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On the cover: Medicinal chemistry covers a broad spectrum of research capabilities including the interaction of drugs and toxic substances with biological systems.

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Glycans are carbohydrate portions of glycoproteins which play important roles in protein folding, and protein solubility. They are often crucial for activities and functions of the proteins. Therefore, glycan analysis plays an important role in the bio-technological and pharmaceutical industries, as well as in clinical research.

Prof Sam Li and Dr Huatao Feng from the Department of Chemistry are working with Thermo Fisher Scientific / Life Technologies to develop a platform for glycan analysis. The project will leverage on current multi-channel capillary electrophoresis (CE) products dedicated for genetic analysis and extend the unique separation power and flexibility of these techniques for glycan analysis.

The high separation efficiencies and high-throughput capabilities achievable in multi-channel CE make it an attractive strategy when large numbers of targeted compounds are involved with extremely complicated sample matrices. Glycan analysis is one such example.

The researchers intend to investigate the performance of multi-channel CE separation using different separation mechanisms and sample pre-treatment methods to identify optimal methods for analysing complicated glycan samples. The novel techniques developed are expected to meet the demands for fast, powerful and economical analytical methods.

As the biological mechanism for the generation of glycan profiles is still not fully understood and glycan reference standards are not readily available yet, the research outcomes from this project are expected to create new applications and market opportunities for CE products in the bio-pharmaceutical industry and the clinical diagnostics market.

Prof Li reckons that this project will not only generate new strategies based on multi-channel CE for glycan analysis, but bring with it the capabilities for many other areas requiring rapid analytical methods with high selectivity and sensitivity.

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NUS launched a science and technology exhibition, entitled “Building Our Nation through Science and Technology” on 9 April 2015 in celebration of its 110th anniversary. The 10-day exhibition featured more than 20 exhibits showcasing the innovative ideas of researchers from the Faculty of Science, Faculty of Engineering and School of Computing. The displays were featured along five themes: Digital Nation, Multimedia Nation, Smart Nation, Sustainable Nation and Healthy Nation. Apart from viewing the demonstrations, prototypes and poster displays, visitors to the exhibition also interacted with some of the exhibits. The Faculty of Science showcased five exhibits in quantum technologies, plant genetics, food science, data science and biosensors.
Environmental effects of floating solar panels in reservoirs

In 2011, the Economic Development Board (EDB) and PUB, the national water agency, announced a pilot project to explore the use of floating solar panels in Tengeh Reservoir to generate electricity and optimise the space occupied by Singapore’s reservoirs. The Freshwater and Invasion Biology Laboratory led by Prof Darren YEO, NUS, is currently collaborating with PUB to examine the ecological impacts of the solar panels (specifically on the reservoir’s aquatic biodiversity) through field studies conducted at Tengeh Reservoir.

A variety of target organisms are being monitored, including fishes and macroinvertebrates, water-dependent birds, and zooplankton and phytoplankton to assess the potential effects across a broad coverage of biodiversity. Changes in numbers and diversity of these organisms are effective indicators of the potential impacts there might be from the solar panel installation/operation. The ecological knowledge will also contribute towards the conservation of reservoir faunal and floral communities, and is particularly important if more floating solar panels are implemented in the future.

“This study addresses a need for site- and context-specific data for analysis of the effects of the solar panels on biodiversity as well as on the environment in general,” says Prof YEO. He added that although potential impacts could be extrapolated from literature, these would be at best postulations, which cannot fully replace actual studies. In addition, the biodiversity data obtained from this study can complement concurrent studies by external collaborators (e.g. on chemical aspects such as water quality and physical aspects such as evaporation rates) to provide a more comprehensive picture of the effects of the floating solar panels.
Advanced fluorescent probes

Fluorescent sensors can be used to visualise almost any chemical substance, whether biological or environmental

“Give me a place to stand on, and I will move the Earth” — Archimedes.

Introduction

Since the earliest days man has walked the planet, human beings have collectively wondered when encountering something new, “What is this stuff?”, “What is it made of?” and “Is it safe to eat or drink?”, etc. With time, the level of inquiry has become more sophisticated and examples include “What are the constituents?”, “How much of each component?”, “Is there a disease vector present?”, etc. The analysis involved in answering these questions would require a considerable amount of time but increasingly, the world is demanding a rapid or instantaneous response. It is no longer acceptable to wait days or even hours for analytical results.

Small molecule fluorescent sensors offer a perfect approach to tackle this situation, owing to their non-invasive nature, high sensitivity, fine spatiotemporal resolution and emission / target tunability, etc. Prof Young-Tae CHANG has pioneered the “Diversity Oriented Fluorescence Library Approach” (DOFLA), a new paradigm for sensor development, and demonstrated its applicability to a broad range of biological and environmental analytes. Using combinatorial techniques, the dye collection already contains more than 10,000 compounds. This is one of the biggest toolboxes available for chemical sensor development.

DOFLA: Construction and Application

The construction of DOFLA hinges on one key term: diversity - both structural and photo-physical (Figure 1A). Starting from one fluorophore and a series of diversity elements, one can rapidly expand the library following a combinatorial approach, i.e. on one fluorophore there are three diversity sites and there are 10 diversity elements to attach to each site. One can produce \(10 \times 10 \times 10 = 1,000\) different fluorescent sensors using a single fluorophore. Following this concept, fluorophores with substantial structural differences and extensive emission range covering the visible light spectra were selected as a starting point. The molecules were purified and categorised according to their properties to build small libraries, each containing less than 100 compounds and embedded into 96-well plates.

An image-based high-throughput screening platform (Figure 1A) was developed to screen and generate prototypes. By irradiating the sensor plate with an excitation lamp, the background image of the library (with the excited fluorophores) is obtained using an off-the-shelf camera to serve as a reference image. After the addition of the analyte to the sensor plate, another image is captured and compared with the reference image. Changes in emission intensity or colour of the dye are identified as a hit and subjected to further analysis.

Environment, Food Safety and Social Security: What DOFLA Can Do

With this screening platform, a variety of analytes covering a large category of materials had been tested (Figure 1B):

- Environmental pollutants - multiple heavy metal ions;
- Food safety - caffeine, bisphenol A and milk fat; and
- Social security - date rape drugs (e.g. GHB and GBL).

All of these analytes require instrumental analysis which is both costly and time-consuming but the research team had developed fluorescent sensors that respond instantaneously to them. This has opened up the possibility of real-time monitoring even by non-technical people. The strength of the fluorescence technique is that by using
small molecules, one is able to observe clear colour changes visible to human eyes in the presence of targeted substances.

As an example, fluorescent sensors have been developed for the date rape drugs GHB and GBL. GHB “turns off” the fluorescence signal while GBL “turns on” the fluorescence signal. In practical situations such as in a bar, one could carry a test strip of sensors or perhaps apply them as nail polish. By spraying their beverages on them, one could easily detect if the drink has been adulterated.

Other than the GBL and GHB sensors, other fluorescent sensors are being identified using the screening platform. In Prof CHANG’s group, researchers are actively developing a fluorescence detector prototype (Figure 1A, part 4). This will be an affordable yet compact handheld device comprising an excitation lamp, a detector / photodiode and a signal output element. Possible applications of this portable device could include routine monitoring of the environment.

In the Body: DOFLA Sheds Light on Biomedical Imaging

Countless biological events occur in the human body every moment: signals transduce from the brain to every organ through the nervous system; tissues respond to both external and internal stimuli; cells undergo mitosis and apoptosis, etc. A goal in biological science has been to understand and clearly observe events taking place in the human body. However, this is not a trivial task even with state-of-the-art imaging techniques due to the extent of biological diversity (there are more than 200 types of cells in the human body, each containing more than 20 different organelles) and similarity among cells and tissues.

With the help of chemists, this biological dilemma is being resolved. During the past decade, DOFLA has produced unique and selective fluorescent sensors targeting more than a dozen cells, tissues and other biological events (Figure 2). Depending on the biological events, the screening format and targets can vary, but the key approach remains the same: select the target together with several bio-related analogs and achieve differentiation among these species. For instance, when targeting T cells, not only the related B cells are selected as an analog but it also includes the red blood cells, white blood cells and platelets. This capability allows medical researchers to identify key cellular tissues (e.g. neural stem cells, mesenchymal stem cells and pluripotent stem cells) and also important biological events (e.g. inflammation induced macrophage growth). Through the DOFLA approach, bioimaging probes are being developed to advance medical knowledge.

Future Prospects

Although a series of environmental and biomedical fluorescent sensors have already been discovered, a more important task would be to identify the common features in them which cause these fluorescence changes. With this knowledge, one day the research team would be able to claim: “Give me a fluorescent sensor, and I will illuminate the world”.

References
**New ways of making molecules**

Reducing waste in chemical synthesis by exploring the “borrowing-hydrogen” concept

**Introduction**

Synthetic chemistry, the subject on the preparation of diverse chemical structures from simple building blocks, is considered to be the “central science” as it provides the toolbox for various applied areas such as pharmaceuticals, chemical biology and material science. Due to resource constraints, the current trend in synthetic chemistry is not simply about preparing molecules of specific interest, but rather more on the methodology of how to prepare them in a highly efficient yet economical manner. Pharmaceutical companies are constantly looking for new innovations in the large-scale synthesis of pharmaceutical drugs as this would allow them to manufacture the drugs in a more efficient manner with less wastage, translating to a lower production cost.

The concept of “redox economy” which focuses on minimising synthetic steps that only adjust the oxidation state of the intermediates without generating structural complexity is an important consideration at the strategic level for chemical synthesis. Redox-neutral transformations that circumvent such redundant steps, reduce waste production and make efficient use of material resources are highly sought after since they also tend to be environmentally benign and sustainable processes.

“Borrowing Hydrogen” Catalysis

Figure 1 illustrates the process for the production of amines which have ubiquitous functionality in pharmaceuticals and many other fields. Traditionally, amines are produced from readily available alcohols through a multi-step process involving several processes: oxidation of an alcohol to a ketone, condensation with amine to imine followed by a reduction step. As both the oxidation and reduction steps in this process utilise stoichiometric reagents, waste products are generated.

The concept of “borrowing hydrogen” has been proven to be a much more efficient process. In this method, the alcohol starting material can be directly converted to the amine product using a one-pot synthesis strategy without any stoichiometric waste except for water (Figure 1a)!

![Figure 1: “Borrowing hydrogen” - an important catalytic concept.](image)

Figure 1: “Borrowing hydrogen” - an important catalytic concept.

This intriguing and highly attractive process has been enabled by the use of transition metal catalysis (Figure 1b). The transition metal catalyst enables the initial oxidation of the alcohol to an aldehyde/ketone. After the imine is formed via condensation of the ketone and amine, the metal complex that takes an “H₂” equivalent from the alcohol can now return it to the imine intermediate to form the amine product. Overall, the transition metal catalyst promotes efficient oxidation and reduction in a catalytic fashion (as it is regenerated after each cycle of redox chemistry) so no additional external reagent is required and no waste product is produced. Although this process has been under research for many years, only certain simple amines can be produced at industrial scale using this method. Also, the efficiency of the existing catalytic systems is far from ideal. Prof ZHAO Yu’s research group is working in this field to identify and develop more efficient catalytic systems.

**Enantioselective Amination of Alcohols**

A molecule is considered chiral if there exists another molecule that has identical chemical composition, but which is arranged in a non-superimposable mirror image. The chiral molecules that are mirror images of one another are known as enantiomers. Prof ZHAO’s group has recognised that if the last step in the catalytic cycle shown in Figure 1b can be rendered enantioselective (whereby one enantiomer is preferentially produced), chiral amines can be produced in high efficiency in this way.
ZHAO Yu obtained his B.S. degree from Peking University and his Ph.D. from Boston College. After working at the Massachusetts Institute of Technology as a postdoctoral associate, he was awarded the NRF Fellowship and joined the Department of Chemistry, NUS as an assistant professor in 2011. His research interests focus on catalyst and methodology development for efficient chemical synthesis. Please visit zhaoyu.science.nus.edu.sg for more information about his research group.

References


The human body is an environment with large biological molecules (e.g. proteins) present in a homochiral form (having only one enantiomer) and it is widely accepted that most drugs which are often chiral have to be prepared in one form of its enantiomers. The demand for chiral compounds, often as single enantiomers, has escalated sharply in recent years, driven particularly by the demands of the pharmaceutical industry. For these reasons, chiral amine synthesis has been an extremely important goal in organic synthesis. The preparation of amines using the borrowing hydrogen method, while being highly economical, was never used for chiral amine preparation until their work was reported in Angewandte Chemie in early 2014 (Figure 2; [1]).

As shown in Figure 2, the starting alcohol (in a racemic form having equal amounts of left- and right-handed enantiomers) and the primary amine can be transformed into the chiral amine product with high yield and enantioselectivity without the need for any additional external reagent. The chiral iridium complex (denoted as compound 2 in Figure 2) can catalyse the redox chemistry effectively. A chiral acid (denoted as compound 1 in Figure 2) also proved to be an essential compound: it not only promotes the condensation of the ketone intermediate with amine to form the imine, but also helps control the enantioselectivity in the reduction step. The efficiency of this system, however, is far from being practically useful. The high loading of expensive catalysts is a significant limitation.

Recently efforts at Prof ZHAO’s laboratory have identified a much more efficient iridium system as well as an alternative iron-based complex for related transformations, which are much more attractive from an economical and process sustainability viewpoint.

Recently, Prof ZHAO’s group has extended the previous discovery to a dynamic kinetic asymmetric amination process (Figure 3; [2]). In this study, the alcohol substrates possess two stereogenic centres and exist as a mixture of four isomers. By performing this asymmetric amination reaction, only one isomer out of four possible products was obtained! This highly selective synthesis is enabled by the effective conversion of all isomers of the starting material to an achiral intermediate followed by stereoselective construction of the amine product by the catalyst.

The research to develop highly efficient methods for synthesising chiral compounds will inevitably lead to more efficient synthesis of drug candidates and commercial drugs to meet the demands of the pharmaceutical industry.
Bacterial outer membrane biogenesis

Outer membrane assembly in Gram-negative bacteria, specifically lipid transport, is an ideal target for the discovery of novel antibiotics.

Introduction

Since the advent of antibiotics after the WWII, fatality rates due to bacterial infections have decreased sharply and remained at a low level. Over the past few decades, however, bacterial pathogens that are resistant to many clinical antibiotics have emerged, rendering treatment against these bacterial infections ineffective. The rapid rise in the incidences of multi-drug resistant pathogens, termed “superbugs”, poses a major threat to global public health. In 2011, an outbreak of one such superbug, *Klebsiella pneumoniae* carbapenemase (KPC), which is resistant to even the last resort drug used in the clinic, killed seven individuals at the National Institute of Health in the United States. KPC belongs to a class of bacteria known as the Gram-negatives, which include more commonly known strains such as *Escherichia coli*, *Salmonella enterica* and *Pseudomonas aeruginosa*. Infections caused by Gram-negative bacteria are particularly difficult to treat due to the limited repertoire of antibiotics that can penetrate the unique cell envelope of these organisms, and are hence effective against them. Beyond this intrinsic resistance to many antibiotics, Gram-negative bacteria become multi-drug resistant by acquiring ways to destroy antibiotics or pump them out of the cell. In the case of KPC, an enzyme that destroys the carbapenem antibiotics was acquired; no other drug is now available to treat these infections. This episode underscores the urgent need to develop new strategies to combat infections caused by these superbugs.

The Gram-negative Bacteria Cell Envelope and Antibiotic Resistance

The cell envelope of Gram-negative bacteria consists of two lipid bilayers: an inner membrane that encloses the aqueous cytoplasm and an outer membrane that faces the extracellular environment (Figure 1). Between these two membranes is a second aqueous compartment known as the periplasm, which contains peptidoglycan (cell wall) that determines the shape of the bacterial cell. This unique double-membrane envelope renders Gram-negative bacteria intrinsically more resistant to many external insults. The outer membrane is an asymmetric lipid bilayer in which the inner and outer leaflets are composed of different lipids, namely phospholipids and lipopolysaccharides, respectively. In the outer leaflet, lipopolysaccharides, which are negatively-charged glycolipids that contain six hydrocarbon chains each, pack together in the presence of positively-charged ions to form an impervious polyelectrolyte with gel-like characteristics in the hydrophobic interior. The resulting lipopolysaccharide leaflet exhibits markedly decreased fluidity and makes the outer membrane a very effective permeability barrier.

The outer membrane is essential for the survival of most Gram-negative pathogens. In addition, compromising outer membrane integrity can enable the use of many antibiotics currently only effective against the other major class of bacteria, the Gram-positives (which lack the outer membrane). Therefore, the molecular machines that build the outer membrane have great potential as new targets for antibiotic discovery. Furthermore, the machines that are known to assemble outer membrane components are highly conserved among Gram-negative bacteria, making them possible targets for broad-spectrum antibiotics. In this regard, a mechanistic understanding of how the outer membrane is assembled would be extremely valuable.

Gram-negative Outer Membrane Assembly

The Gram-negative outer membrane presents a simple system for studying membrane assembly. All four major outer membrane components (lipopolysaccharides, phospholipids, integral β-barrel proteins and outer membrane lipoproteins) are synthesised in the cytoplasm or the inner membrane and must be transported across the periplasmic space to be incorporated into the outer membrane (Figure 2). The unique challenge of the Gram-negative organism is to assemble the outer membrane outside the cytoplasm, in an environment that lacks an obvious energy source (i.e. adenosine triphosphate (ATP)). Owing to their hydrophobic nature, outer
membrane components do not freely diffuse across the aqueous periplasm. Separate machinery that transports outer membrane lipoproteins across the periplasm (the Lol system) or assembles β-barrel proteins into the outer membrane (the Bam complex) has been characterized extensively. Both these outer membrane components are transported across the periplasm via soluble intermediates bound by protein chaperones. Furthermore, the trans-envelope machinery that transports lipopolysaccharides across the periplasm and assembles them into the outer leaflet of the outer membrane (the Lpt complex) has been studied and characterized as part of Prof. CHNG Shu Sin’s previous work. Surprisingly, very little is known about how phospholipids, the most basic membrane building block, get to its final destination in the inner leaflet of the outer membrane.

**Understanding Phospholipid Trafficking in Gram-negative Bacteria**

Research in Prof. CHNG’s laboratory focuses on understanding phospholipid transport in Gram-negative bacteria. Molecular machinery involved in phospholipid transport in the context of outer membrane assembly has not been identified. Early research work did, however, provide information on some requirements of phospholipid transport in the model Gram-negative organism *Escherichia coli*. First, the phospholipid composition is different between the inner and outer membranes. Next, it is known that transport of phospholipid from the inner to outer membrane is dependent on the proton motive force, and not cellular ATP (as is for lipopolysaccharides or lipoproteins). Furthermore, it has been established for a long time that, unlike outer membrane lipoprotein and lipopolysaccharide assembly, phospholipid transport between the inner and outer membranes is bidirectional (Figure 2). The cell requires forward transport to deliver phospholipids to the outer membrane. Backward phospholipid transport may be important in lipid homeostasis at the outer membrane, i.e. controlling the levels of phospholipids relative to lipopolysaccharides, in order to form a stable asymmetric lipid bilayer. It is not known if forward and backward phospholipid transport occur via the same pathway. Nonetheless, phospholipid molecules have been proposed to be shuttled between the inner and outer membranes via soluble chaperone-phospholipid intermediates (analogous to outer membrane lipoprotein trafficking), through protein bridges (similar to lipopolysaccharide transport), or via membrane bridges (akin to stalk formation during vesicle fusion) that may provide the route for diffusion of phospholipids between the two membranes. How phospholipids are transported between the inner and outer membranes remains an important unsolved problem. A mechanistic understanding of these processes can surely be exploited in the design and development of novel antibiotics against Gram-negative pathogens. In addition, membranes form the basis for life, physically defining cells and organelles, and modulating the chemical environments within these compartments for optimal metabolism and growth. How phospholipids are transported to and from the bacterial outer membrane may serve as a good model for understanding membrane/organelle biogenesis in other systems, such as eukaryotic cells, and eventually provide insights into the formation of the modern cell as we know it.

![Figure 2: Processes involved in outer membrane (OM) assembly. OM lipoproteins are targeted to the OM via the Lol chaperone system while lipopolysaccharides (LPS) are transported and assembled via the Lpt protein bridge. The Bam complex folds and inserts β-barrel proteins into the OM. How phospholipids (PL) are transported from the inner to the outer membrane (and back) is not known.](image)

Shu Sin CHNG obtained his Ph.D. in Chemistry at Harvard University (MA, USA) in 2010, where he was awarded the Eli Lilly Organic Chemistry fellowship and the Christensen Prize for outstanding research achievement. After a short post-doctoral stint at the Harvard Medical School (MA, USA), he joined the Department of Chemistry, NUS as an assistant professor in August 2011.

References


Platinum agents with new modes of action
Engineering new platinum-peptide prodrugs for targetted chemotherapy

Cisplatin, an Effective Anticancer Drug

It was 1999 when Lance Armstrong won the first of seven Tour de France races, the most celebrated and prestigious of all the peloton races, but he would ultimately be forced to relinquish these titles following his admission of doping. But three years prior, Armstrong was diagnosed with advanced metastatic testicular cancer and the disease had spread to his brain, lungs and abdomen. At 25 years of age, this was devastating news, given the poor prognosis of his condition. It put his promising competitive cycling career in jeopardy. In the ensuing year, Armstrong underwent aggressive chemotherapy with a cocktail of chemotherapeutic drugs, administered together with surgery. In 1997, after a year of treatment, he was declared cancer-free. The chemotherapeutic drug that rid him of the cancer was cis-diamminedichloroplatinum(II), more commonly known as cisplatin.

Since the serendipitous discovery of its anti-tumour properties by Barnett Rosenberg in 1968, cisplatin has become one of the most important agents in the clinician’s arsenal against cancer. Together with carboplatin and oxaliplatin, they constitute a class of platinum-based drugs being used in almost half of all cancer chemotherapeutic treatments (Figure 1). Before cisplatin, testicular cancer was a deadly disease to an adult male, with low survival rates at about 10%. Since the introduction of cisplatin, however, survival rates have increased significantly to almost 80%, making testicular cancer amongst the most managed malignant tumour disease. Second generation platinum drugs carboplatin and oxaliplatin are now being applied routinely against ovarian and colorectal cancers, respectively. Yet despite their widespread use in the clinic, platinum drugs are limited by their high toxicity and severe side-effects, often dose-limiting, as well as incidences of drug resistance. These limitations, in spite of their clinical successes, provide the motivation for the search of new metallodrugs with different modes of activity.

Platinum(IV) Prodrugs of Cisplatin

From a chemical structure standpoint, cisplatin is a highly unusual drug because it does not contain a single carbon atom! Not surprisingly, its mechanism of action against cancer cells stems largely from its inorganic metallic character. Cisplatin enters the cancer cell largely intact through diffusion or transport mechanisms. Once within the cell, it undergoes a process of chemical aquation whereby the chloride ligands are substituted by water. Aquated cisplatin is highly reactive and it binds DNA to form stable DNA lesions that interfere with DNA processing mechanisms, particularly RNA transcription. Consequently, the cell attempts to repair and remove these lesions but if the damage is too extensive, programmed cell death is initiated. Because cisplatin cannot discriminate between diseased and healthy cells due to the lack of selectivity, systemic toxicity is high.

To address this poor selectivity, one way is to engineer cisplatin so that it can actively seek out and destroy cancer cells. Homing peptides that bind specifically to receptor protein biomolecules overexpressed on cancer cells, not on healthy cells, may be utilised to direct the cell-killing cisplatin payload to targetted cells. To realise this concept, a synthetic strategy that facilitates the conjugation of peptides to platinum(IV) prodrugs of cisplatin was developed [1]. Platinum(IV) prodrugs belong to an interesting class of compounds that are non-active in their native form but they can be converted to cisplatin when subjected to a reducing environment, typically found within cells. The new synthetic strategy allowed the conjugation to take place under mild conditions and in very high yields, a marked

Figure 1: Platinum drugs in clinical use.

Figure 2: Induction of targetted necrosis in HER2-positive cancer cells.
Wee Han ANG received his Ph.D. from the École Polytechnique Fédérale de Lausanne (EPFL) in 2007, where he was conferred the EPFL Doctorate Award for his doctoral work. He pursued postdoctoral research work at the Massachusetts Institute of Technology on the NUS Overseas Postdoctoral Fellowship. He has been an assistant professor in the Department of Chemistry since joining NUS in 2009.

References


The importance of being “drug-like”
Systematic modification of molecules to discover better drug candidates

Introduction

In 1998, the International Union of Pure and Applied Chemistry defined Medicinal Chemistry as a multidisciplinary science that is focused on drugs (“biologically active agents”). It encompasses their invention, discovery, design, identification, preparation, study of their metabolism, construction of structure-activity relationships and interpretation of their mode of action. Fifteen years on, one would ask if this definition is still relevant, especially since the intervening period has seen an onslaught of buzz words like chemical biology, chemo-genomics, chemical proteomics, chem-bioinformatics into the medicinal chemistry lexicon, with each staking a claim as an independent field or discipline related to drug discovery and development. These are undoubtedly useful technologies that will push forward the boundaries of drug discovery. Some may even attribute the recent upswing in FDA drug approvals to the contribution by these approaches. The long gestation period for drug discovery suggests that these approvals, in all likelihood, originated from studies initiated in the early 2000s. A significant event which took place then was the proposal by a group of medicinal chemists from the global pharmaceutical company, Pfizer of a mnemonic for identifying molecules which are likely to have limited absorption in humans because of suboptimal solubility or permeability. These rules (“Rule of five”) which are simple and easy to use, moved rapidly up the hierarchy of medicinal chemistry concepts. Widely applied and commonly misused, the “Rule of five” defines certain necessary, but not sufficient, conditions for a drug candidate. By drawing attention to the need for “drug-like” features in a candidate molecule, it has helped improved the overall quality of drug candidates. Although medicinal chemists have not been entirely weaned off their addiction to “potency”, they now realise the need to strike a balance between the physical attributes and drug activity. Only then will their candidate compounds achieve pharmacologically effective concentrations in the body when administered orally (the preferred route).

Drug candidates which have achieved this “sweet spot” have a higher chance of clearing early clinical trials and advancing down the development pipeline. In the following paragraphs, two case studies are presented to illustrate approaches to incorporate “drug-like” features into candidate molecules with the necessary potency but not the required physical attributes.

Improving “Drug-like” Properties of Cysmethynil

The first case study centres on inhibitors of the enzyme Isoprenylcysteine Carboxylmethyltransferase (ICMT). ICMT plays an important role in the processing of proteins for control of cell growth and the onset of cancer. Inhibition of ICMT results in events that mitigate uncontrolled proliferation of cancer cells. By developing agents that specifically inhibit ICMT, a clearer understanding of the effect of ICMT on other oncogenic signalling pathways can be obtained. This could potentially yield clinically useful agents for the treatment of cancer.

Cysmethynil (Figure 1) is a small molecule that specifically inhibits ICMT. In the presence of cysmethynil, the growth rate of cancer cells slows down and ultimately ceases, followed by cell death. When administered to mice with implanted tumours, cysmethynil causes regression of tumour sizes. In spite of its promising effects on cancer cells, cysmethynil has several limitations. It has limited potency and lacks several “drug-like” features (i.e. good solubility, weak binding to plasma protein) which would hamper its development as a clinically useful drug.

To address these problems, a detailed structure-activity relationship study was carried out and it revealed two interesting findings, namely that the carboxamide group in cysmethynil (position 3) could be replaced by an amino group and that the m-tolyl group at position 5 was optional for...
Mei Lin GO obtained her Ph.D. in the area of antimalarial drug discovery from NUS. She is an associate professor in the Department of Pharmacy, NUS and is currently the Deputy Head in charge of Education at the department. She is also a member of the Drug Development Unit at NUS which works on the development of new drug candidates. Her research interests lie in the area of design and synthesis of bioactive small molecules with drug-like properties, establishing structure-activity correlations and mode of action studies by biochemical and pharmacological approaches. She has authored more than 80 peer reviewed publications and is the co-inventor of three patents on biologically active entities.

Please visit http://www.pharmacy.nus.edu.sg/staff/phagoml/main.html for more information about her research work.

activity (Figure 1). The latter implied that functionalities moderating physical properties can be introduced at position 5 with minimal effects on ICMT inhibition. Two compounds (2 and 15) were then synthesised and evaluated. Compound 2 was comparable to cysmethynil in terms of ICMT inhibition but it was significantly more potent in curtailing the growth of cancer cell lines. In spite of the inclusion of the polar amino group, solubility of compound 2 was poor, likely due to the retention of the non-polar m-tolyl ring at position 5.

Hence, modifications at position 5 were explored and this resulted in compound 15 which now bears the polar heterocyclic aminopyrimidine ring at position 5. Compound 5 exceeded expectations in that it was not only 100x more soluble than cysmethynil and compound 2, but was also a more potent inhibitor of ICMT. When tested in tumour-bearing mice, compound 15 was well-tolerated and attenuated tumour growth. Both compounds 2 and 15 were subsequently patented.

Making Isoindigo Water Soluble

The second case study is based on a group of compounds called bisindoles and specifically, on one member meisoindigo which has been used for the treatment of a type of leukemia (chronic myeloid leukemia) in China. Reports on the mode of action of meisoindigo suggest that it has several targets including DNA. A significant drawback of meisoindigo is its poor aqueous solubility. This is expected from the “brick-dust” nature of its scaffold which would promote tight crystal packing and hamper solvation. Tellingly, most literature reports on this scaffold focus on modifications to enhance its solubility. Unfortunately, in most instances, greater solubility was achieved at the expense of biological activity. It became clear that designing biologically active bisindoles with acceptable solubility-permeability profiles and potent activity would be a challenge.

Investigations into structure activity relationships identified key positions on the isoindigo scaffold that were sensitive to modification. The most significant finding was that the N-methyl of meisoindigo could be replaced by N-aryalkyl side chains with gains in activity. As these modifications would exacerbate poor solubility, an alternative approach was to append solubilising basic heterocycles to the isoindigo scaffold.

Compound 5-4, a water soluble analog was developed by introducing a 4-methylpiperazin-1-ylethyl side chain. Compound 5-4 was modestly more potent than meisoindigo on a leukemic cell line but with greatly enhanced aqueous solubility. When administered to tumor-bearing mice, compound 5-4 increased survivability. Under similar experimental conditions, meisoindigo had no demonstrable effects on tumour growth. The in vivo efficacy of compound 5-4 could be attributed to its improved physicochemical profile. It is proposed that its modest cell-based potency has been compensated or even overcome by its favourable solubility profile. Mechanistically, compound 5-4 mimicked meisoindigo in its effects on cell cycle arrest and apoptosis.

Taken together, these case studies illustrate how well-reasoned and relatively simple structural modifications can be used successfully to address problems related to “drug-like” properties.
A lifetime of contributions to Science

Professor Louis CHEN obtained his B.Sc. (Hons) in Mathematics from the University of Singapore (now National University of Singapore) in 1964 and his M.S. and Ph.D. in Statistics from Stanford University in 1969 and 1971, respectively. During his 43 years with the University, Professor CHEN was Head of the Department of Mathematics (1996 - 2000) and Head of the Department of Statistics and Applied Probability (2002 - 2004). From July 2000 to December 2012, he served as the founding Director of NUS’ Institute for Mathematical Sciences, which he built into one of the finest in the world. Internationally, Professor CHEN was elected President of the Bernoulli Society for Mathematical Statistics and Probability (1997 - 1999) and President of the Institute of Mathematical Statistics (2004 – 2005). In his seminal work on Poisson approximation in 1975, he developed a method of approximating the probabilities of occurrences of dependent rare events. This method, which is now commonly known as the Chen-Stein or Stein-Chen method, has found wide-ranging applications in many fields, with lasting impact in science and technology. It has developed into one of the most important areas of discrete probability.

Professor Chong Kim ONG from the Department of Physics joined NUS in 1981. For his outstanding research work, he received the NUS Outstanding Researcher Award in 2010. As a physicist, he has a charismatic ability to inspire and motivate young scientists in conducting basic research on functional materials, and exploring their application in the development of new devices. The sophisticated tools and facilities in his research laboratory have been specially designed and built in-house based on his original research, at low cost. Many of these are not available commercially. Professor ONG is a tireless researcher and educator; he has published more than 450 papers in international journals and authored a book entitled “Microwave Electronics: Measurement and Materials Characterisation”. He is an esteemed fellow of the Singapore Institute of Physics and the Institute of Physics, UK and has served on the advisory boards of several international journals. In the local academic community, Professor ONG was the Vice-President of the Singapore National Academy of Science (1998 - 2000) and the President of the Institute of Physics, Singapore (1996 - 2000).

Professor Frank WATT moved from the Department of Nuclear and Particle Physics, Oxford University, where he founded and was director of the Oxford Scanning Proton Microprobe group, and joined the Department of Physics, NUS in 1992. In Singapore, he established the Research Centre for Nuclear Microscopy and pioneered the techniques of nuclear microscopy and proton beam writing lithography. The Research Centre for Nuclear Microscopy was expanded and renamed the Centre for Ion Beam Applications (CIBA). He remained director of this Centre until 2011. Professor WATT extended CIBA's position as the premier nuclear microscopy and proton beam writing facility worldwide. He initiated the conference series (Nuclear Microprobe Technology and Applications) which is now the major conference in this field. The 15th conference in this series was held in Padua, Italy in 2014. He also initiated the International Workshop series on Proton Beam Writing, the first of which was held in Singapore in 2004. In 2010, he was awarded the Institute of Physics Singapore President’s Medal for his outstanding research achievements. He has received numerous faculty awards for teaching, as well as the NUS Annual Teaching Excellence Award (ATEA) in 2011.