Drugs of the Future 2018, 43(8): 627-633

CCC: 0377-8282/2018
DOI: 10.1358/dof.2018.043.08.2856625

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Meeting Report

Current Trends in Drug Discovery – Young Scientists and Tomorrow's Medicines: Highlights from a joint Society for Medicines Research and British Pharmacological Society meeting

King's College London, London, UK – June 7, 2018

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Summary

This well-attended 1-day meeting brought together presentations from both internationally recognized keynote speakers and early career scientists presenting their work either as oral or poster presentations. There are several early career symposia organized by the various professional bodies focused on their areas of interest. This meeting, however, specifically aimed to bring together early career scientists from all life science and drug discovery related disciplines and from academia and industry to share their work and experiences. A broad range of topics were covered during the day through posters and oral presentations and included medicinal chemistry, chemical biology, cheminformatics, biochemistry, safety assessment, application of artificial intelligence, assay development and aspects of regulatory approval. Prizes were awarded for the best presentation and best poster. The best presentation prize was awarded to Hannah Lithgow (Strathclyde University and GlaxoSmithKline [GSK]) for her talk "Investigating the impact of covalency on Protac-mediated degradation of BTK." The best poster prize was awarded to Cassie Messenger (GSK) *for her poster "Molecular availability and accessibility – measuring and understanding cellular drug concentrations." All presenters of posters and oral papers also received free membership of the Society for Medicines Research (SMR) for a year. The meeting organizers gratefully acknowledge sponsorship by AstraZeneca and GlaxoSmithKline.*

Key words: Ibrutinib – Iniparib – Serine racemase inhibitors – Antibody–drug conjugates – NPR-C agonists – Artificial intelligence – AZD-1419 – AZD-14202

How Do We De-risk New or "Undruggable" Targets, Quickly?

Prof. Chas Bountra, University of Oxford

Prof. Bountra presenting the first keynote talk—which used no visual aids—began with a summary of his perspective on the state of drug discovery today. Specifically, Prof. Bountra highlighted that discovery of new medicines is too risky, too slow and too costly. For example, the cost of developing a new drug is now estimated to be up to USD 4 billion. Cancer therapy has increased in cost about 10-fold over a 10-year period. To demonstrate the slow speed of the drug discovery process, Prof. Bountra cited an analysis that identified 529 compounds in phase I-III in 2002; by 2013, 45 had reached the market, 389 had failed for various reasons while 95 were still in development.

Furthermore, there is massive duplication of effort on a relatively small number of molecular targets and reproducibility of published data is being increasingly questioned increasing wastage. Finally, reflecting the duplication of effort, only approximately 40% of FDA approvals can generally be described as novel each year. In some therapeutic areas no novel entity had been introduced in decades. For example, in Alzheimer's disease the last new therapy was approved in 2002 and the last novel mechanism in antibiotics was introduced in 1984. Some of the limitations in drug discovery described by Prof. Bountra were difficulty in diagnosing patients, the general lack of predictive in vivo models and lack of objective, quantifiable biomarkers.

The validation of pioneer targets for drug discovery remains a major challenge. The fact that major pharmaceutical companies had a 50-60% failure rate in reproducing academic data was a case in point. Prof. Bountra outlined the strategy he has adopted to improve validation and hinges on pre-competitive pooling of resources from industry, the Wellcome Trust and multiple patient groups and philanthropists. This resource is being used to work on novel targets to identify high-quality chemical starting points for further investigation. These tools are extensively characterized and demonstrate cellular activity and have been peer reviewed by a group of experts prior to release. A key element in this work is that results are made freely available to all with complete transparency encouraging collaboration between groups. An additional element in both the identification and screening of new targets is access to primary human cells.

Research targets are focused on the epigenetics field with over 70 tools now generated. Particular therapeutic interest is in cancer metabolic disorders, inflammation and neuropsychiatric disorders with a panel of industry and academic experts to help prioritize targets for investigation.

Finally, Prof. Bountra outlined separate new collaborative initiatives on diseases associated with aging (Birmingham, Dundee, Oxford) and neuropsychiatry (Bristol, Cardiff, Oxford). The impact of Prof. Bountra's talk on the audience was clearly significant as several speakers and poster presenters referenced his talk during the rest of the meeting.

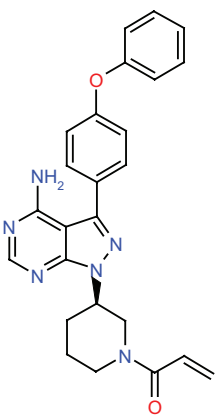
Investigating the Impact of Covalency on Protac-mediated Degradation of BTK

Ms. Hannah Lithgow, University of Strathclyde and GlaxoSmithKline (GSK)

Ms. Lithgow gave a brief review of proteolysis targeting chimeras (Protacs) as a strategy for catalytic targeting of protein degradation (1) followed by a description of her work on comparing reversible versus irreversible Bruton tyrosine kinase (BTK) inhibitors as Protacs.

Protacs are heterobifunctional small molecules which allow targeted protein degradation via the ubiquitin-proteasome system. A Protac comprises a targeting molecule, (e.g., an enzyme inhibitor), a variable linker chain and a motif that is recognized by E3 ligase (e.g., a VHL or IAP binder). By simultaneous engagement of both a target protein and an E3 ligase, a Protac promotes formation of a ternary complex, allowing ubiquitin transfer from the E3 ligase onto the target protein ('event-driven pharmacology'). The ubiquitinated target protein is then recognized and subsequently degraded by the proteasome.

The acrylamide-based irreversible BTK inhibitor **ibrutinib** was chosen for elaboration into a covalent Protac to probe the effect of covalency on Protac-induced degradation. Ibrutinib irreversibly binds to BTK through cysteine 481 in the active site (2, 3) which, if incorporated into a Protac, would be expected to remove the catalytic ability of the Protac. Furthermore, BTK was previously identified within



Ibrutinib

the GSK laboratories as a protein which is amenable to degradation via the Protac approach, and was therefore deemed a suitable target for this investigation. It was hypothesized that a covalent Protac would reduce the degradation efficiency of BTK compared to a reversible Protac but should still theoretically act as a stoichiometric degrader. To assess this hypothesis, a reversible Protac based on the ibrutinib scaffold was also prepared.

An irreversible BTK Protac inhibitor based on ibrutinib was prepared by elaborating ibrutinib via the acrylamide to attach the linker and E3 ligase IAP binding motif. A reversible BTK Protac inhibitor was prepared by reduction of the acrylamide. The irreversible binder was demonstrated by mass spectrometry to covalently modify BTK while, as expected, the reversible Protac did not. In contrast the reversible inhibitor gave reductions in BTK in cells while the irreversible Protac had no impact. Modification of the linker chain length made no difference to the observed results. These data suggest that more work is required to understand whether non-catalytic Protacs are suitable for a therapeutic approach.

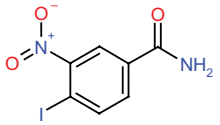
Target-probe Assessment Resource: Objective Assessment of Chemical Probes to De-risk Chemical Biology and Follow-up Drug Discovery

Dr. Albert Antolin, The Institute of Cancer Research

Dr. Antolin described the development of a new open access tool, Probe Miner (http://probeminer.icr.ac.uk) to help in the selection of chemical probes for use in chemical biology, for understanding biological systems and target validation.

Chemical probes are essential reagents in chemical biology and drug discovery for understanding biological systems and for target validation (4, 5). However, selection of chemical probes is largely subjective and prone to historical and commercial biases (2, 3). Despite many publications discussing the aspirational properties of chemical probes and the proposal of 'fitness factors' to be considered when assessing chemical tools, scientists often select probes through web-based searching or previous literature that are heavily biased toward older and often flawed probes or use vendor catalogues that do not discriminate between probes (5, 6). The prevalent use of low-quality chemical probes challenges many chemical biology studies, their reproducibility and significance and consequently their translation into follow-up drug discovery.

To illustrate this problem, Dr. Antolin highlighted the case of **iniparib**, the first compound reported as a poly [ADP ribose] polymerase (PARP) inhibitor to enter the clinic. It duly failed in clinical trials and was subsequently identified as a non-specific cysteine protease inhibitor but is still described as a PARP inhibitor in some studies. True PARP inhibitors are now successful drugs.



Iniparib

Dr. Antolin described how the Institute of Cancer Research team integrated publicly available curated databases such as CHembl and canSAR to analyze the scope and quality of bioactive molecules. The data set included over 1.8 M compounds assessed against 2,200 human targets. A set of six chemical probe scores—potency, selectivity (the most challenging to define), cell potency, SAR, availability of inactive analogues and PAINS—mirroring the proposed 'fitness factors' to enable the objective, quantitative and data-driven assessment of chemical tools using public data were used to develop the online Probe Miner resource to democratize access to these scores for an improved approach to chemical probe assessment and selection.

The Probe Miner team is working closely with the Chemical Probes Portal, which is a complementary tool for helping to select appropriate literature compounds as tool probes.

Identification of a Novel Inhibitor of Serine Racemase Through a Fragment-based Drug Discovery Strategy

Dr. C. Koulouris, University of Sussex

Dr. Koulouris discussed her approach to identify serine racemase inhibitors that will inhibit the production of D-serine, a co-agonist of *N*-methyl-D-aspartate (NMDA) receptors (7). NMDA receptors are highly expressed in the CNS and drive excitatory synaptic transmission and synaptic plasticity and their dysfunction has been implicated in Alzheimer's disease, neuropathic pain, schizophrenia and depression. Thus, reduction of D-serine levels may have therapeutic benefit in a range of CNS conditions associated with NMDA receptor hyperfunction.

Human SR (hSR) was successfully expressed, purified and crystallized in-house (8). A coupled biochemical assay was optimized to measure SR activity, in which produced D-serine is degraded by D-amino acid oxidase (DAO) with a horse radish peroxidase-mediated chemiluminescence readout. After the assay was characterized, fragment libraries were screened to identify active serine racemase inhibitors. As inhibitors of DAO would also read out as positive, all initial hits were counter screened for DAO activity and those showing DAO activity removed from further analysis.

In all, 60 hSR-fragment hits were identified that were inactive in the DAO counter screen and demonstrated IC_{50} values against hSR of around 1 mM or below. These hits were

characterized with biophysical methods adapted for hSR (thermal shift, microscale thermophoresis) to determine K_D values. Three distinct structural series were identified and used to further probe SAR. Excitingly, binding of one fragment F01 (K_D = 960 μM) was confirmed by X-ray crystallography to a resolution of 1.8 Å.

Further analogue studies and crystallization trials for the remaining hits are in progress.

The Kinetics of Drug Action; Signaling in a New Era

Prof. Steven Charlton, University of Nottingham

Prof. Charlton in his keynote presentation started by explaining that physiological processes occur over a wide range of time scales, from sub-second neurotransmission through to longer-term events such as cell proliferation or differentiation that can occur over hours or even days. To accommodate this, the signaling pathways that control these processes are fundamentally dynamic, with the onset and duration of effect influenced by a complex multitude of interacting factors, including the nature of the signaling molecule, receptor effector protein expression levels, substrate depletion and pathway desensitization, among many others. Despite this, in drug discovery programs signaling pathways are often measured at a single time point, most commonly at the peak response. In addition, many analytical pharmacology approaches currently in use are based on equilibrium models, including the operational models currently used to describe the relative strength of agonist efficacy at different pathways. This can result in misinterpretation of agonist pharmacology and highlights the need to look beyond a single snapshot in time. Prof. Charlton also highlighted the importance of understanding the kinetics of ligand binding and the dynamics of pathway activation/desensitization on drug efficacy at different pathways. Examples were given where differences in the kinetics of ligand binding could be misinterpreted as pathway selectivity or "biased agonism." Furthermore, Prof. Charlton spoke about how leveraging the theoretical potential of biased agonism provides several challenges for drug discovery, using drugs at the dopamine D_2 receptor and salmeterol time-dependant induction of β-arrestin, as examples. Some of these challenges are due to the techniques used to quantitatively analyze biased agonism; other challenges include the need to robustly identify in a very early phase which cell type harbors the cellular target of the drug candidate, to understand which signaling pathway leads to the desired therapeutic effect, and how these pathways may be modulated in the disease to be treated (9). He also discussed how low agonist efficacy can influence signaling strength over time and perhaps avoid acute on-target side effects. The example was given of the relatively recent introduction of FRET- and BRET-based pathway biosensors that have allowed the study of the kinetics of signaling cascades at a temporal resolution previously not possible. Finally, Prof. Charlton argued for new analytical pharmacology approaches that incorporate rate of signaling to better predict clinical efficacy and tolerability.

An Antibody–Drug Conjugate Approach to Selective Inhibition

Dr. Elizabeth Love, LifeArc

Dr. Love highlighted the potential for antibody–drug conjugates (ADCs) as targeted biotherapeutics combining the high specificity and long circulating half-life of an antibody with the cell killing potency of a small-molecule payload. Over the last decade the field of ADCs has gained significant momentum, with four ADCs currently on the market which employ either DNA-damaging or tubulin-targeting cytotoxic payloads. Dr. Love described a novel ADC approach which brings selectivity to a broad-spectrum inhibitor, focusing on a problematic target—matrix metalloproteinases (MMPs). MMPs are a family of structurally related proteolytic enzymes, implicated in many degenerative conditions including cancer, inflammation and cardiovascular disease. Over the past 30 years, significant drug discovery effort has been invested into generating small-molecule MMP inhibitors. Many of these inhibitors reached late-stage clinical trials, however their lack of MMP-subtype selectivity led to negative results. Dr. Love used this as an example of an extracellular ADC in which both antibody and small molecule bind to a single target, bringing selectivity to a formerly nonselective inhibitor.

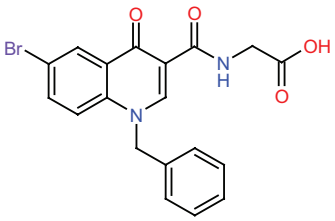
Synthesis and Evaluation of Novel Nonpeptide Agonists of NPR-C

Dr. D. Conole, University College London

Endothelium-derived C-type natriuretic peptide (CNP) possesses cytoprotective and antiatherogenic functions that regulate vascular homeostasis and smooth-muscle relaxation. The vasoprotective properties of CNP are mediated by the natriuretic peptide receptor type-C (NPR-C), thus the latter represents a novel therapeutic target for the treatment of cardiovascular diseases. Dr. Conole described his work in the design and development of an array of biophysical and physicochemical methods to help assess small-molecule drug-like mimetics of CNP agonists at NPR-C.

Of the biophysical methods explored, surface plasmon resonance (SPR) proved most useful in tracking the SAR of a series of small-molecule ligands based on the lead compound ([**I**]). These ligands were identified from a systematic design program that identified key binding features of the natural ligand CNP and the peptide antagonist M-372049 derived from the published crystal structure of NPR-C (10, 11)

Fluorescence polarization (FP) and thermal shift (TS) assays detected natural ligand and peptide antagonists



[I]

such as M-372049 but were not generally sensitive enough to detect interaction of small-molecule agonists with the receptor.

Encouragingly, Dr. Conole reported that selected small-molecule agonists demonstrated vasorelaxation in small mesenteric artery, but not other tissues, and a reduction in blood pressure in vivo indicating translation of activity seen in SPR to tissues. Further investigation of the series is in progress.

The Use of Surrogate Markers in EMA Fast-track Approval Pathways—Are They Validated?

Dr. Catherine Schuster Bruce, The William Harvey Research Institute

In situations of unmet medical need, Conditional Marketing Authorisation (CMA) and Accelerated Assessment (AA) pathways are fast-track routes enabling early European Medicines Agency (EMA) marketing approval. CMA is conditional on completing EMA-imposed postmarketing studies. AAs require full safety and efficacy data while shortening regulatory assessment time but have no imposed conditions. Surrogate outcomes (or endpoints), intended to predict desired clinical outcomes, reduce drug development time permitting drug discovery benefits to reach patients relatively quickly (12). Dr. Schuster's work focused on determining the numbers of CMAs and AAs granted according to clinical and surrogate outcomes, and assessing whether surrogates were validated and used the European Public Assessment Reports (EPARS) to assess CMAs and AAs during January 2011-January 2018. Dr. Schuster concluded that for two fast-track approval pathways, CMA and AA, nonvalidated surrogate outcomes supported most of the EMA authorizations issued between 2011 and 2018. While surrogate outcomes may expedite delivery of drug discovery benefits to the public, there is inherent uncertainty in using nonvalidated surrogates to support marketing authorizations. Post-authorization obligations imposed on CMAs address this to some extent. However, medicines granted AAs based on nonvalidated outcomes should be re-evaluated as clinical efficacy evidence emerges.

Using Zebrafish Larvae to Model Hemorrhagic Stroke

Ms. Siobhan Crilly, University of Manchester

Ms. Crilly started with an introduction to intracerebral hemorrhage (ICH), which is a type of stroke caused by the spontaneous rupture of blood vessels in the brain and is responsible for ~6% of all global deaths annually. Primary injury following an ICH event arises due to toxic blood components damaging neuronal cells in the brain parenchyma and secondary injury due to recruitment of immune cells to the site of injury, inducing swelling and increasing pressure on the intracerebral structures. Mammalian disease models have been employed thus far to investigate disease mechanisms, however a zebrafish larval model of ICH offers many experimental advantages such as enabling noninvasive live imaging and observation of pathological cellular events occurring in real time. Ms. Crilly described the development of an in vivo model using the bubblehead genetic mutant zebrafish larvae to model spontaneous ICH. She described the utilization of this model to examine the outcomes of interleukin-1 (IL-1) inhibition on disease progression. Ms. Crilly's data indicated that the pathological response to brain bleeding is conserved between zebrafish and humans in the context of ICH, thereby validating the use of this in vivo system as an alternative approach for studying ICH preclinically. She also showed that chemical inhibition of IL-1β is protective against disease pathology in this model.

Beyond the Hype, How AI is Disrupting Drug Discovery and Development

Dr. Beeta Balali-Mood, Mood Biopharma Consulting

In her presentation, Dr. Balali-Mood highlighted the efforts to investigate why the pharmaceutical industry is lagging behind in adopting this approach and discussed the potential of non-pharma technologies. Artificial intelligence (AI) is gaining traction at several stages of development in the drug discovery process, especially in target selection, data mining and identification of drug–disease connection and relevant biomarkers as predictive tools for efficacy. Another approach involves processing data from diseased and healthy tissues and identifying targets through supervised learning. In lead discovery, progress has been made in studying target engagement and prediction of drug-like properties. Dr. Balali-Mood highlighted synthetic accessibility of small molecules as one of the areas where machine learning could have a significant impact and mentioned that the challenge here is lack of sufficient data and more fundamentally, unrecorded data. AI has the potential to revolutionize drug discovery. With her talk, Dr. Balali-Mood intended to raise awareness about the machine learning tools at hand. Furthermore, Dr. Balali-Mood highlighted the importance to both academia and industry in leveraging the potential of non-pharma technologies to streamline

the drug discovery process and ultimately bring benefit to patients.

Characterizing the Catalytic Mechanism of a Methyltransferase for Use in Oncology Drug Discovery

Ms. Alice Eddershaw, AstraZeneca

Understanding the catalytic mechanism of target enzymes in drug discovery enables development of balanced assays for hit finding screens and aids profiling mechanisms of new inhibitors, so it could improve translation to the clinic. The objective of Ms. Eddershaw's study was to elucidate the mechanism of a methyltransferase (MT), an attractive enzyme class of epigenetic oncology targets. Ms. Eddershaw described the use of a discontinuous MTase-Glo (Promega) assay to study human MT with two substrates—*S*-adenosyl methionine (SAM) and a 21-residue histone peptide with a methyl-accepting residue—and showed that initial velocity data fitted best to the Ping Pong model suggesting formation of a methyl-bound MT intermediate. Product inhibition was consistent with the Theorell Chance, Ping Pong, and Rapid Equilibrium random mechanism with dead-end EAP and EBQ complexes. Lack of uncompetitive inhibition by dead-end inhibitors indicated random order substrate binding, ruling out Ping Pong and Theorell Chance mechanisms.

Discovering New Therapeutic Options for Chronic Respiratory Disease

Prof. Maria Belvisi, AstraZeneca and Imperial College

Following a brief outline of an interesting career trajectory resulting in her current dual role as an academic researcher and the leader of a major pharmaceutical company respiratory research group, Prof. Belvisi discussed several themes during her talk.

Firstly, Prof. Belvisi outlined the extent of the challenge for research in respiratory diseases highlighting the enormous and increasing healthcare and economic burden across Europe with over 600,000 deaths a year and 6 million hospital admissions with total costs exceeding EUR 380 billion per year (in 2013) (13). Globally, respiratory diseases are among the leading causes of death, with chronic obstructive pulmonary disease (COPD) now the fourth and fifth ranked cause of death in Western Europe and worldwide, respectively (14). More effective treatments are needed for patients with severe asthma who cannot be controlled on existing therapies. While for example cancer research benefits from a large charity sector spend of some 30% of economic burden, respiratory is much less well served with only 0.2% comparable spend.

As a second theme, Prof. Belvisi discussed the challenges of bringing new respiratory medicines to market with reference to changes in focus undertaken by AstraZeneca to increase research productivity. Despite the significant burden, respiratory medicine appears to have fewer new approved therapies than other common disease areas such as cardiovascular, metabolic and neurological diseases with fewer drug candidates and a higher failure rate. Respiratory is identified as one of the highest risk therapeutic areas in terms of attrition rates during development with a 3% success rate compared with 6-14% for other disease areas. This problem is probably multifactorial with many factors coming into play such as lack of mechanistic understanding, poor translation from preclinical to clinical studies, difficulties of developing inhaled therapies and the lack of investment in respiratory research.

To reduce failure rates, AstraZeneca embarked on a major revision of its R&D strategy with the aim of improving R&D productivity, which was below the industry average in 2005-2010. The main conclusion was to focus decision-making on 5 technical determinants (right target, tissue, safety, patient, commercial). A sixth determinant was subsequently identified as the right culture. In a recent publication assessing the success of the application of this 5R strategy, AstraZeneca is now shown to be among the most productive of the research organizations studied (15, 16).

In the third theme of her presentation, Prof. Belvisi outlined the strategy being developed within AstraZeneca to address unmet needs in chronic lung diseases by focusing on novel and diverse drug discovery for both treatment and disease modification. Approaches cover a range of modalities, biologics, small molecules, immunotherapeutics, protein engineering and perhaps unique for this therapeutic area, the key role of delivery devices. The strategy involves both internal research but also collaborations with a wide range of academic and biotech groups.

For example, a collaboration with Bicycle Therapeutics is looking at the role of bicyclic peptides in modulating protein–protein interactions. Ethris is addressing delivery of stabilized mRNA and Dynavax is collaborating with AstraZeneca on oligonucleotide TLR9 agonist **AZD-1419**, in phase IIa study for asthma. The final example was given in the form of **AZD-14202**, an inhaled IL-4Rα antagonist aimed to disrupt the key drivers of T2 asthma, which is in early clinical development.

Finally, the importance of diagnostics was highlighted by the approach to develop diagnostics available in the doctor's surgery to give increased confidence in the use of new and inevitably expensive medicines.

Disclosures

The authors are in paid employment of their respective organizations. T. Blackburn is both a member of the BPS and SMR which no remuneration is paid. F. Fallah-Arani and

R.A. Porter are Society for Medicines Research Committee members for which no remuneration is paid.

References

1. Tinworth, C.P., Lithgow, H., Churcher, I. *Small molecule-mediated protein knockdown as a new approach to drug discovery.* Med Chem Comm 2016, 7(12): 2206-16.
2. Honigberg, L.A., Smith, A.M., Sirisawad, M. et al. *The Bruton tyrosine kinase inhibitor PCI-32765 blocks B-cell activation and is efficacious in models of autoimmune disease and B-cell malignancy.* Proc Natl Acad Sci U S A 2010, 107(29): 13075-80.
3. Pan, Z., Scheerens, H., Li, S.J. et al. *Discovery of selective irreversible inhibitors for Bruton's tyrosine kinase.* ChemMedChem 2007, 2(1): 58-61.
4. Bunnage, M.E., Chekler, E.L., Jones, L.H. *Target validation using chemical probes.* Nat Chem Biol 2013, 9(4): 195-9.
5. Workman, P., Collins, I. *Probing the probes: fitness factors for small molecule tools.* Chem Biol 2010, 17(6): 561-77.
6. Arrowsmith, C.H., Audia, J.E., Austin, C. et al. *The promise and peril of chemical probes.* Nat Chem Biol 2015, 11(8): 536-41.
7. Traynelis, S.F., Wollmuth, L.P., McBain, C.J. et al. *Glutamate receptor ion channels: structure, regulation, and function.* Pharmacol Rev 2010, 62(3): 405-96.
8. Wolosker, H., Sheth, K.N., Takahashi, M., Mothet, J.P., Brady, R.O., Jr., Ferris, C.D., Snyder, S.H. *Purification of serine racemase: biosynthesis of the neuromodulator D-serine.* Proc Natl Acad Sci U S A 1999, 96(2): 721-5.
9. Michel, M.C., Charlton, S.J. *Biased agonism in drug discovery-is it too soon to choose a path?* Mol Pharmacol 2018, 93(4): 259-65.
10. Moyes, A.J., Khambata, R.S., Villar, I. et al. *Endothelial C-type natriuretic peptide maintains vascular homeostasis.* J Clin Invest 2014, 124(9): 4039-51.
11. Veale, C.A., Alford, V.C., Aharony, D. et al. *The discovery of non-basic atrial natriuretic peptide clearance receptor antagonists. Part 1.* Bioorg Med Chem Lett 2000, 10(17): 1949-52.
12. Ciani, O., Buyse, M., Drummond, M., Rasi, G., Saad, E.D., Taylor, R.S. *Time to review the role of surrogate end points in health policy: state of the art and the way forward.* Value Health 2017, 20(3): 487-95.
13. Gibson, G.J., Loddenkemper, R., Sibille, Y. et al. The European Lung White Book: Respiratory Health and Disease in Europe. European Respiratory Society (ERS): Sheffield 2013.
14. Lozano, R., Naghavi, M., Foreman, K. et al. *Global and regional mortality from 235 causes of death for 20 age groups in 1990 and 2010: a systematic analysis for the Global Burden of Disease Study 2010.* Lancet 2012, 380(9859): 2095-128.
15. Cook, D., Brown, D., Alexander, R., March, R., Morgan, P., Satterthwaite, G., Pangalos, M.N. *Lessons learned from the fate of AstraZeneca's drug pipeline: a five-dimensional framework.* Nat Rev Drug Discov 2014, 13(6): 419-31.
16. Morgan, P., Brown, D.G., Lennard, S. et al. *Impact of a five-dimensional framework on R&D productivity at AstraZeneca.* Nat Rev Drug Discov 2018, 17(3): 167-81.