free-field-bared scoring function approach to estimate binding affinities by calculating the non-bonded interactions based on position of a ligand and rank ligands by their predicted binding affinities.\textsuperscript{15}

E. EXPLORING BIOLOGY WITH SMALL ORGANIC MOLECULES

Small organic molecules have proven to be invaluable tools for investigating biological systems, but there is still much to learn from their use. To discover and to use more effectively new chemical tools to understand biology, strategies are needed that allow us to systematically explore ‘biological-activity space’. Such strategies involve analysing both protein binding of, and phenotypic responses to, small organic molecules. Finally, although most small molecules are not drugs, the occasional development of a small molecule into a drug can motivate researchers to use small-molecule tools to study biology.\textsuperscript{16}

Oxygen derived free radicals such as superoxide anions, hydroxyl radicals and hydrogen peroxide are \textit{cytotoxic} and give rise to tissue injuries. Therefore there is a need for more effective less toxic and cost effective synthetic antioxidants that can increase the antioxidant capacity in the plasma and reduce the risk of certain disease such as cancer, heart disease. It is considered must that the New Chemical Entities (NCEs) which can generate free radicals has to be screened for antioxidant activity before it is processed for other important biological evaluations like anticancer activity.

National Cancer Institute (NCI) has approximately screened around 2,00,000 test compounds over a period of 20 years. The identified anticancer
agents were grouped based on their mechanism of action. However, there is no evidence available for the treatment of majority of solid tumors. One of the reasons identified for such failures is the emergence of drug resistance. Further, the existing antineoplastic drugs are also highly toxic with lower therapeutic indices. These drugs frequently cause myelosuppression with a risk of hemorrhage and infection owing to their cytotoxic effect on rapidly proliferating tissues. The unavailability of safer and more specific anticancer drugs have led to the arousal of interest in our study as the present therapy is considered inevitable. This apparent failure necessitated the exploration of new avenues for finding effective anticancer agents.

A number of high-throughput phenotypic assays have been developed, including assays that measure cell viability or proliferation. Such assays measure the presence of intact cell membranes, the abundance of cellular energy (ATP concentration), or the presence of cellular reductases or esterases, which are found in nearly all cells. Recently, gene-expression signatures have been developed into high-throughput, phenotypic assays. By measuring the effects of small molecules on the appearance of this gene signature, it is possible to determine whether each compound changes the cell state (for example, induces differentiation of neutrophil precursors into neutrophils). Such new methods for automating and rapidly performing such measurements would be of value.\textsuperscript{17,18}

**F. FISSURE IN DRUG DISCOVERY**

Designing new chemical entities with which to perturb biological systems requires a systematic evaluation of the properties of existing tools. Although
large-scale measurements of the effects of small molecules on proteins and phenotypes can be challenging, the resulting data sets can be useful in probing biological-activity diversity. New ways to increase the complexity and sophistication of the \textit{in silico} methods and \textit{in vitro} assays that can be performed on vast arrays of molecules will prove valuable. In so doing, the study move closer to understanding the roles of the diverse molecules that are responsible for life, death and disease. The unavailability of safer and more specific anticancer drugs have led to the arousal of interest in our study as the present therapy is considered inevitable. This apparent failure necessitated the exploration of new avenues for finding effective anti-cancer agents.

To substantiate the above statements, the research attempt has been undertaken to frame the strategy as per the protocol of drug discovery to design, synthesis new chemical entities of diverse heterocyclic scaffolds like \textit{Spiro-oxindole (SV), Pyrazole \alpha - amino phosphonates (PV) and Acenaphthylene Pyrrolidine (VAZN)} analogues and screening for anti-cancer activity because highly reputed research organisations like CSIR-CLRI working on hard synthetic strategies, but have left many compounds without exploring for its biological activity. So there is a real scope for those compounds when hunted, to investigate as a anti-cancer drug candidate in future.

G. REFERENCES


