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ICBS 2021 – Looking Towards the Next Decade of Chemical Biology

Clinton G. L. Veale¹; Arindam Talukdar², Boris Vauzeilles³

Hosted virtually by Emory University, through its co-chairs, Haiyan Fu (Emory University), Sally-Ann Poulsen (Griffith University), Huw Davies (Emory University) and Alanna Schepartz (UC Berkeley), the International Chemical Biology Society (ICBS) held its 10th Annual Conference from 10-13th November 2021. Building on the progress from previous meetings,¹⁻⁷ the theme of this year's event "*Looking Towards the Next Decade of Chemical Biology*" blended a diverse array of established and early career speakers working under the cross-disciplinary umbrella of Chemical Biology. In what was a tightly packed schedule, ICBS 2021 consisted of 14 focused sessions and two trainee presentation sessions, in which a total of 69 'podium' presentations were given. In addition, attendees enjoyed six invited keynote lectures and the ICBS Global Lectureship Award. The scientific lecture schedule was complimented by forums discussing the early career chemical biology space, and the next decade of chemical biology respectively, in addition to two networking and professional development sessions, live poster sessions, and the ICBS business meeting,

DAY 1: PRE-CONFERENCE TRAINEE SYMPOSIUM

Trainee Presentation Session 1. In appropriate harmony with the theme of ICBS 2021, and looking toward the future of chemical biology, the pre-conference trainee symposium was organized and hosted by members of the ICBS trainee chapters. The first session of the trainee symposium, chaired by **Aylin Binici (Max Planck Institute of Molecular Physiology)** and **Kun Qian (Emory University)** featured short presentations from six young chemical biologists. The first presenter, **Brodie Bailey (Walter and Eliza Hall Institute of Medical Research)** presented their work in optimizing and biologically characterizing an exciting new class of anti-plasmodials, with slow to moderate activity against the erythrocytic stage,⁸ as well as their ongoing work in identifying and validating a putative mitochondrial carrier as a potential biological target for this class. **Guoliang Zhu (East China University of Science and Technology)** described their recent work where mining of their final genome

library and molecular networking-based dereplication, resulted in the identification of acyl coprogen siderophores as a promising class of non-bactericidal small molecules, capable of preventing *Pseudomonas aeruginosa* infection in a *Caenorhabditis elegans* infection model.

Catia Pierotti also from the **Walter and Eliza Hall Institute of Medical Research** provided details of their study, where the employment of a range of target engagement strategies identified a small molecule capable of interacting with three key effectors involved in necroptosis, MLKL, RIPK1 and RIPK3. This resulted in the inhibition of necroptosis in vitro, which translated into protective activity in a murine model of systemic inflammatory response syndrome (SIRS).⁹ **Laura Depta (Technische Universität Dortmund)** discussed the role of protein kinase Akt in cell signaling and diseases pathogenesis, and their efforts to develop cellular systems capable of discerning isoform-selective Akt inhibitors. This model system in conjunction with a structure-based drug design campaign, resulted in the development of a cohort of isoform-selective covalent-allosteric Akt inhibitors, which can act as probes to tease out specific roles of each isoform.¹⁰ Cholesterol transport proteins (CTP), are responsible for intracellular cholesterol transport, as well as playing an important role in processes such as autophagy, and viral entry. However, the lack of suitable selective inhibitors has hampered efforts to elucidate a broader picture of their cellular function. Accordingly, **Thomas Whitmarsh-Everiss (Technical University of Denmark)** presented their scaffold fusion approach leading to the identification of a diverse cohort of sterol-inspired compounds which inhibit the Aster family of CTPs, as well as displaying excellent selectivity for Aster-C, over the A and B isoforms.¹¹ **Receptor Activity Modifying Proteins (RAMPS)** play an important role as allosteric modulators of G-Protein Coupled Receptors (GPCR), and their downstream biological functions. Therefore, the identification, and characterization of these PPIs is important in the GPCR drug discovery space. To this end, **Ilana Kotliar (The Rockefeller University)** described the development of a multiplexed suspension bead array immunoassay, through which they mapped the protein-protein interactions (PPIs) between secretin-like G-protein coupled receptors and their corresponding partners bringing new insight into this important area.¹¹

Career Talk and Panel Discussion. In what was an extremely informative forum, **Shannon Miller (Harvard University)**, moderated a career talk and panel discussion with five early-

career chemical biologists, **Kali Miller (Stanford University)**, **Brooke Bullock Lao (Pfizer)**, **Lao, Guillaume Garivet (BASF SE)**, **Jiafeng Geng (Emory University)** and **Adam Kositzke (LabCorp)**. Through this interactive session, the speakers gave useful insights into their paths through their PhDs, and post-doctoral space and moved onto their experiences in navigating an early career, including tips for interviewing, hiring cycles and identifying the correct employment opportunities.

Trainee Presentation Session 2. The second trainee presentation chaired by **Dacheng Fan (Emory University)** featured an additional five short presentations from young chemical biologists. To open the session, **Joomyung Jun (Massachusetts Institute of Technology)** presented their progress toward the chemoselective bioreversible esterification of acidic residues, in order to enhance the traceless entry of therapeutically relevant proteins into the cytosol.¹² Their presentation went on to disclose the expansion of this approach, to allow further late-stage modification of the protein labels. **Anneliese Gest (University of California, Berkeley)** followed with their work in developing methodology to visualize and quantify the membrane potential of neurons,¹³ and progress toward neuron specificity through attachment to genetically encoded cell surface receptors.¹⁴ **Zhi (Lindsey) Lin (Harvard University)** provided insight into the development of photo-lenalidomide, as a chemical proteomic tool to identify the direct targets of lenalidomide. Through this approach, they identified eIF3i as a new target, which forms a ternary complex with lenalidomide, and CRBN but, is not ubiquitinated, or degraded. Furthermore, they found that sequestration of eIF3i was critical for the anti-angiogenesis phenotype of lenalidomide.¹⁵ The session was closed by **Brendan Dwyer (The Scripps Research Institute)** who described the total synthesis of naturally occurring anticancer curcusions. Through the synthesis of alkyne tagged analogues, and a biotin tagging/ streptavidin enrichment strategy, they identified the 'difficult to drug' oncoprotein BRAT1 as the target of the curcusions,¹⁶ as well as an undisclosed E3 ligase, which is involved in mitochondrial dynamics and apoptosis, whose promise as a new E3 ligase for PROTAC development was demonstrated.

Keynote Lecture 1. The first keynote session chaired by **Chad Altobelli (UC San Francisco)** and delivered by **Jason Gestwicki (UC San Francisco)** provided a fascinating end to the opening day, highlighting their recent progress in characterizing C-terminal PPIs through two case studies. In the first case study, they explored how alterations to the HSP70 and HSP90 C-terminal motifs may influence interaction to the E3-ligase co-chaperone, CHIP. Through a positional scanning synthetic combinatorial library, they revealed that CHIP could bind a range of peptide sequences with greater affinity than those found on HSP70 and 90. Having hypothesized that this phenomenon allows CHIP to interact with a wider range of proteins, they determined that latent C-termini, of cleaved tau and caspase-6 exposed through caspase activity, interact with CHIP, leading to their ubiquitination. This finding has important implications for understanding the pathogenesis of Alzheimer's disease as well as the link between post-translational modification and proteostasis.¹⁷ Through a similar approach, the second case study discussed the elucidation of a conserved peptide motif, which mimics the C-terminal binding motifs of 20s binding proteasome activators. Inclusion of this motif in a functional chimera, resulted in activation and gate opening of the 20s ribosome. The ability to activate the 20s ribosome is an attractive pathway in combatting neurodegenerative diseases.¹⁸

DAY 2

Opening Remarks. To open the main conference, delegates were welcomed by ICBS President, **Jonathan Baell, (Monash Institute of Pharmaceutical Sciences)**, and President-Elect **Zaneta Nikolovska-Coleska, (University of Michigan)**. They emphasized the importance of the 10 year milestone of ICBS meetings, and as part of the theme 'Looking towards the next decade of chemical biology' hoped that the community will emerge stronger from the present pandemic situation and, further hoped that the next ICBS meeting will be in person. On behalf of the organizing committee and ICBS2021, **Hian Fu** provided a historic overview of ICBS and its aim to promote global research at the interface of chemistry and biology. This included an overview of past meetings, as well as key partnerships formed over the years, such as those with EU OPENSREEN, ECBS, the **E**uropean **F**ederation of

Medicinal chemistry and Chemical biology (EFMC), the ICBS Pharma-LifeSci Advisory team in 2018 as well as the establishment and spread of ICBS student trainee chapters.

Joint Session 1: Global Advances in Chemical Biology. This opening session, chaired by **Cheryl Arrowsmith (University of Toronto, Princess Margaret Cancer Centre)** sought to take stock of the advances in chemical biology over the past decade and look forward to the future. The first presenter, **Bahne Stechmann, (EU-OPENSREEN)** elaborated on their European Infrastructure of Open Screening Platforms for Chemical Biology, operating from their 24 partner sites jointly funded by research ministries of eight member countries. Through joint grant applications, training, sharing of compound collections, common operational procedures, transparency, shared IP practices and collaborations with scientists from both academia and industry they have built a strong scientific community, which promotes translational research. The next speaker **Stephan Schurer (University of Miami)** discussed the US government-sponsored program; **Illuminating the Druggable Genome (IDG)** whose goal was to improve understanding of properties and functions of proteins that are not well studied within commonly drug-targeted protein families.¹⁹ This included the development of a data and resource-generating center, resource dissemination and outreach center, and knowledge management center with highly interconnected informatics tools, to facilitate collaboration with academic investigators as well as the biopharma industry. This success was illustrated through three case studies. The first from the labs of Roth and Soichet focused on Illuminating the druggable GPCR,²⁰ and the generation of commercially available probes for orphan and understudied GPCRs. The second case study described work on the druggable kinome, where they built a valuable **Kinase Chemo-Genomic Set (KCGS)** consisting of more than 1200 kinase inhibitors as a useful starting point to identify new kinase inhibitors.²¹ The final example discussed the development of the computational hit finding platform; **Computational Hit finding Critical Assessment of Computational Hit finding Experiments (CACHE)** which is a public benchmarking project which aims to accelerate the output, improve the reliability, and facilitate dissemination of computational chemical biology.²²

Yan Zhang (National Natural Science Foundation of China) gave a history and overview of the chemical biology initiatives currently underway in China and included aspects on

initiation, targeting policy and general information on chemical biology research. From a Chinese perspective, a project entitled 'chemical problems in the life process' which began in 1991 from the lab of Tang Youqi was key in initiating chemical biology research in China. After 2000, the number of interdisciplinary chemical biology projects funded by the NSFC under their 'major research plan' increased dramatically. Between 2007 – 2015 chemical biology funds were focused toward the utilization of small chemical probes for signal transduction, dynamic modification of bio-macromolecules and chemical interventions. In line with the growth of chemical biology in China, since 2018 the NSFC has earmarked a separate budget for the chemical biology division for promoting chemical biology, as well as the establishment of the department of interdisciplinary sciences in November 2020. **Bridget Wagner (Broad Institute)** described the educational opportunities that ICBS is undertaking through the recently formed education committee, whose aim is to increase ICBS' presence across the globe and to provide networking opportunities for trainees and introduce them to industry scientists or academic mentors. The key activities included arranging ICBS pre-conference trainee symposia which provide opportunities for postdoctoral and graduate students to discuss their on-going research and the career talk and panel discussion to foster the next generation of chemical biologists. Related to the trainee symposia is the increasing number of trainee chapters. The currently active trainee chapters are located in Dortmund, Atlanta, Boston, San Francisco, Michigan, Melbourne and Shanghai and interact through monthly seminars. To start a chapter please contact icbs@chemical-biology.org for more information. **Cheryl Arrowsmith**, who leads the Toronto site of the Structural Genomics Consortium (SGC), discussed the development of the SGC in the context of its mission to accelerate the discovery of new medicines through pre-competitive, open science. Having begun in 2004 as a structural biology programme focusing on the kinase, epigenetics, and ubiquitination signaling, the SGC now includes ~ 150 scientists with a global network of more than 300 collaborators, attracting funding from pharmaceutical companies, charities, and government agencies, and is regarded as a model public-private partnership. Their lecture touched on several success stories, including the development of over 50 epigenetic chemical probes with a special emphasis on the methyltransferases,²³ the **Donated Chemical Probes (DCP)** programme where comprehensively characterized chemical probes, donated by several

pharmaceutical companies, are curated and made available for the purposes of open science²⁴ and the chemical probes portal (www.theSGC.org/chemical-probes/), a public resource established by SGC, containing 350 small molecule inhibitors, agonists, antagonists, PROTACS and degraders.²⁵ Looking toward the future, SGC is affiliated with the Innovative Medicine Initiative (IMI) funded EUbOPEN project, whose goal is to develop high-quality tool compounds for 1000 human proteins, in line with the objective of Target 2035.²⁶

Keynote Lecture 2. The second keynote session chaired by **Shuibing Chen (Weill Cornell Medicine)** featured a two-part lecture by **Peter Schultz (Scripps Research Institute)** who provided some insight into several pioneering contributions to expanding the toolkit for synthetic biology. The first segment discussed recent progress in expanding the genetic code to facilitate the incorporation of non-canonical amino acids (ncAAs), which can site selectively introduce unique properties to a protein.²⁷ The first example discussed genetic encoding of a Her-2 specific antibody, allowing for the incorporation of *p*-acetylphenylalanine. Subsequent orthogonal chemistry facilitated the formation of an antibody-drug conjugate with auristatin F, as a targeted therapy of breast cancer.²⁸ Additional examples included incorporating photo-crosslinking ncAAs for probing interactomes,²⁹ developing ncAA dependent organisms, with potential for vaccination or biocontainment³⁰ and using ncAAs to enhance protein stability.³¹ This segment concluded with their recent success in expanding the genetic code of human hematopoietic stem cells, which can stably encode ncAAs.³² This led naturally into the second segment, and their work in understanding genome evolution. Here the initial discussion surrounded the introduction of a third DNA base pair, where through metabolic engineering, they were able to replace the majority of 2'-deoxycytidine in the *E. coli* genome with its 5-hydroxymethyl analogue (5hmC).³³ In an attempt to increase the uptake of 5hmC, they unexpectedly developed a bacterial mutant with a chimeric genome featuring substantial ribonucleotide incorporation.³⁴ While this discovery has important implications for genome manipulation, it also provides a potential mechanism for the putative RNA to DNA transition in early cell evolution. This led to a description of an elegant stepwise development of a respiration deficient *Saccharomyces cerevisiae* mutant, whose survival and replication was dependent on the presence of endosymbiotic ADP/ATP translocase expressing *E.coli*, thus

providing new insight into possible mitochondrial evolution and the prokaryote to eukaryote transition.³⁵

Concurrent Session 1: Bioengineering Commodities of the Future. This synthetic biology-oriented session was chaired by **Baulke Albada (Wageningen University and Research)** and **Abhishek Chatterjee (Boston College)**. The first speaker, **Michelle Chang (University of California, Berkeley)** presented their hybrid cellular – heterogeneous catalysis approach for the conversion of renewable glucose, a universal building block, to olefins³⁶ as an entry into the use of new resources to produce petrochemical-like hydrocarbons. Using the thiolate pathway to redesign a fatty-acid synthase (FAS)-independent pathway for hydrocarbon chain elongation in cells led after optimization to 4-5 g of C8-C10 products per liter. A bioinspired-approach based on the decarboxylation-dehydration of 3-hydroxy acids using Nb₂O₅ Lewis acid heterogeneous catalysis leads to terminal heptenes and nonenes, which can further be cracked to lower olefins, oligomerized to higher olefins, or isomerized. **Pamela Peralta-Yahya (Georgia Institute of Technology)** introduced the bioconversion of CO₂ to value-added chemicals in order to anticipate the potential need for bioproduction of rocket propellant. They introduced a biotechnology enabled ISRU (**In Situ Resource Utilization**) strategy using engineered cyanobacteria for 2,3-butanediol production as a propellant, and how this process could be implemented on Mars.³⁷ The next speaker was **Michael Jewett (Northwestern University)**, who targets the use of cell-free systems for on-demand biomanufacturing, introducing a cell-free antibacterial vaccine production platform from freeze-dried components – *in vitro* conjugate vaccine expression (iVAX) platform which could be engineered towards a portable, inexpensive and rapid biomanufacturing without a cold chain.³⁸ Another aspect of his research deals with the expansion of Nature's repertoire of ribosomal monomers, enlarging the range of ribosomal substrates to cyclic and expanded-backbone structures such as α , β , γ , δ , ϵ , and ζ - amino acids.³⁹⁻⁴² **Angela Steinauer (Eidgenössische Technische Hochschule Zurich)** described their efforts to engineer and evolve lumazine synthase from *Aquiflex aeolicus* to encapsulate its own genome. Lumazine synthase is a bacterial microcompartment that catalyzes the last step of riboflavin biosynthesis. Installation of an RNA binding peptide on the inside of the capsid and 3 rounds of directed evolution allowed

for the selection of capsids that package their own RNA genome and behave like natural virus capsid. Interestingly, this strategy led to change in the structure and size of the capsid, and to the evolution of RNA packaging signals, allowing for more efficient packaging.^{43,44}

Concurrent Session 2: Versatile Tools for the Chemistry-Biology Interface. This session, co-chaired by **Parasuraman Jaisankar, (CSIR-Indian Institute of Chemical Biology)** and **Peng Cheng, (Peking University)**, featured five speakers who delivered illuminating updates of their respective research activities. The first talk, delivered by **Andrea Rentmeister (WWU Munster)**, described an interesting strategy to reversibly modify DNA. Through the assistance DNA Methyltransferase, modified *S*-adenosyl-L- methionine (AdoMet) analogs, featuring a photo-caging (PC) group on the sulfur atom, can install the PC group into the DNA with site or sequence specificity. Subsequent triggering with light results in removal of the PC from the target site, and offers a new tool to reversibly modulate DNA functionality.⁴⁵ Similarly, they described a two-step enzymatic cascade by RNA methyltransferase for site-specific mRNA modification starting from stable methionine analogs,⁴⁶ as well as engineering a SAM synthetase from *Cryptosporidium hominis* for efficient generation of AdoMet analogs.⁴⁷ *N*-6-methyladenosine (m6A), and its subsequent modifications are a vital component of nuclear RNA that regulates several mammalian and plant biological processes. **Guifang Jia (Peking University)**, discussed their groups work in determining m6A as a major substrate for the human RNA demethylase, Fat Mass and Obesity-Associated Protein, (FTO), resulting in its oxidative demethylation.⁴⁸ This insight led their group to transgenically express FTO in commercially important crops, where they showed that active FTO transgenes increased the biomass of rice and potatoes by up to 50%, representing a promising strategy for future agricultural requirements.⁴⁹ Returning to the podium, **Anneliese Gest**, elaborated further on the development of fluorescence lifetime microscopy methodology to measure membrane potential of neurons. They emphasized the importance of membrane potential (V_{mem}) in numerous cellular processes, ranging from neurotransmitter release to cell cycle control and tissue patterning. However, existing tools for V_{mem} quantification have certain limitations. Electrode-based strategies are most effective in short-term cellular recordings, whereas optical strategies provide easier access to diverse samples but generally only detect relative

changes in V_{mem} .⁵⁰ The group developed a fluorescence lifetime-based (VFFLIM) approach for imaging and optical quantification of V_{mem} at single-cell resolution in mammalian cell culture.¹³ **Hankum Park (Seoul National University)** provided insight into early endosome proteomics and its application to spatial snapshots of amyloid precursor protein (APP) intramembrane processing. Here, characterization of the endosomal protein and lysosomal protein by pull-down methods showed a clear overlap of endosomal and lysosomal proteins. Multiplex targeted proteomics recapitulate the APP processing in endosomes and lysosomes.^{51,52} The last speaker of the session was **Himadri Sekhar Sarkar, (CSIR-Indian Institute of Chemical Biology)** who reported the development of a new set of photo reversible molecules named azopyrazolequinazolines (azoPyQs), capable of binding to *E.coli* DHFR and modulating enzymatic activity through reversible photo-isomerization. Importantly, this new class of photo-switchable ligands displayed greater selectivity, for *E.coli* over humans isoforms of DHFR, than in previous studies, and expands the utility of this method for elucidating protein functions in living cells.

Joint Session 2: Industrial Contributions to Advancing Chemical Biology. This session chaired by **Douglas Auld (Novartis)**, focused on industrial research as well as industrial-academic partnerships. It opened with a presentation by **Paul Brennan (University of Oxford)** who tackles research in Drug Discovery with an industrial mindset. Friedrich ataxia, is an autosomal recessive neurodegenerative disorder resulting from reduced expression of frataxin. Screening of the Structural Genomics Consortium epigenetic probe collection in a cellular phenotypic assay led to the identification of an inhibitor of H4K20 methylation (A-196) leading to increased levels of frataxin expression,⁵³ which their group are further optimizing towards a probe showing longer half-life for *in vivo* use. **Jeremy Jenkins (Novartis Institutes for Biomedical Research)** used augmented intelligence to create a sustainable Mechanism of Action library, (the MoA box), systemizing chemical genetics by introducing a set of well-characterized molecules and targets.⁵⁴ Development of a tool score and a set of additional criteria allowed for the selection of 3500 compounds covering 2247 targets. **Digital RNA with perturbation of Genes (DRUG-seq)** proved able to cluster compounds by mechanism of actions (MoAs) based on their intended targets.⁵⁵ Finally, **Frances Rodriguez-Rivera**

(Merck & Co) presented the μ MAP strategy, based on the introduction of a photocatalyst on the target for proximity activation of photolabels such as diazirines. Iridium-based photocatalysts proved able to activate diazirines using at short-range Dexter Energy Transfer.⁵⁶ These approach allowed for labeling intracellular targets such as BRD4 using a BRD4 ligand – Ir photocatalyst conjugate (JQ1-G2-Ir), which allows time-dependent labeling in live cells. This strategy could be extended to covalent labeling of cell surface targets such as the A2aR receptor, in a native cellular context.⁵⁷

Concurrent Session 3: Antimicrobial Discovery and Resistance: Tackling an Upcoming Health Crisis. This session, which highlighted one of the most urgent areas of contemporary drug discovery, was chaired by **Claudia Jessen-Trefzer (University of Freiburg)** and **Stephan Hacker (Leiden University)**. The opening speaker, **Anna Hirsch (Helmholtz Institute for Pharmaceutical Research Saarland)** described their collaborative project, which aimed to indent and develop inhibitors of the LasB virulence factor in *Pseudomonas aeruginosa*. The initial portion of this lecture described their successful identification and characterization of a mercaptoacetamide containing a hit fragment.⁵⁸ Inspired by the natural substrate, a subsequent fragment merging, elaboration and growing strategy, resulted in a small molecule candidate with substantially improved activity, compared to the fragment hit, coupled to in vivo activity in *Galleria mellonella* larvae model of infection.⁵⁹ **David Hilko (Griffith University)**, delivered a lecture describing the incorporation of an improved synthesis of ethynyl-2'-deoxyuridines (EdU) and its application as a chemical probe for the fluorescent labeling of DNA. Introduction of a 3-*N*-nitro protecting group on the pyrimidine, facilitated stable and stereo selective modification of the 2' position of the arabinosyl sugar.⁶⁰ Incorporation of these suitably modified DNA bases, into *Trypanosoma cruzi* DNA, followed by 'clicking' to a fluorescent azide, allowed for effects of inhibitory drugs on parasite replication to be assessed. This therefore, represents a promising probe to assess the killing characteristics of future anti-trypanosomals.⁶¹ **Snehalata Saini (IISER Bhopal)** elaborated on their work toward exploiting the bacterial signal recognition particle (SRP) as a target for antibacterial drug discovery. This strategy utilized antisense peptide nucleic acids (PNA), which feature an uncharged, and hydrolysis resistant pseudo peptide backbone, which binds

to bacterial specific regions of SRP RNA, and disrupt bacterial protein transport. Confirmed interaction between the designed PNAs and bacterial RNA translated into dose-dependent inhibition of bacterial cell growth.⁶² **Chris Meier (Hamburg University)** discussed the development of broad-spectrum antivirals, which disrupt host targets involved in the viral life cycle. His lecture focused primarily on anthranilic acid derivatives, which starved the virus of critical pyrimidines, via inhibition of host dihydroorotate dehydrogenase (DHODH). X-ray co-crystallographic analysis suggested that these human DHODH inhibitors bound to the entrance tunnel of co-substrate ubiquinone. Optimization of this class of compounds led to potent low nanomolar target activity, which translated into broad-spectrum anti-viral activity against several high profile viruses including, Ebola virus, Lassa virus and Zika virus. Furthermore, these compounds displayed promising metabolic and pharmacokinetic properties.

Concurrent Session 4: Drug Screening During a New Pandemic: Meeting the Balance Between Drug Repurposing and Novel Viral Biology. This particularly topical session chaired by **Arnab Chatterjee (Calibr at Scripps Research)** and **Shuibing Chen**, opened with **Sumit Chanda (Scripps Research Institute)** who discussed a collaborative effort during the initial stages of the pandemic to develop a high-throughput screening format for SARS-CoV-2 in Vero cells, followed by hit validation, triaging and mechanism of action studies. From the REFRAME library, this in-depth, repurposing study, ultimately identified clofazimine as an attractive clinical candidate for treating COVID-19, and possibly for future viral pandemics.^{63,64} Staying with the theme of preparing for future pandemics, **Matthew Frieman (University of Maryland)** presented their approach to developing a broad spectrum anti-viral platform. Through a yeast suppressor screening approach, they identified influenza A (IAV) and MERS viral proteins, which interact with eukaryote proteins, which they reasoned might act as host replication factors. From both screens, the SKI complex was identified and validated as a promising broad-spectrum anti-viral target. Subsequent computer-aided drug design led to the identification of three compounds, with IAV, MERS, Ebola, Marbug, SARS-CoV and SARS-CoV-2 antiviral activity, that interfere with viral RNA production.⁶⁵ **Johan Neyts (University of Leuven)** described their recent effort in identifying small molecules for

the treatment and prophylaxis of SARS-CoV-2 infection. Through their work, they found that high doses of the influenza antiviral favipiravir, showed encouraging in vivo antiviral efficacy in a Syrian hamster model,⁶⁶ and acted synergistically with molnupiravir.⁶⁷ Through application of the same model, they showed that the oral protease inhibitor PF-07321332 completely protected from intranasal infection and transmission.⁶⁸ **Samuel Constant (Epithelix)** detailed their technology, which uses reconstructed human airway epithelial cells, for the development of 3D human airway models as a tool to study SARS-CoV-2 pathogenesis. Amongst other important discoveries, including the ability to evaluate the efficacy of antiviral therapies, application of their technology found that SARS-CoV-2 infection impairs mucociliary clearance,⁶⁹ that ciliated and goblet cells are more prone to infection and that SARS-CoV-2, alters the innate immune transcriptional signature of nasal and bronchial models.⁷⁰ Based around their own epithelial cell screening, validation and mechanistic platform, **Sara Cherry (University of Pennsylvania)** discussed the identification of a cohort of 16 antiviral nucleoside analogues, including molnupiravir and remdesivir, which all displayed selective antiviral activity. Following the confirmation of their mechanisms of action, they found that inhibitors of *de novo* pyrimidine biosynthesis acted synergistically with molnupiravir or remdesivir, where this activity translated into physiologically relevant tissues. Interestingly, this synergy wasn't observed with purine biosynthesis inhibitors, alongside molnupiravir or remdesivir.⁷¹

Keynote Lecture 3: The third keynote session, chaired by **Laura Kiessling (Massachusetts Institute of Technology)** was delivered by **Deborah Hung (Broad Institute)** who opened by discussing the challenges of bacterial infection. These included the evolution of resistance, the sub-optimal duration of antibiotic therapy regimens, as well as how antibiotics tend to be primarily selected for their pathogen inhibiting or killing capacity, rather than improved clinical outcomes.⁷² Through this lens, the lecture moved toward their systems chemical biology approach to antibiotic discovery, and the challenges associated with traditional target and whole cell-based screening approaches, which tend to render compounds, which are either inactive in biologically relevant space, or fail to inhibit new target classes. Focusing on a recent *Mycobacterium tuberculosis* (MTb) project, they described their **PR**imary screening

Of Strains to **P**rioritize **E**xpanded **C**hemistry and **T**argets (PROSPECT), in which an essential MTb gene is depleted, thus generating a series of inhibitor hyper-susceptible hypomorphs, whose inhibition simultaneously confirm cell entry and target inhibition. Through a screen of reference compounds, they found that the strain response could infer an MOA, and facilitated the prediction of new inhibitory scaffolds against a target. Screening of compound libraries against pools of numerous modified MTb strains identified a new compound (BRD-8000), which un-competitively inhibited the essential efflux pump EfpA. Subsequent medicinal chemistry optimization, introduced activity against wild-type strains, with in vivo activity in a murine TB model. Selective generation of resistance to BRD-8000 indicated that resistance all occurred through the same single residue mutation.⁷³ Application of the reference strain response, resulted in the identification of an additional EfpA inhibitor (BRD-9327) whose resistance profile was different to BRD-8000. Importantly, they also found that high level resistance to one of the BRD compounds increased sensitivity and hindered the emergence of resistance to the other BRD compound, and thus offers a compelling option for MTb combination therapy.⁷⁴

Business Meeting. ICBS president **Jonathan Baell** led the annual business meeting, which was primarily focused on organizational aspects of ICBS as a geographically inclusive and diverse society. As a not for profit organization, the society, relies primarily on sponsorship, conference income and membership fee', which due to the society's growth has put ICBS in a financially sound position. It was further highlighted that the growth and reach of ICBS is inherently linked to the web of committees. Accordingly, efforts to grow, develop and cement these structures have been a key element of recent ICBS work. This was followed by brief reports from, the communications committee, membership outreach and services committee, industry, partnership and fundraising committee as well as the education committee. Within their portfolio's, each committee reported encouraging signs of growth and development, including increasing membership in new regions, arranging, symposia, networking events and mentorship for young chemical biologists, new partnerships and collaborations with industry and related societies and improved mechanisms of communicating with members, including a dynamic social media presence. **Haian Fu** took the opportunity to thank

Jonathan Baell for his significant contribution to the society and the enormous achievements in his tenure, particularly with respect to the development of new structures which have seen steady growth. This was followed by welcoming the new ICBS president, **Zaneta Nikolovska-Coleska** who briefly gave their vision for the future of the society including further expansion of outreach initiatives, mechanisms to recognize the achievements of ICBS members and support for emerging chemical biologists.

Networking and Professional Development Sessions. As has become custom with online meetings, the organizers, arranged two networking and professional development sessions. These included live poster sessions, where 29 outstanding posters were shared, meet and greet sessions with journal editors whom supported the event, an exhibitor booth and live demonstrations as well as well-attended networking socials.

Breakthrough Chemical Biology in the Oceania/ Asia Region. This session, chaired by **Yan-Mei Li (Tsinghua University)** and **Mikiko Sodeoka (Riken)**. Featured a special opening remark from ICBS president **Jonathan Baell** who reminded listeners that this session embodied the ethos of ICBS in embracing geographical diversity of participants. The opening speaker, **Feng Shao (National Institute of Biological Sciences)** discussed the role of pyroptosis and the inflammasome in antibacterial and antitumor activity. The group showed that while typically the inflammasome plays a defensive role against Gram-negative bacterial infection, pyroptosis mediated by mouse caspase-11 did not protect mice from *Shigella flexneri* infection. They determined, that this occurred due to specific ADP-ribosylation of caspase lysine's, which acts as a bacterial virulence instrument, and paralyzes pyroptosis defense.⁷⁵ Moving into the anti-cancer space, the group developed a bioorthogonal system, which selectively released a pro-pyroptosis client protein into mouse tumor cells. Data from these experiments revealed that induction of pyroptosis in as little as 15% of tumor cells was sufficient to clear entire breast tumor grafts, and as such indicates substantial potential for the role of the inflammasome in cancer immunity and therapy development.⁷⁶ **Rebecca Ritchie (Monash University)** discussed their contribution to the field of formyl peptide receptor (FPR) antagonists, as agents for managing ischemic injury.

The FPR family of GPCRs contribute to regulation of host inflammation. Given the role of inflammation in myocardial infarction (MI), FPRs offer the opportunity for therapeutic intervention. Accordingly, they identified a small-molecule FPR1/FPR2 agonist which protected mice against myocardial ischemia-reperfusion injury.⁷⁷ This was followed by a focused medicinal chemistry campaign, resulting in optimized small molecules, with potential and biased FPR agonist activity.⁷⁸ The next speaker, **Hyun-Woo Rhee (Seoul National University)** detailed their recent discovery where following application of proximity labeling methodology,⁷⁹ they identified a mitochondrial matrix protein OPax, which has NADPH-quinone oxidoreductase activity, and can protect mitochondria from ROS. Their study revealed that in the case of diseases such as optic neuropathy related to oxidative damage in mitochondria, OPax might prove a useful target. **Junko Ohkanda (Shinshu University)** discussed interactions of 14-3-3 proteins, and their role in numerous biological processes, as well as the fusicoccin A, a naturally occurring 14-3-3 stabilizer produced by fungi that helps in stomatal opening in plants.⁸⁰ Through, the rational design of model derivatives of fusicoccin, they were able to elucidate that the presence of a C-12 hydroxyl moiety hampers the formation of a ternary complex between fusicoccin analogues, a 14-3-3- protein and phosphopeptide ligands. Furthermore, their data indicated that the absence of the C-12 hydroxyl was correlated with improved cytotoxicity, thus indicating that the mode of action of fusicoccin analogues is innately linked to its capacity to initiate interactions with 14-3-3 proteins.⁸¹

DAY 3

Keynote Lecture 4. In an inspiring start to the day chaired by **Sally-Ann Poulsen, Dame Carol Robinson (University of Oxford)** walked delegates through the astonishing utility of mass spectrometry (MS) techniques for modern chemical biology. The first segment of their lecture focused on three protein assemblies critical in the SARS-CoV-2 life cycle. The first illustrative example detailed a study of the SARS-CoV-2 main protease (M^{pro}), where native MS, provided fine-grained details of the monomer/dimer equilibria, measurement of the rate of substrate turnover/proteolysis, and helped identify allosteric modulator of M^{pro}.⁸² The second example described how through charge-detection MS identified an unexpected heterogeneity in glycosylation of the SARS-CoV-2 S-protein timer, which they postulated was

a means of confounding host immune responses.⁸² Finally, looking at viral proliferation, through native MS they were able to characterize multiple different proteoforms of the SARS-CoV-2 nucleocapsid (N) protein, and elucidate their unique, RNA, immunophilin and antibody interactions. Furthermore, they were able to visualize a direct N-protein – cyclophilin interaction, which could also be disrupted with cyclosporine, thus providing a tool for future small molecule screening.⁸³ The second segment provided an insight into their groundbreaking work investigating protein structure and conformation of membrane proteins and methods to quantify the presence of interfacial lipids, and their influence on oligomer stability.⁸⁴ Application of this platform in the context of GPCRs, identified a specific endogenous lipid that stabilizes a specific Gα – β1 adrenergic receptor interaction, increases GTP turnover and modulated ligand interactions, and thus has important implications for drugging GPCRs.⁸⁵ This progressed onto the development of methodology, to identify and characterize ligands bound to membrane protein assemblies, and thus directly combining native MS with elements of metabolomics, proteomics and lipidomics, a technique which they termed ‘nativeomics’.⁸⁶ As an illustrative example, they showed how this powerful experiment was used to resolve fine-grained structural information of a V-ATPase, and understanding of its mechanism of assembly,^{86,87} and finally an example of expanding this approach to develop a light-sensitive method of measuring the rhodopsin transmembrane signaling cascade.

Concurrent Session 5: Chemical Biology Approaches to Unravel Cell Death Pathways.

This session, co-chaired by **Kamyar Hadian (Helmholtz Zentrum München)** and **Zaneta Nikolovska-Coleska (University of Michigan)** was the occasion of fascinating discussions on exploring different cell death mechanisms and the potential impact of their selective triggering to fight cancer and other diseases. Ferroptosis is a type of cell death by peroxidation of phospholipids containing polyunsaturated fatty acids (PUFA) tails. GPX4 is one of the key factors in the peroxidized lipids repair system, and Sedaghatian-type spondylometaphyseal dysplasia (SSMD) is a rare bone disease related to deficiency in GPX4. **Brent Stockwell (Columbia University)** identified three new SSMD patients with a R152H homozygous point mutation in GPX4. This mutation induces loss of stability of loop 124-132, indirectly resulting in instability of the catalytic triad, and simultaneously R152H GPX4 becomes more resistant

to degradation, because two lysines which are potential site of ubiquitylation are present in this loop.⁸⁸ In terms of potential treatment attempts for these patients, it appears that deuterated-linoleic acid most effectively rescues their fibroblasts. **Mitochondrial Outer-Membrane Permeabilization (MOMP)** triggers cell death either through apoptosis, or if caspase activity is inhibited, as **Stephen Tait (University of Glasgow)** explained, through **Caspase-Independent Cell Death (CICD)**, a pro-inflammatory process inducing an anti-tumor immune response, which beats apoptosis at inhibiting tumor growth and potentially avoids apoptosis resistance.^{89,90} Caspase inhibition might be a promising anti-cancer approach, proving that “how a cancer cell dies matters”.⁹¹ **Yilong Zou (Westlake University)** identified that clear cell carcinomas are sensitive to ferroptosis due to high polyunsaturated lipid levels, and GPX4 inhibition results in cell ferroptotic death. However, the cancer cell lipidome is plastic, and cancer cells can downregulate polyunsaturated lipids to evade ferroptosis *in vivo*.⁹² Further studies showed that polyunsaturated fatty acids are sufficient to induce ferroptosis in selected cancer cells. Sensitivity to ferroptosis for potential cancer treatment can be stratified by the PALP (**P**hotochemical **A**ctivation of **L**ipid **P**eroxydation) technique both *in vitro* and *in situ*.⁹³ This session continued with two flash talks. In their second stint at the podium, **Catia Pierotti** explained how necroptosis, a form of programmed cell death, is controlled by 3 proteins, RIPK1, RIPK3 and MLKL. They developed an optimized ligand which binds to human MLKL, as well as RIPK1 and RIPK3.⁹ This ligand, which is able to bind also in cells and inhibit necroptosis, showed efficacy *in vivo* in a mouse TNF-SIRS model. Interested in identifying which membranes are essential for lipid peroxidation in ferroptosis, **Nikolai von Krusenstiern**, also from **Columbia University**, showed that PUFAs deuterated at their bisallylic position localize to lipid droplets which are not relevant to the mechanism of ferroptosis and the at the endoplasmic reticulum, which is probably the primary site of action of these compounds in arresting ferroptosis. Interestingly, while PUFAs act as potentiators, MUFAs (mono unsaturated fatty acids.) are protective against ferroptosis.

Concurrent Session 6: The Power of Phenotypic Screening to Uncover New Cellular Targets. This session, chaired by **Bridget Wagner** opened with **Jason Sello (University of California, San Francisco)** who discussed the development of small molecules, which inhibit

retrograde trafficking in the host as a strategy for developing broad-spectrum antimicrobials. Early application of this approach resulted in the identification of small molecule inhibitors of polyomavirus and vaccinia virus,^{94,95} in addition to numerous other viral and non-viral microbes. In-depth mechanism of action studies indicate that these compounds inhibit retrograde trafficking by disrupting the transmembrane domain recognition complex pathway,⁹⁶ and thus offers a compelling target for host modulating anti-microbial discovery. Given the central role of phosphatidic acid in a variety of biological functions **Jeremy Baskin (Cornell University)**, described their progress in developing tools to help understanding elements of its cellular regulation. Through their chemoenzymatic phospholipase D (PLD) mediated labelling strategy termed **Imaging Phospholipase D Activity with Clickable Alcohols via Transphosphatidylation (IMPACT)**, they were able to identify cellular and sub-cellular locations of phosphatidic acid and PLD activity.^{97,98} Co-application of IMPACT with a genome-wide CRISPR screen was used to identify regulators of PLD signaling, from which they identified glycogen synthase kinase 3 (GSK3) as a PLD signaling regulator.⁹⁹ In a concise and informative lecture, **Douglas Selinger (Plex Research)** discussed the development of a search algorithm-based AI for drug discovery called Plex. Through a live demonstration of the software, they predicted GSK3 β to be the putative target of series 7 of the TCAMS antimalarial library. Importantly, the answer dataset included additional information underpinning the prediction, which is therefore, a useful tool for assisting phenotypic screening approaches. The final lecture in the session, given by **Laura Doherty (Harvard University)**, revolved around developing unbiased approaches to understanding function and therapeutic potential of deubiquitinating enzymes (DUBs) as they relate to cancer. By integrating multiple orthogonal data sets, they were able to uncover new insight into well-characterized DUBs, as well as explore the biological roles of understudied DUBs.¹⁰⁰

ICBS Global Lectureship: This session was chaired by **Jin Zhang (University of California, San Diego)**, who was the recipient of the 2020 ICBS Global Lectureship Award. Aiming at better understanding how natural electrophiles such as 4-hydroxynonenal (HNE) react and interact with the proteome, **Yimon Aye (Ecole Polytechnique Fédérale de Lausanne)** developed the T-REX™ methodology (**T**argetable **R**eactive **E**lectrophiles and **o**Xidants

Delivery). This technology is based on bi-functional photocaged probes with a targetable ligand (Halo-Tag) and on-demand liberation of a photocaged electrophilic signal of choice allowing spatiotemporal precision, ligand dosage control and chemotype control. This electrophile can react with the protein of interest in proximity, allowing to link on-target engagement directly to function. Applications of this technology was first validated on known electrophile responsive pathways in the field of antioxidants and stress response.¹⁰¹⁻¹⁰⁸

In terms of new targets identification, the oncogene Akt3 was recognized as a privileged first responder to native electrophile HNE.¹⁰⁹ In the context of potential drug development for triple negative breast cancer, mechanistic insights gained by these studies allowed for an Akt3 isoform-specific inhibitor.¹¹⁰ Further studies targeted the characterization of the protein players accounting for the drug response to the approved covalent drug dimethyl fumarate.¹¹¹ Keap1 is a critical sensor of reactive electrophiles, modification of Keap1 by reactive electrophiles ultimately results in Nrf2 transcription factor activation, that drives the upregulation of a battery of cytoprotective and antioxidant genes. RNA-Seq reveals downregulation of neutrophil and macrophage-related genes following Keap1-specific electrophile modification. Phenotype monitoring in live zebrafish larvae showed that Keap1-alkylation induced innate immune cell loss. Combination of T-REX with SILAC identified the altered interactome of modified Keap1 allowing the identification of Keap1/Wdr1 interaction as the target. Keap1 modification by an electrophile induces dissociation of Wdr1 from the Keap1/Wdr1 association, resulting in triggering of the Bax channel immune cell apoptosis. Alternatively to T-Rex, a G-Rex platform was developed for non-targeted mapping of novel electrophile sensors, in precise space and time,¹⁰⁶ and the most recent results involve evolution of a dual G-Rex/T-Rex platform.¹¹²

Concurrent Session 7: Carbohydrate Chemistry: From New Methodologies to Applications in Chemical Glycobiology. This session focusing on glycochemistry and chemical glycobiology was co-chaired by **Carmen Galan (University of Bristol)** and **Boris Vauzeilles (Institut de Chimie des Substances Naturelles CNRS)**. **Sabine Flitsch (University of Manchester)** presented several applications of galactose oxidase in the context of cell-free biocatalysis for bioconjugation.¹¹³ Galactose oxidase from *Fusarium sp.* was

submitted to directed evolution to expand its substrate range.¹¹⁴ It could also be immobilized on Purolite for the oxidation of benzyl alcohols and recycled,^{115,116} exploited in the chemoenzymatic synthesis of 3-deoxy-3-fluorofucose,¹¹⁷ or for the generation of DNA-encoded libraries.¹¹⁸ Other developments involve glycoprotein labeling,¹¹⁴ oxidation of Neu5Gc-containing glycans,¹¹⁹ or even the labeling of polysaccharides.¹²⁰ **Christophe Biot (University of Lille)** then reported on the use of the Chemical Reporter Strategy to image sialylation in live cells. This approach was developed as a tool to image Congenital Disorders of Glycosylation.¹²¹ Combination of two alkyne-functionalized metabolic reporters, namely ManNAI and SiaNAI, allowed imaging of both sialin-dependant and sialin-independent incorporation pathways.^{122,123} **Cristina De Castro (University of Napoli)** described their work on viral glycosylation of giant chloroviruses, on which they identified many original glycans with a new core structure, highly conserved between all chloroviruses. These glycans can be introduced on various peptidic sequences with no typical consensus.¹²⁴⁻¹²⁷ *Paramecium bursaria* Chlorovirus 1 (PBCV-1) encodes several glycosyltransferases, some of which are conserved through other chloroviruses.^{126,128,129} These glycosyltransferases might become useful tools for chemical glycobiology. Lipopolysaccharide of *Alcaligenes faecalis* shows promising properties as an adjuvant for vaccines. **Koichi Fukase (Osaka University)** described the chemical synthesis of its Lipid A, allowing for further studies and showing good immunostimulatory activity for hexaacylated lipid A. This chemically synthesized *Alcaligenes* Lipid A allowed for the development of a potent and safe nasal vaccine using *Streptococcus pneumoniae*'s pneumococcal surface protein A (Psp A),¹³⁰ inducing *Streptococcus pneumoniae*-Specific IgA and Th17 mediated protective immunity.

Concurrent Session 8: Chemical Proteomics in Health and Disease. **Raymond Moellering (University of Chicago)** and **Christina Woo (Harvard University)** were co-chairs of the session, focused on advances in chemical proteomics as a tool for understanding biological pathways and developing targeted therapeutics in the context of human health. **Christopher Parker (Scripps Research Institute)**, described the concept of **Fully Functionalized Fragment (FFF)** probes as an integrated strategy to identify chemical probes and ligands for a greater proportion of the human proteome. By combining the efficiency of

chemical fragments for exploring chemical space, with the utility of biorthogonal chemistry assisted proteomics, their technology was capable of identifying new protein-ligand interactions, identifying targets following a phenotypic screen, and offered new insights into the design of chemical probes.^{131,132} As an illustrative example, they showed how they elucidated new details of the role of the solute carrier protein, SLC15A4 in cell signaling, where they identified first-in-class SLC15A4 inhibitors which suppressed IFN1 production and disrupt both TLR7/8/9. As part of his ongoing contributions to spatiotemporal proteomic profiling^{133,134} **Peng Zou (Peking University)**, discussed their recent advances in spatiotemporally resolving subcellular glycosylation and phosphorylation post-translational modifications (PTMs). Subsequent analysis of the ER phosphoproteome, gave new insight into the role of PTMS to protein secretion and translocation.¹³⁵ Given the challenges associated with selective protein labeling in a crowded biological environment where endogenous protein expression is low,¹³⁶ **Tomonori Tamura (Kyoto University)** elaborated his ligand-directed N-alkyl-N-alkylsulfonamide (NASA) chemistry, which facilitates rapid modification of non-catalytic lysine residues of intracellular and membrane proteins located in close proximity to a ligand binding site. Through the modification of a known HSP90 inhibitor, and subsequent covalent inhibition of HSP90 activity, they demonstrated the utility of LD-NASA for irreversible inhibition of protein function in the cellular environment.¹³⁷ Similarly, the applicability of LD-NASA for inhibiting PPIs in cells, was demonstrated by the targeted covalent inhibition of the p53 – MDM2 PPI.¹³⁸

Keynote Lecture 5. Huw Davies (Emory University) chaired this fascinating session. More than 90% of proteins do not possess known binding pockets that small molecules can bind, and are considered as the undruggable proteome. In the case of cancer, these include KRAS, c-MYC, p53, p21. **Daniel Nomura (University of California, Berkeley)** uses chemoproteomics to discover covalent ligands against undruggable targets, expand the scope of targeted protein degradation and discover new induced proximity-based therapeutic modalities. Mapping complex proteomes using mass-spectrometry-based ABPP (MS-ABPP) identified over 100 000 ligandable hotspots across more than 16 000 human proteins using reactivity-based probes. In parallel, thousands of covalent ligands have been identified to

build libraries of compounds susceptible to be used in competition with reactivity-based probes for the identification of ligands for these hotspots.¹³⁹ Nimbolide, a natural product able to impair triple negative breast cancer cell proliferation was shown to target E3 ubiquitin ligase RNF 114, selectively alkylating an *N*-terminal cysteine located in an intrinsically disordered region.¹⁴⁰ Nimbolide inhibits ubiquitinylation of p21 by RNF114 by disrupting their interaction, leading to accumulation of p21 and p57 tumor suppressors. Nimbolide can therefore be exploited as a novel E3 ligase recruiter, as shown by degrading BRD4 or other targets such as the fusion oncogene BCR-ABL, with different specificity compared to CRBN and VHL-based degraders (selective degradation of BCR-ABL vs c-ABL).¹⁴¹ Target-based screening of their covalent cysteine reactive library led to the identification of a fully synthetic RNF114 recruiter for targeted protein degradation applications (EN219).¹⁴² Moreover, their team identified ligandable cysteines in over 98% of the human E3 ligases.¹⁴³ FEM1B is an E3 ligase that regulates reductive stress by turning on mitochondria to generate oxidative stress. Screening identified EN106, which covalently engages FEM1B in cells, and is usable as a new E3 ligase handle for PROTAC applications.¹⁴⁴ Dan Nomura also alluded to the development of E3 ligase **Swarm-T**argeting **Chimeras** (SwarmTAC) for recruitment of core ubiquitin proteasome machinery such as E2 ligase or cullin adapters, as well as understanding rules for rational design of molecular glue degraders. They finally described deubiquitinase-targeting chimeras (DUBTACs) for Targeted Protein Stabilization. An example of DUBTAC stabilizes mutant CFTR in human bronchial epithelial cells, and increases chloride current in primary human donor bronchial epithelial cells from $\Delta F508$ -CFTR patients.¹⁴⁵

Concurrent Session 9: Next Generation Drug Targets for Cancer Therapy. Following welcomes from the chair of this EFMC session, **Rui Moreira (University of Lisbon), Yimon Aye** returned to the podium to discuss their **Targetable Reactive Electrophiles and OXidants** (T-REX) technology, in which a toolkit of biocompatible, photocaged electrophiles, which facilitate selective, single-protein precision targeting of nucleophilic cysteines.¹⁰⁸ The efficient identification of functionally responsive cysteines, which are susceptible to covalent inhibition via small molecules, has been expanded into mammalian cells, *C. elegans* and zebrafish in a genome wide format.^{106, 111,146} As an illustrative example, they detailed, how

this method was used to identify a specific isoform of Akt (Akt3), as a target for triple negative breast cancer, which led to an electrophilic inhibitor, with oral *in vivo* activity in a murine breast cancer model.¹¹⁰ Through two case studies, **Tiago Rodrigues (Universidade de Lisboa)** described their work in applying machine learning (ML) to prioritize and identify targets of small molecules, and demonstrated the immense power of ML in contemporary drug discovery. The first example detailed, how their workflow, which employs **Self-Organizing MapS** (SOMS) to identify 5-lipoxygenase (5-LO) as the putative target of β -lapachone, with subsequent validation studies confirming that β -lapachone is a potent allosteric modulator of 5-LO.¹⁴⁷ The second case study, detailed, how SOMs were used to establish a relationship between the human transient receptor potential vanilloid 2 (hTRPV2) channel and the anti-cancer activity of piperlongumine, where it was found to allosterically modulate hTRPV2 mediated calcium influx, resulting in the impairment of glioblastoma progression.¹⁴⁸ Returning to the concept of covalent inhibition, **Stephen Hacker** closed the session describing their contribution to residue-specific proteomics, to identify nucleophilic residues on a target protein, which are susceptible to attack from small molecules.¹⁴⁹ While the majority of work in this area has focussed on nucleophilic cysteine residues, they described the development of light-activated methyl tetrazole probes, which facilitated global profiling of nucleophilic aspartate and glutamate residues in gram-positive and gram-negative bacteria.¹⁵⁰ This approach was efficiently expanded to develop probes capable of proteome-wide monitoring of nine different amino acids as well as protein N-terminii,¹⁵¹ and is now primed to assist in profiling covalent inhibitors against a more diverse set of amino acids.

Concurrent Session 10: Hijacking and Targeting the Ubiquitin System. The session chaired by **Stefan Knapp (Goethe University Frankfurt)** and **Xiangbing Qi (National Institute of Biological Sciences, China)**, was dedicated to recent progress in the field of chemical biology targeting the ubiquitin-mediated cellular function. **Monique Mulder (Leiden University)**, opened the session by introducing the basic concepts inerrant to ubiquitination and deubiquitin and their role in several cellular processes including the proteasome, transcription, enzyme activation, DNA replication, endocytosis and metastasis.¹⁵² This was followed by a fascinating description of their contributions to

developing activity-based probes for interrogating enzymatic cascades of ubiquitin and ubiquitin like modifiers.¹⁵³ This was followed by the recent development of a direct linear synthesis of SUMO-based probes, which facilitates labelling at both the *N*- and *C*-termini and the dual incorporation of dyes and reactive warheads, to help probing SUMO specific enzymes.¹⁵⁴ **Ting Han (National Institute of Biological Sciences, China)** introduced the concept of molecular glues for targeted protein-degradation and the substantial challenges associated with their development, as well as their potential against ‘undruggable’ targets. Through this, they described the development of a HTS assay, which led to the identification of a small molecule, which promoted the interaction between CDK12 and the DDB1-CUL4-RBX1 E3 ligase, leading to degradation of Cyclin K. In addition, to this effect leading to cancer cell death, these results also present new modalities for targeted protein degradation.¹⁵⁵ In a related story, they described resolving the MOA of the anti-cancer sulfonamide, indisulam, as well as some closely related compounds, which they found promotes binding of RBM39 to the CUL4-DCAF15 E3 ubiquitin ligase, resulting in disproportionate degradation of RBM39.¹⁵⁶ **Georg Winter (Austrian Academy of Science)**, discussed chemical genetics as a tool to identify and characterize small-molecule degraders. Using genome wide CRISPR-Cas9 screening, in conjunction with small molecules known to interact with different substrate receptor of cullin ring ligases (CRL), they were able to map the common and degrader unique factors, which influence degrader activity.¹⁵⁷ In a study related to work discussed previously in the session, they described a comprehensive screening and characterization study, identifying anticancer molecular glues, which promote the degradation of Cyclin K, through interaction between the CDK12–cyclin K complex with a CRL4B ligase complex.¹⁵⁸ In the final lecture of the session, **Xiulei Mo (Emory University)**, discussed the concept of hypomorph surface amino acids residues as built-in molecular glues that hold protein complex together, and how these can direct us to regions to target when developing molecular glues. By initially identifying hypomorph mutations, which disrupted the SMAD4-SMAD3 PPI and resulted in a loss of tumor-suppressing TGF- β signalling function, they were able to rationalize the importance of this region to PPI formation. Subsequent small molecule screening, led to a bisindolylmaleimide, which promoted PPI formation and reversed the loss of function, which

provides a powerful systematic approach for identifying therapeutically relevant molecular glues.¹⁵⁹

Joint Session 3: Reflecting on the First Decade of ICBS. As the first keynote speaker of the first ICBS conference held in 2011, the sole speaker for this special session was **Christopher Austin (CEO partner, Flagship Pioneering)** who opened by highlighting the poor transition between clinical or scientific observation and application in patients. There have been numerous important milestones in the field's recent history, including the creation of the human genome toolbox in 2003, to make the human genome public without any restrictions, and the molecular libraries program, which offered access to the large-scale screening capacity required to identify chemical probes as tools to perturb genes, to interrogate biological targets and pathways. During the last decade, the translational discordance has continued to widen between science and therapeutic interventions with scientific discoveries outstripping the ability to address human disorders associated with them. To address the dearth of scholarly literature on methodology focusing on preclinical drug development and HTS techniques, a freely accessible ebook by Lilly-NIH 'The assay guidance manual' helped to promulgate knowledge in drug development and chemical biology. The national center for advancing translational science (NCATS) was established ten years back to bridge the gap between the translational process and therapeutic outcome for the patient's cure. They concluded by saying that during the last decade the chemical biology community has reached out to explain the critical processes of developing chemical probes and target identifications, which catalyzes opportunity for the advancement of science, medicine as well as reducing the translational gap. Historically chemists and biologists outside of the Pharma industry did not see themselves working in the same area. ICBS was created to be an intellectual home for those committed to the study of chemistry in living systems.

DAY 4

Keynote Lecture 6. This session, chaired by **Hiroyuki Osada (Riken Center for Sustainable Resource Science)**, introduced **Dan Yang (Westlake University)**, who

developed 5 generations of fluorescent probes for different types of Reactive Oxygen Species and Reactive Nitrogen Species over 15 years of work. For example, HKGreen-4 detects peroxynitrite with high sensitivity and selectivity through an oxidative *N*-dearylation reaction.¹⁶⁰ A similar trigger allowed the development of HKYellow-AM for fluorescent imaging of peroxynitrite in animal models.¹⁶¹ Interestingly, oxidative *O*-dearomatization of a 2,6-dichlorophenol proved selective for HOCl, as in the HKOCl-3 probe.¹⁶² Similarly, HKOCl-4, a yellow fluorescent probe for HOCl, can be used for the detection of hypochlorous acid in living cells and tissues. Superoxide has been shown to react with triflate esters, leading to HKSOX-1r for its detection in live cells and *in vivo*,¹⁶³ or its targeted alternatives such as HKSOX-2m for mitochondria targeting, or HKSOX-2l for lysosomes. Completing the collection, 2,6-diiodophenols proved selective for hydroxyl radicals, and their implementation led to HKOH-1,¹⁶⁴ which allows for screening HO° scavengers, and second-generation HKOH-2, which was developed as lysosome targeting and halo-tag versions. A tandem Payne/Dakin reaction was used to design HKPerox-1 for hydrogen peroxide detection,¹⁶⁵ as well as a ratiometric probe HKPerox-3, for the quantification of cellular H₂O₂ during starvation.¹⁶⁶ A new generation, HKPerox-4, allows imaging in zebrafish embryo and the detection of glucose or other metabolites through the suitable oxidase, concomitantly converting O₂ into H₂O₂. Clinical translation might enable detection of H₂O₂ from exhaled breath condensates, in order to develop diagnostic kits. Chemiluminescence versions of such probes enabled real-time monitoring of H₂O₂ in rat brain,¹⁶⁷ recurring real-time imaging of HOCl in mice,¹⁶⁸ or the detection of endogenous HO° production in High Fat Mice. In another strategy, their group designed mitochondria-targeted, cardiolipin-selective probes with a diamino group matching the distance and charges of the diphosphate polar head of cardiolipin, allowing confocal imaging in live cells. And finally, they designed fluorogenic probes for carbapenemase detection in the context of antibiotic resistance,¹⁶⁹ ligand-directed light activatable probes for fluorogenic protein labeling in living cells,¹⁷⁰ and NapIM, a probe for real-time imaging of DNA-G quadruplexes in living cells.

Joint Session 4: Rising Stars Award Presentation. Haian Fu, provided a background to the award, with a particular emphasis on the importance of recognizing emerging talent in

the society. In recognition of their outstanding early-career achievement in chemical biology, **Lesley Aldrich (University of Illinois at Chicago)**, **Nir London (The Weizmann Institute of Science)** and **Jeremy Baskin**, were the worthy recipients of the 2021 ICBS Young Chemical Biologist Award. In an invited podium presentation, **Lesley Aldrich** described their approach to developing high throughput assays for detecting autophagy inhibiting PPI inhibitors as a means of disrupting cancer cellular homeostasis. In the first example, they described the development of a NanoBRET assay format, which identified a small molecule, inhibiting the Beclin 1-ATG14L PPI, and leading to selective disruption of the VPS34 Complex 1 and subsequent autophagy inhibition.¹⁷¹ As an alternative means of disrupting autophagy, they sought to inhibit the formation of the LC3-II complex, by disrupting the ATG5-ATG16L1 PPI, which they successfully achieved through a compound-multiplexed fluorescence polarization assay.¹⁷² **Jeremy Baskin** returned to the podium to provide additional insight into their IMPACT technology, particularly, in their successful efforts to speed up the requisite biorthogonal reactions and enhance the spatiotemporal imaging of phosphatidic acid in the cellular environment. In addition to facilitating the elucidation of lipid trafficking kinetics, these probes also act as tracers to investigate non-vesicular lipid transfer pathways,¹⁷³ as well as the role of phosphatidic acid in cell signaling pathways.^{174,175} **Nir London** opened their presentation with an overview their contribution to covalent ligand discovery including fragment screening, PROTACS and *in silico* methodology.¹⁷⁶⁻¹⁷⁸ This progressed onto the focus of the lecture which detailed the development of α -substituted methacrylamides as a new class of electrophiles, with tuneable reactivity and whose mechanism of covalent bond formation results in the release of a leaving group in the β -position. Modification of the known covalent BTK inhibitor ibrutinib with these new warheads resulted in increased selectivity and inhibitory potency. Furthermore, incorporation of fluorescent leaving groups allowed for covalent inhibition to be measured by fluorescence, in a generic technology they called **Covalent Ligand Directed Release (CoLDR)**,¹⁷⁹ which has been expanded to incorporate a chemiluminescent probe. Further investigation of **CoLDR** showed that translocation of the carbonyl group altered the mechanism of covalent bond formation, so that the active site binding compound was eliminated, leaving a site specifically tagged protein. This approach

was shown to be useful in incorporating numerous functionalities for biorthogonal chemistry.¹⁸⁰

Future Forum: Looking Forward to the Next Decade of Chemical Biology. The moderators for the session **Milka Kostic (Dana Farber Cancer Institute)**, and **Will Pomerantz (University of Minnesota)**, alongside a group of highly motivated panelists (**Gonçalo Bernardes, University of Cambridge; Jay Bradner, Novartis Institute for Biomedical Research; Masatoshi Hagiwara, Kyoto University; Laura Kiessling, Massachusetts Institute of Technology; Yamuna Krishnan, University of Chicago; and Zaneta Nikolovska-Coleska, University of Michigan**) facilitated this open forum for sharing ideas for bridging the gap between chemistry and biology. The early discussion highlighted that the strength of chemical biology emanates from its interdisciplinarity, which provides insight, which is not achievable using traditional chemical or biochemical tools in isolation. This has been augmented by rapid development of sophisticated methodologies including AI and the community should collectively progress with an unapologetic translational mindset for the good of humanity. It was agreed that as a cross cutting field, chemical biology has made a significant impact through the development of probes to understand the druggable genome and proteome, which has had a significant impact on drug discovery. Related to this have been breakthrough technologies related to drugging PPIs, targeted degradation, and ever advancing assay platforms, which allow deeper probing of druggable chemical space.

Moving onto weaknesses and blind-spots, the panel broadly agreed that global engagement and collaboration, seeking expertise outside one's own, integration of scientists from different backgrounds and partnerships with industry are areas which require work. In closing the session, the panel were asked to discuss their thoughts on the future of global chemical biology. Point raised here included maintaining the momentum of the last decade, and the appetite for knowledge, to ask unconventional questions; facilitating growth and training by improving scientific inclusion, including the participation of young researchers across the globe from diverse fields.

Closing Remarks. The incoming president of ICBS **Zaneta Nikolovska-Coleska**, closed ICBS 2021, by thanking all the delegates, attendees, sponsors, exhibitors, and the conference organizers, who together ensured that this year's meeting retained the tradition of the high quality science associated with the ICBS. From a particularly competitive field, the selection committee selected **Laura Depta, Catia Pierotti, Joomyung Jun** and **Zhi (Lindsey) Lin**, as the recipients of the ICBS 2021 Trainee Presentation Awards. Furthermore, the ICBS 2021 Poster Awards, were presented to three excellent young chemical biologists: **Beatrice Chiew, (Monash University, Design in the dark – illuminating the druggability of 53BP31 for BRCA1 breast cancer), Dipika Sarkar (Indian Institute of Chemical Biology, Modulating Agonism and Antagonism in Toll-Like Receptor 7 and Structural Optimization Leading to Antagonists relevant to Psoriasis)** and **Dr Ejaz Ahmad, (University of Michigan Identification and characterization of small-molecule inhibitors targeting Pregnancy-Associated Plasma Protein A), with Ilana Kotliar, (Rockefeller University) and Tulsi Upuhay, (Indian Institute of Science and Education Bhopal)** receiving honorable mentions.

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