IonoChem

Improved Efficacy Through Lipophilisation

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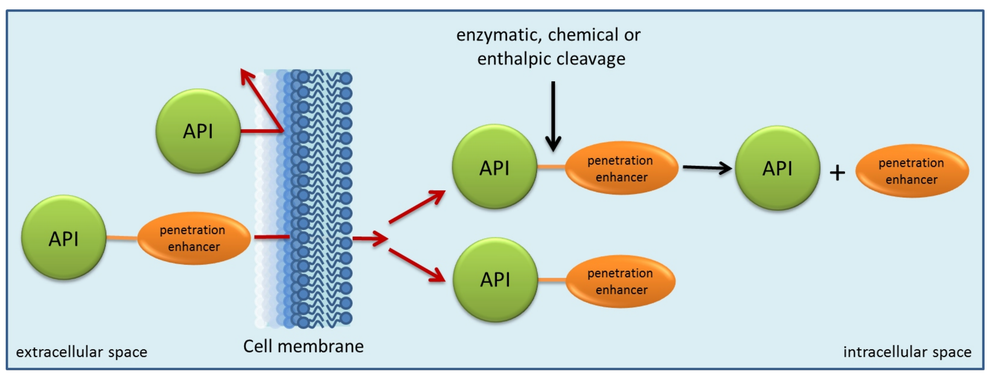
Approach

Attrition rates of drugs are still extremely high, and this is mainly due to lack of efficacy or for reason of toxicity. Arguably these issues are mainly due to the fact that drug candidates are typically isolated on the basis of their potency in a screen against a molecular target, and only subsequently are they tested in organisms *in vivo*. Since most modern targets have enjoyed a rather high degree of validation, it is likely that the issue of ostensible potency *in vitro* but lack of efficacy *in vivo* is not so much with the target but with the ability of drugs to reach and thereby interact with their target.

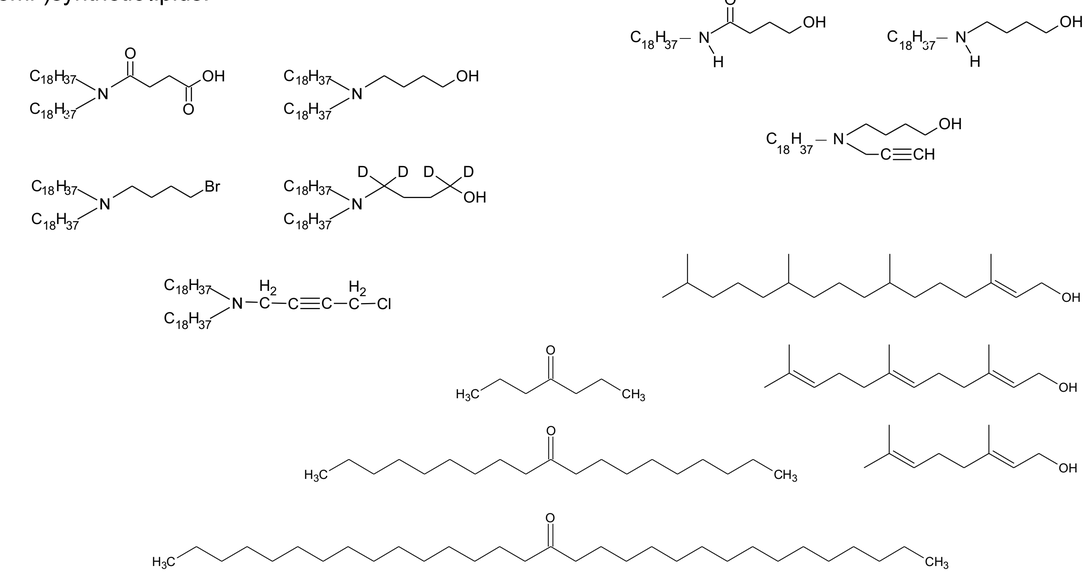
Since most drugs address intracellular targets they have to cross cellular membranes. These lipophilic barriers are especially for hydrophilic compounds an intractable obstacle leading not only to a poor bioavailability but also a too short biological half-life of Active Pharmaceutical Ingredients (APIs).

IonoChem addresses these issues and offers various methods to improve membrane permeation capabilities of APIs. One approach is the design and formulation of lipid based compounds, another an equivalent prodrug concept (**below**). Common ground is a selective derivatisation of APIs with a broad spectrum of lipohilic moieties, which are covalently coupled to the metabolic active structure. These penetration enhancers comprise mono- and double-tailed natural and (semi-)synthetic lipids, that can be introduced into various positions of therapeutic molecules.



Lipid Tool Box

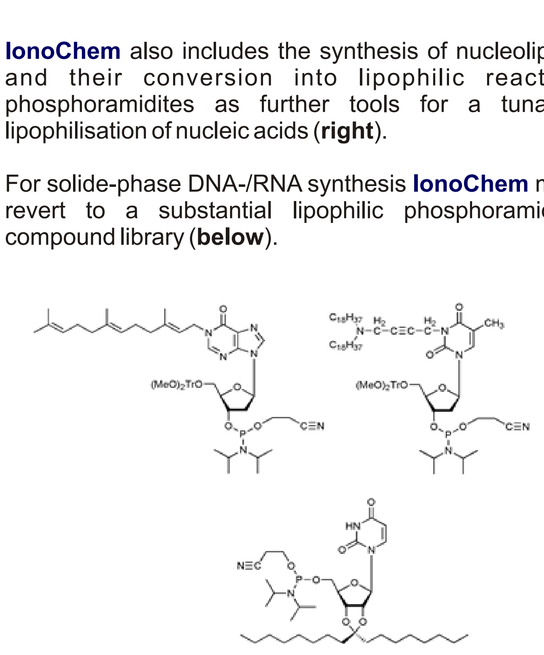
IonoChem offers a broad spectrum of lipids for the lipophilisation of APIs. It comprises various classes of lipids ranging from natural plant origin, like isoprenoids to functionalised mono- and double tailed natural or (semi-)synthetic lipids.

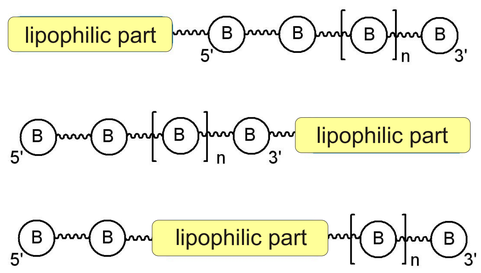


Lipophilic Oligonucleotides

IonoChem also includes the synthesis of nucleolipids and their conversion into lipophilic reactive phosphoramidites as further tools for a tunable lipophilisation of nucleic acids (**right**).

For solide-phase DNA-/RNA synthesis **IonoChem** may revert to a substantial lipophilic phosphoramidite compound library (**below**).





Delivery of therapeutic molecules of complex nature like small-interfering (si)RNAs across plasma membranes remains one of the largest obstacle for their *in vivo* application.

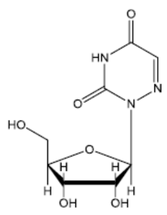
IonoChem adresses this challenge by the creation of a specific lipo-DNA as a trafficking tool for the targeted membrane transition of siRNAs (**below**).

Literature and further readings

- PCT/EP2013/069936, Reactive, lipophilic nucleoside building blocks for the synthesis of hydrophobic nucleic acids.
- Knies C., (2013), Synthese und onkologische Testung von Nukleolipiden verschiedener Pyrimidinnukleoside. Master Thesis, University of Osnabrück.
- Köstler, K., Werz, E., Malecki, E., Montilla-Martinez, M. and Rosemeyer, H. (2013), Nucleoterpenes of Thymidine and 2′-Deoxyinosine: Synthons for a Biomimetic Lipophilization of Oligonucleotides. Chemistry & Biodiversity, 10: 39–61.
- Werz, E., Viere, R., Gassmann, G., Korneev, S., Malecki, E. and Rosemeyer, H. (2013), Synthesis of Thymidine, Uridine, and 5 Methyluridine Nucleolipids: Tools for a Tuned Lipophilization of Oligonucleotides. Helv. Chim. Acta, 96: 872–888.

Improved Efficacy of 6-Azauridine

6-Azauridine (z^6U), a synthetic triazine nucleoside and uridine structure analog, is a potent chemotherapeutic agent for the treatment of leukemia, especially of chronic myeloid leukemia (CML). However, 6-Azauridine had to be withdrawn from the market because of severe side effects, like e.g. the lost of motor coordination or sedative effects on the central nervous system. These side effects were aided and abetted by the observed high renal elimination rates of up to 75%, which required high administration rates with high partial doses.

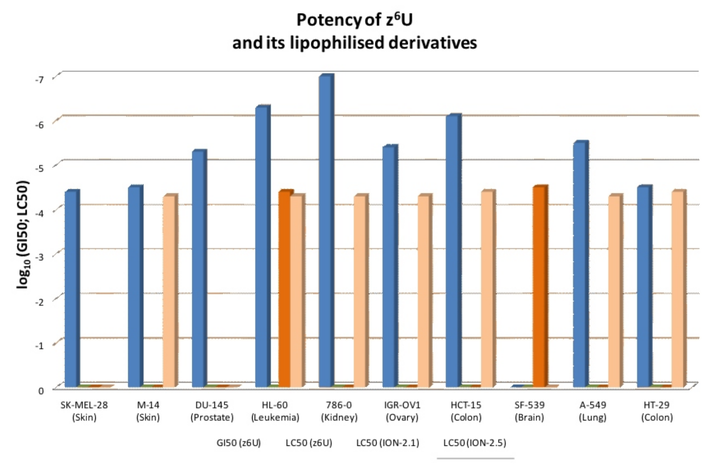


6-Azauridine

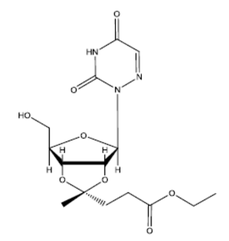
In cooperation with the European ScreeningPort Hamburg various 6-Azauridine derivatives were tested against 10 different human cell lines from the *NCI 60 panel.* Their selective inhibition of cell proliferation and potency to induce cell death were evaluated by means of a standardised luminescent cell viability assay (CellTiterGlo®, Promega) on a Multimode Plate Reader (EnSpire®, PerkinElmer) after 48 and 72 hours, calculating 50% growth inhibition (GI50), and 50% lethal (LC50) concentration values.

While unmodified z^6U shows within nearly all tested cell lines only cytostatic effects **(below**, **blue bars**), the lipophilised derivatives ION-2.1 and ION-2.5 are both highly cytotoxic (**orange bars**) with LC50 values between 30 - 50 µM.

ION-2.1: Improved potency and tumor selectivity



ION-2.1

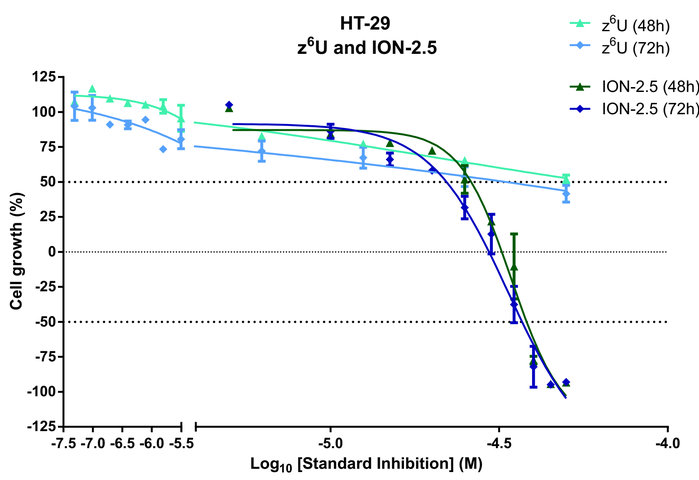


Ketalisation of the z^6U ribose unit with ethyl levulinate generates a highly selective antitumor nucleolipid with only HL-60 and SF-539 cells being sensitive (**dark orange**).

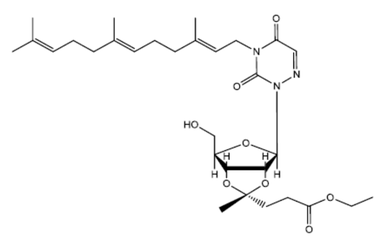
ION-2.5: Improved intrinsic activity

A further lipophilisation of z^6U by introduction of a farnesyl moiety leads to a broad activity within the testedt cell lines.

Remarkably, while z^6U shows only a slight cytostatic effect on HT-29 cells, ION-2.5 exerts a much stronger, nearly 100% toxic effect on this particular cell line (**below**).



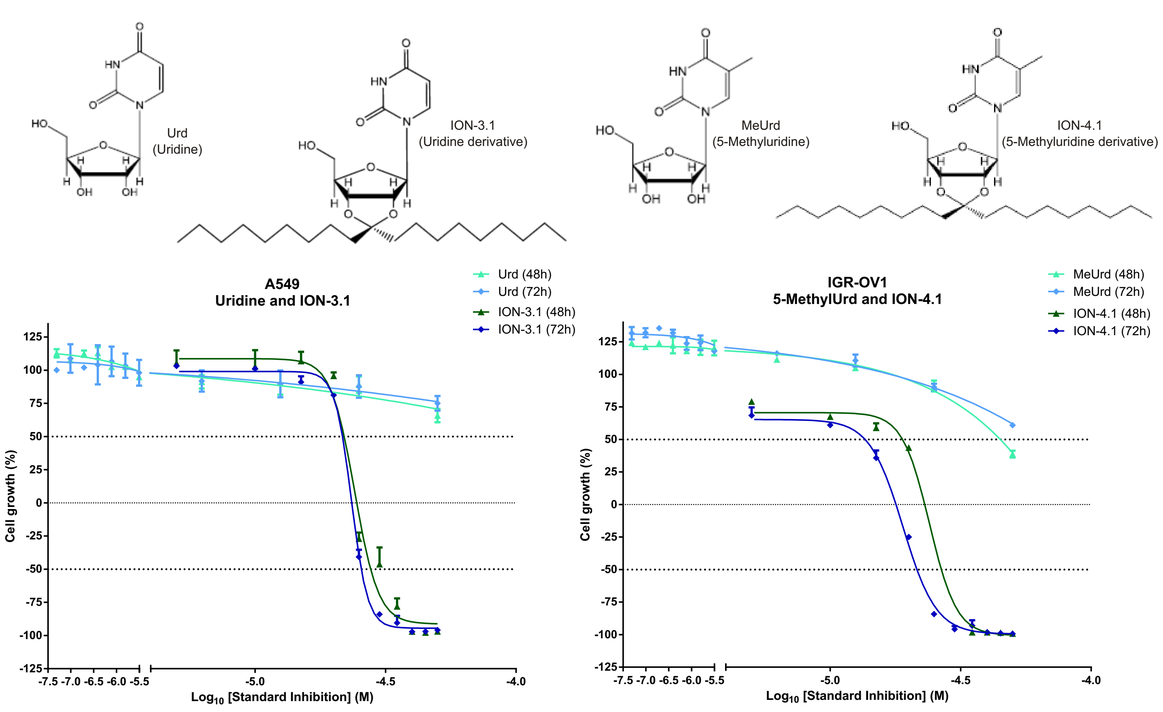
ION-2.5



Dose response of z^6U and ION-2.5 on the colon carcinoma cell line HT-29 after 48 and 72 hours incubation.

Acquired Cell Toxicity Through Lipophilisation

A condensation reaction of the two non-toxic nucleosides uridine and ribothymidine with the also non-toxic near-naturally lipid nonadecane-10-one leads to nucleolipids with high cell toxic properties. ION-3.1 as well as ION-4.1 both show activity within all tested cell lines with ION-3.1 having the most pronounced, nearly 100% toxic effect within the lung carcenoma cell line A-549 and ION-4.1 in the ovarian cell line IGR-OV1.



Cooperations

European ScreeningPort

University of Osnabrück

European ScreeningPort Hamburg GmbH

UNIVERSITÄT OSNABRÜCK