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SHORT REVIEW

Enhancing the Properties of Natural Products and Other Drugs: Deuterium: A Novel Approach

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Abstract

Deuterium substitution is a new approach used to enhance the metabolic profile of a drug. The carbon-deuterium bond seems to be stronger than a usual carbon-hydrogen bond. It leads to improved biological half-life and prolonged action of the drug. The deuterated drugs also show improved pharmacokinetics of the drug and reduce the dosing frequency. This paves the way for drugs from natural sources with good therapeutic effects but poor pharmacokinetic profiles, which can be deuterated for improved properties. Though this seems to be an alternate pathway, the isotope exchange with hydrogen has to be studied well for toxicity and the safety profile of the drug should be evaluated. The present review provides an outline of the deuterium approach in natural products and other drugs which are opted for deuterium to improve the metabolic profile.

Keywords: Deuterium, Kinetic Isotopic Effect, Metabolism, Natural Compound

1. Introduction

The drug discovery process is time-consuming and expensive and out of every ten thousand molecules, only one becomes a drug. The main reason for this is poor metabolic stability and toxicity[1]. Medicinal chemists employ a variety of techniques to improve the efficacy and safety of compounds in the development process. One approach is bioisosterism, in which one substructure is replaced with another to improve one or more features of the parent drug while retaining its biological action[2]. For example, the replacement of hydrogen by a deuterium atom is considered a novel approach to alter the Absorption, Distribution, Metabolism, and Excretion (ADME) properties of drug molecules[3]. Deuteration is a chemical reaction involving the replacement of a covalently bonded atom with a deuterated atom. The typical type is a reaction involving hydrogen and deuterium. An isotope of hydrogen that occurs naturally is stable, nonradioactive, and has similar chemical and physical properties as deuterium. Thus, the distinctive feature of selective deuteration retains the therapeutic effect of a drug[4]. From the perspective of the drug development process, one of the important properties of deuterium is that due to its higher atomic mass than hydrogen, it forms an exceptionally strong bond. It is challenging to break a deuterium-carbon connection since it is almost 6–10 times more stable than the isotope hydrogen-carbon link[5-6]. When one of the reactant atoms is replaced with one of its isotopes, the Kinetic Isotope Effect (KIE), is a relative change in the reaction rate, occurs. It is the ratio of the rates of reaction between the reactants that have been substituted with hydrogen (kH) and deuterium (kD)[4]. This aspect may affect interactions between deuterated drug molecules and the enzyme systems involved in drug metabolism[7]. The C-D bonds are more resistant to oxidative processes because of the Deuterium Kinetic Isotope Effect (DKIE), which has a significantly higher potential. This gives deuterated medications an advantage in metabolism[8]. Deuteration of drugs decreases the rate of metabolism, particularly oxidation in the gastro intestine or liver and may cause the original drug to

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enter into the systemic circulation thus increasing the bioavailability. Mostly, the systemic clearance rate remains unaffected. The advantage of a drug with this property is less frequent dosing and less metabolite production. Because gastrointestinal irritation refers to the amount of medicine administered rather than the concentration of the drug in the blood, this impact may serve to improve tolerability[9]. As a result, deuterium inclusion reduces the metabolism of certain medications with the pathways involving hydrogen-carbon bonds[10]. In the present review the possible application of deuterium on the enhancement of pharmacokinetic properties of natural compounds and other drugs is discussed (Figure 1).

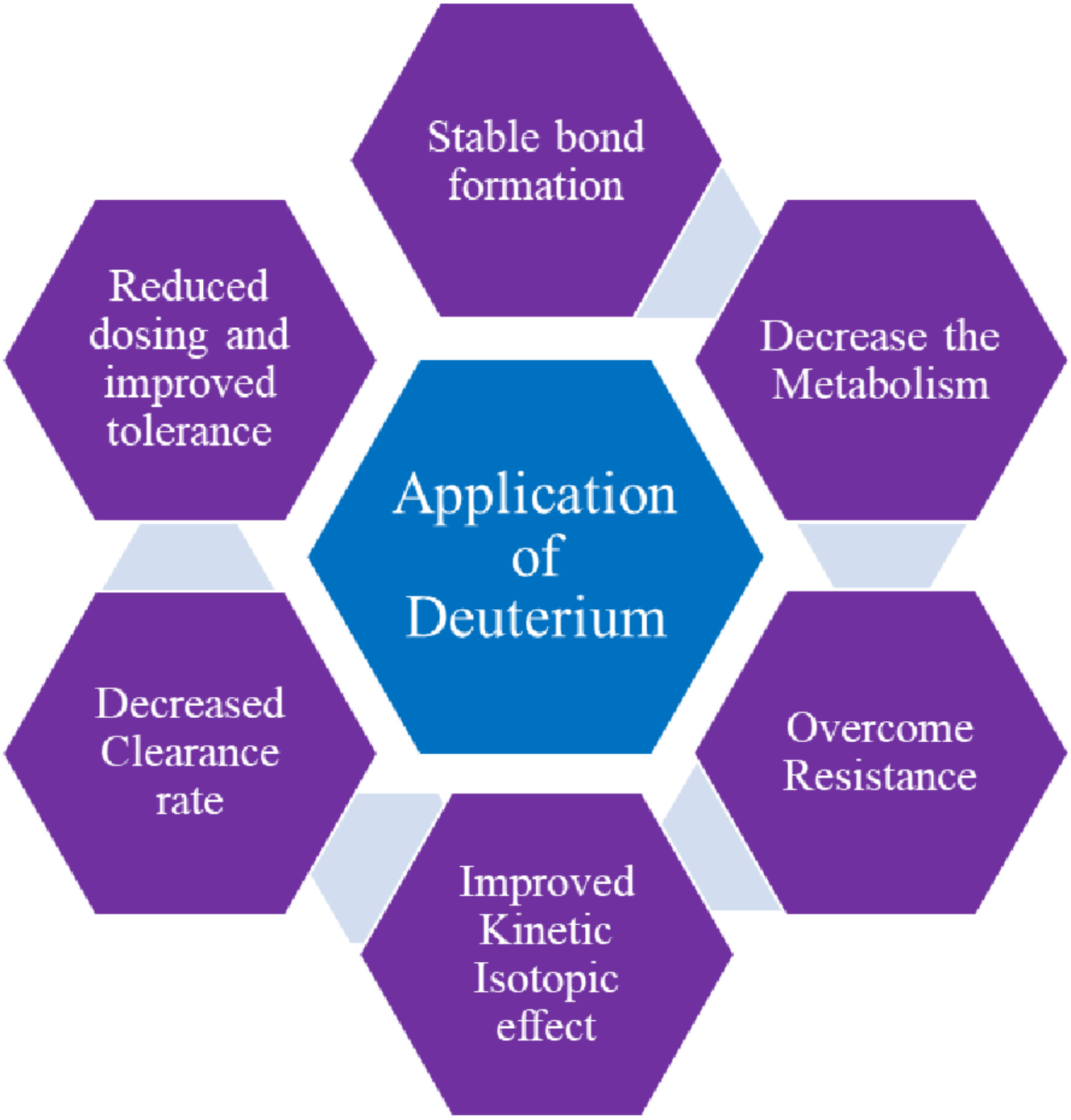


Figure 1. Application of deuterium to improve the pharmacokinetic properties of drugs.

2. Deuterium an Alternative Approach for Natural Products

In the case of anticancer drugs, various natural substances exhibit *in vitro* preclinical investigations but several obstacles persist in converting these outcomes into *in vivo* or clinical trials, leading to failed outcomes. Because of their low water solubility, poor absorption, poorer bioavailability, and shorter retention duration in the biological system, natural compounds have not yet reached their full therapeutic potential despite their amazing health advantages. Following ingestion, natural agents must engage with diverse physico-chemical barriers that may modify their composition and impact their antitumor efficacy. To preserve the natural compound's properties, new synthetic techniques are used to stop the compound's breakdown and maintain its parent structure[11]. The natural product drugs which are potential but have some issues related to pharmacokinetic properties are discussed further so that deuterium can be an alternative approach to address these issues.

For example, although Curcumin has been shown to have potential, its low bioavailability has hindered its usage in clinical settings and is responsible for one of the main observations of low serum levels following oral treatment[12]. Moreover, its rapid metabolism, limited absorption, and quick excretion from the human body pose the biggest obstacles to its widespread usage. It is important now to find a novel approach to overcome this issue[13]. The low bioavailability of Curcumin, its solubility and stability, the ideal dosage, pharmacokinetics, and the mechanism of action for a particular disease are still emphasised as key recommendations for future research, despite its efficacy and safety[14].

The limited solubility of Parthenolide in water and resulting lower bioavailability limits its potential clinical application as an anticancer medication, leading to relatively weak pharmacological qualities. Nevertheless, several Parthenolide derivatives were developed to address this problem. While Resveratrol has demonstrated some effectiveness in cancer patients, its usage is restricted by its low bioavailability. Consequently, attempts have been undertaken to alter Resveratrol to increase its bioavailability and decrease its toxicity[15].

The dosage of Vincristine and Vinblastine is restricted and usually administered as a single dose due to their neurotoxicity. Alkaloids also have limited water solubility and low bioavailability, which makes it challenging to obtain therapeutic dosages that are effective in the tumour target. Therefore, new approaches are required to raise their bioavailability. Vinblastine's gastrointestinal tract absorption is not always consistent. The medication swiftly leaves the bloodstream after intravenous delivery and is absorbed by the tissues. Vinblastine cannot reach therapeutic concentrations in the cerebrospinal fluid and only partially passes the blood–brain barrier. The active metabolite des-acetylvinblastine is formed as a result of extensive metabolism[16].

Although Menthol has been shown to have significant anticancer efficacy, its clinical application has been severely limited due to some issues, including poorly established toxic side effects and pharmacokinetics. Only a small number of studies have evaluated the toxicity and pharmacokinetics of Menthol, even though it has been extensively researched for its potential application in cancer prevention and treatment[17].

Paclitaxel, an anticancer natural product, has a few drawbacks, including low hydro solubility, present adverse effects, and the likelihood of treatment resistance in patients. Despite the proposal of a few possible mechanisms, such as mutations or changes in the binding regions, the establishment of multidrug resistance that has limited its therapeutic efficiency remains largely unexplained. Camptothecin, a naturally occurring chemical with potent anticancer effects, was isolated from the bark of the *Camptothecin acuminata* plant in 1966. Some of the medication's limitations are due to its poor solubility and stability. This is why several stable derivatives are currently being studied in clinical trials. Gingerol, a phenolic compound derived from the young *Zingiber officinale* rhizome, is another phenolic molecule with potent anticancer properties. Gingerol has been shown to have limited water solubility and bioavailability, limiting its widespread application[18].

Although the low aqueous solubility, rapid catabolism, low bioavailability, and poor intestinal absorption of natural compounds limit their clinical applications, various efforts have recently been undertaken to address these drawbacks[19]. One approach among them can be deuterium which is novel and can

cause changes to alter and improve clinical application of these drugs.

Plinabulin is generated from "diketopiperazine phenylahistin," a naturally occurring bioactive chemical found in the sea that has been shown to depolymerize microtubules. Furthermore, Plumbabulin increases cancer mortality, reduces blood supply to tumours, and inhibits angiogenesis. Plunabulin, on the other hand, did not show any efficacy due to its poor pharmacokinetic profile. As a result, MBRI-001, a deuterated plinabulin derivative with improved Pharmacokinetics (PK) and anticancer properties, was created. MBRI-001 quickly finds a home in several organs, with the lung having the largest concentration when compared to other tissues. MBRI-001 was more stable *in vitro* than plinabulin in rat and liver microsomes. Plurabulin was less effective in treating microtubule hepatocellular carcinoma than MBRI-001[20]. Curcumin is beneficial in the treatment of Alzheimer's disease, metabolic syndrome, arthritis, cardiovascular disease, anxiety, neurodegenerative disorders, multiple sclerosis, allergies, AIDS, inflammatory bowel disease, diabetes, nephrotoxicity, psoriasis, lung fibrosis, hyperlipidemia, and antibacterial properties.

Although Curcumin is a safe natural product, preclinical research revealed that it cannot be used to treat any disease due to its low bioavailability, insufficient solubility in aqueous solvents, fast metabolism, and systemic elimination. The hydrophobic properties of Curcumin, such as quick metabolism and faster removal by the liver, are the primary reasons for its low bioavailability. As a result, researchers have conducted extensive research to overcome this disadvantage and improve Curcumin bioavailability using a variety of strategies, such as improving Curcumin solubility, developing Curcumin nanoparticles and liposomal formulations, developing new derivatives, and analogues, and designing hybrid Curcumin molecules. Several studies have shown that deuterated chemicals improve a drug's pharmacokinetic characteristics.

Deuterated substances can affect the drug's pharmacokinetic profile and minimise toxicity by increasing chemical stability. The *in-silico* drug activity prediction of deuterated Curcumin followed Lipinski's 'Rule of Five,' and Swiss ADME data revealed that the Log P and Log S values showed a difference. This difference in the value of partition coefficient and solubility could be mainly due to the exchange of isotope deuterium instead of hydrogen. The slight change in pharmacokinetic profile is related to the difference in bond character between C-H and C-D. As a result, exchanging hydrogen for deuterium at the right place in the chemical structure can considerably slow down metabolism, potentially increasing plasma drug concentration[21].

2.1 Deuteration of Vitamin A

ALK-001 is a deuterium isotope replacement of Carbon-20 in a modified version of vitamin A that is C20-D3-Retinyl Acetate or C20 deuterated vitamin A. It is intended to inhibit vitamin A dimerisation. The accumulation of toxic byproducts is reduced when dangerous vitamin A dimers are reduced. Administration of C20 deuterated vitamin A slowed the rate of A2E production in an animal model. ALK-001 is now being tested in a phase II/III trial in patients with geographic atrophy[22].

3. Other Drugs Being Studied to Improve Metabolism by Deuteration

3.1 Brecanavir

A tyrosyl peptidomimetic that had action against both wild-type and protease inhibitor-resistant HIV was discovered in 2006[23,24]. The drug was found to be safe and well tolerated in clinical studies but low solubility and extensive metabolism seem to be a challenge. Ritonavir, a pharmacoenhancer, was recommended for coadministration to enhance the metabolic profile. However, it is preferable to develop Brecanavir analogues that have a distinctive metabolic profile without using a pharmacoenhancer due to probable side effects and drug-drug interactions. Deuterium can be used in place of hydrogen to improve the ADME features of Brecanavir, where Carbon-Hydrogen bond breaking is the rate-limiting step during metabolism[25]. Brecanavir, a protease inhibitor, has been studied with highly deuterated analogues (Figure 2.) and has been assessed for improvements in metabolic stability However, they had substantially the same *in vivo* character as parent Brecanavir when administered intravenously to rats. One significant modification is that the introduction of deuterium appears to have changed the metabolic profile, but the overall effect of high clearance has not changed[26].

3.2 Clopidogrel

Clopidogrel is a thienopyridine antiplatelet drug used for the diagnosis of cardiovascular diseases such as angina and myocardial infarction[27]. It is available as a prodrug which gets converted into an active drug by the CYP450 enzymes present in the liver[28,29]. The main disadvantage is a majority of Clopidogrel (85%) gets hydrolysed by esterase enzymes to inactive acid and only a small amount of the Clopidogrel left is changed to the active metabolite[30,31]. CYP2C19, a P450 isoenzyme metabolizes the reactions[32] but reduced exposure of metabolisers for CYP2C19 produces less active metabolite leading to little inhibition of platelets. This leads to the development of resistance in Clopidogrel[33,34]. To overcome this resistance, a strategy applied is a synthesis of deuterated analogues of Clopidogrel (Figure 2).

The reaction of Clopidogrel-d3 (Figure 3) turns out to be slower than Clopidogrel in rat whole blood in the *in vitro* condition at 37°C. Even after 70 minutes, the amount of parent Clopidogrel was much less and below the limit of deduction, in contrast to deuterated Clopidogrel, which was still detectable after 2 hours. To examine the blocking of ADP-induced platelet aggregation in rat blood, the antiplatelet effects of Clopidogrel and Clopidogrel-d3 were evaluated using Born's technique. Clopidogrel has a substantial inhibitory impact at a dose of 78 µmol/kg, but only a weak inhibitory effect at a dose of 7.8 µmol/kg. Clopidogrel-d3, on the other hand, is a strong inhibitor even at this modest dose. This seems like a positive outcome to improve the activity of Clopidogrel[35].

3.3 Enzalutamide (ENT)

Enzalutamide is a competitive androgen receptor inhibitor that affects nuclear translocation and binding of DNA, leading to prostate cancer cell death[36]. The FDA authorised it for the treatment of patients with Metastatic Castration-Resistant Prostate Cancer (mCRPC)[37,38]. The metabolic active site is the N-methyl moiety, which gets oxidised to hydroxymethyl-enzalutamide and then is converted to the N-demethyl metabolite by CYP450 enzymes. ENT is also hydrolysed through amide bond cleavage. Deuterated ENT (N-trideuteromethyl enzalutamide, d3-ENT) is an alternative approach similar to the structure of ENT. Deuterium substitution can change the metabolic pathway and increase the exposure to the drug in the body. Hence a study was carried out to evaluate the effect of deuterated ENT (Figure 2). The maximum plasma concentration, Cmax was found to be higher in deuterated analogues of ENT to the orally administered rats in comparison with non-deuterated drugs. Moreover, the AUC_{0-t} of d3-ENT seems to increase significantly in humans and thus the dosing regimen can be reduced accordingly[39].

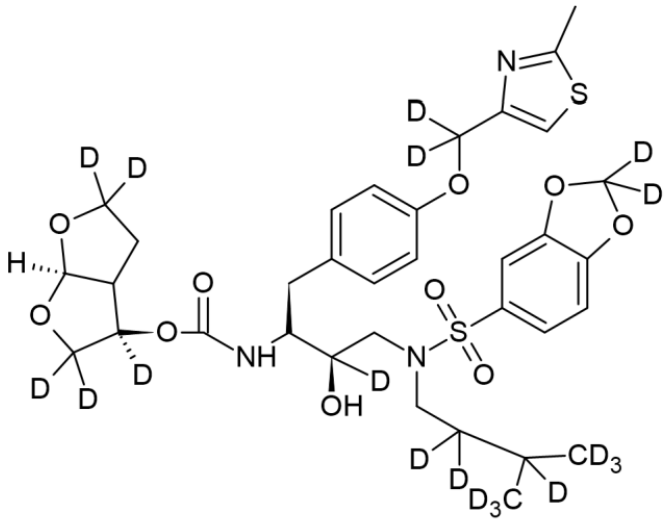
3.4 Apalutamide

ENT and Apalutamide (ARN-509) are similar in their structure and function. They are potent Androgen Receptor (AR) antagonists and have a higher affinity for AR receptors. Apalutamide crosses the Blood-Brain Barrier (BBB) less effectively than ENT, so the chances for the development of seizures are less[40-42]. Due to the similarity of ENT and Apalutamide in structure, to get a more stable AR antagonist, deuteration of Apalutamide was carried out (Figure 2). The deuterated Apalutamide had a similar affinity for the AR *in vitro* and there was a two-fold increase in the exposure of the drug in the rats. Further studies are required to transfer this pharmacokinetics profile to *in vivo* efficacy[43].

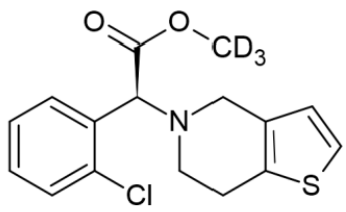
3.5 Vismodegib

The first oral and highly selective Hh signalling pathway targeting medication for basal cell cancer is called Vismodegib (GDC-0449). It inhibits the smoothened receptor by acting as a cyclopamine competitive antagonist, and this inhibition keeps the transcription factors Gli1 and Gli2 dormant, preventing the development of cancer-mediated genes in the Hh signalling pathway[44-46]. It is seen that oxidation of the pyridine or the middle benzene ring leads to the formation of three oxidative major metabolites. Hence to reduce the rate of metabolism, deuterated analogues were synthesised and studied[47,48].

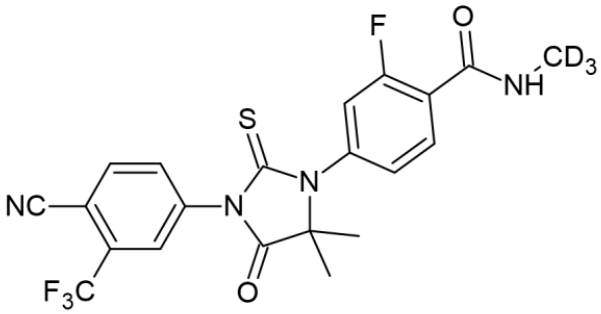
Deuterated compounds showed better pharmacokinetics than the prototype at the same dose. The SKLB-C2211 (Figure 2) compound was found to be more effective. It exhibited an almost 2.24-fold increase in plasma drug concentration when compared with Vismodegib. Moreover, the half-life (t1/2) of the deuterated molecule was also increased to 8.547 h by comparison with Vismodegib (5.617 h) indicating prolonged action. In addition, its peak concentration Cmax was observed to be



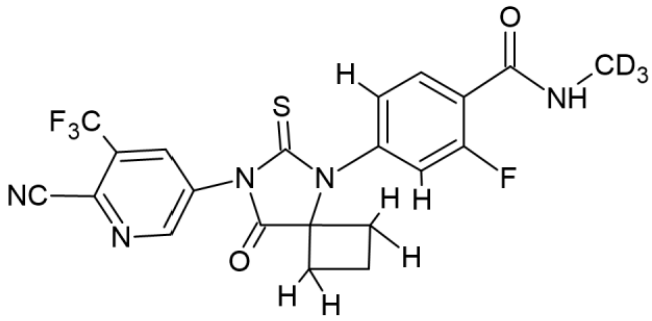
Deuterated Brecanavir



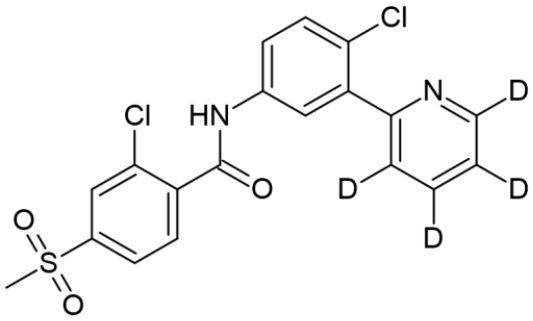
Deuterated clopidogrel



Deuterated enzalutamide



Deuterated apalutamide



Deuterated Vismodegib

Figure 2. List of deuterated drugs to improve metabolism.

higher than Vismodegib, suggesting that the dosage of SKLB-C2211 may be decreased in clinical practice. On the other hand, SKLB-C2211 took the same time for onset of action similar to the parent drug. It is important to note that this deuterated compound has a possible effect in clinical practice to reduce the dose and increase the pharmacokinetics[49].

3.6 Tivozanib

Vascular Endothelial Growth Factor (VEGF) stimulates the signalling pathway of endothelial cells by activating VEGF receptor (VEGFR) tyrosine kinases, which in turn play a significant role in tumour angiogenesis[50]. Tivozanib is strong and particularly it inhibits VEGFR, especially the tyrosine kinase receptors VEGFR-1, -2, and -3[51]. It has been proven to have antitumor effects in the liver, lungs, and colon renal cell carcinoma[51,52]. One drawback is that adverse events (AEs) like hypertension are more common with Tivozanib[53]. So, there is a need for analogues of Tivozanib to overcome the adverse event and have a better pharmacokinetic profile. Hence deuterated Tivozanib (HC-1144) (Figure 3) was synthesised and evaluated. The *in vivo* pharmacokinetics of deuterated tivozanib were studied in rats and it was seen that they exhibited better properties. It showed a 1.5-fold increase in plasma drug concentration as compared to Tivozanib. There is a change in half-life also which increased to (11.10 ± 1.2 h) as compared to that of Tivozanib (9.35 ± 1.09 h), showing HC-1144 had increased bioavailability and prolonged action. Furthermore, the Cmax of the deuterated molecule was found to be higher than Tivozanib, implying that the dosage of HC-1144 may be lowered in clinical use. Both drugs have similar peak times showing that the time taken for action did not change[54].

3.7 Tramadol

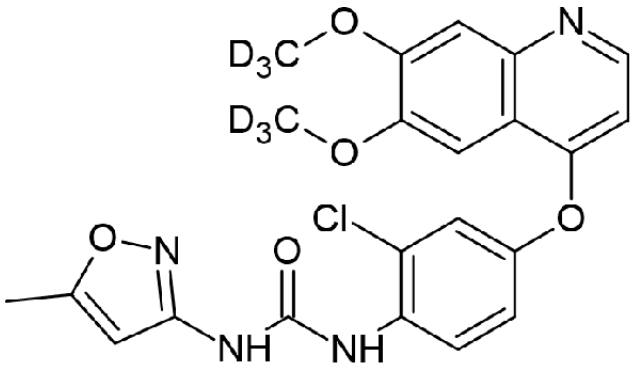
Tramadol (±)-cis can be used to treat moderate to severe pain[55]. It has a relatively short duration of action due to first-pass metabolism and hence is dosed as frequently as 100 mg every 4–6 h. It causes several side effects including constipation, nausea, dizziness, and somnolence. Hence there is a need for similar analogs of Tramadol to reduce the dosing and side effects. Deuterated Tramadol (Figure 3) was synthesized and evaluated for activity. The substituent for deuterium in place of hydrogen did not alter the *in vitro* binding affinity but the formation of primary metabolite is slowed down approximately by fivefold. However, the attempt to reduce the dosing and prolonged action did not turn up in deuterated Tramadol[56].

3.8 Rofecoxib

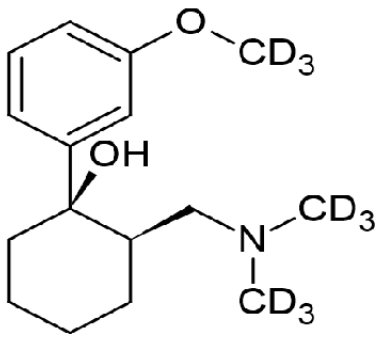
In acute or chronic inflammation, the cyclooxygenase produced is either COX-1 or COX-2[57]. COX-2 inhibitors are chemicals that are classified as Nonsteroidal Anti-Inflammatory Medicines (NSAIDs). They inhibit prostaglandin synthesis, particularly cyclooxygenase-2, at prescribed levels[58]. The systemic availability is low in the COX-2 inhibitor, hence deuterated analogues are synthesised and their pharmacokinetics are evaluated for increased plasma concentration. In the comparative analysis, the deuterated compound (BDD-11602) (Figure 3) exhibited superior plasma concentration. The deuterium isotopes in this position may diminish the first-pass metabolism caused by epoxidation in the 3',4'-position of deuterated Rofecoxib, resulting in improved systemic availability. Furthermore, the insertion of deuterium in positions 2', 3', 4', 5', and 6' of Rofecoxib's phenyl ring did not affect COX-2 inhibition. There was no apparent difference in the COX-2 inhibition of both the parent and the deuterated form of the drug[59].

3.9 Nevirapine (NVP)

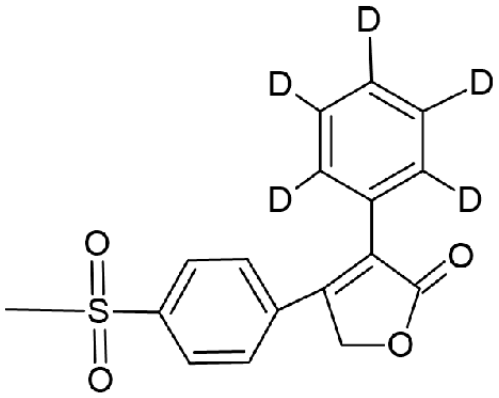
Nevirapine is a first-generation non-nucleoside reverse transcriptase inhibitor. NVP is currently prescribed for treating Human Immunodeficiency Virus (HIV) infections and preventing mother-to-child HIV transmission after childbirth[60,61]. NVP-induced hepatotoxicity incidence is reported which ranges from 6%[62]. Another data states 36.1% of incidence out of which 7.7% includes severe toxicity[63]. The reason for this toxicity seems to be the formation of O-hydroxy NVP metabolite circulating in the blood[64-66]. However, treatment of rats with tri deuterated NVP (12-D3NVP) (Figure 3) showed a decrease in the bioavailability in the blood and the formation of skin rash vs rats treated with NVP[65]. Hence deuterated NVP at position 12 was synthesised and evaluated. Deuterium substitution is shown to be effective in lowering hepatic P450 synthesis of 12-OHNVP in both human and mouse hepatocytes. This could be owing to the kinetic isotope effect of this



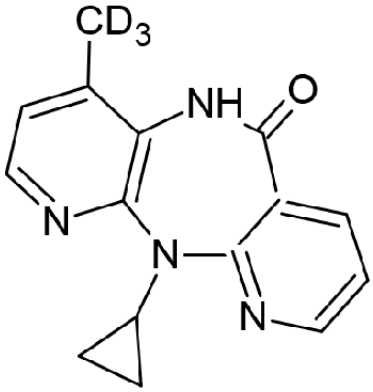
Deuterated Tivozanib



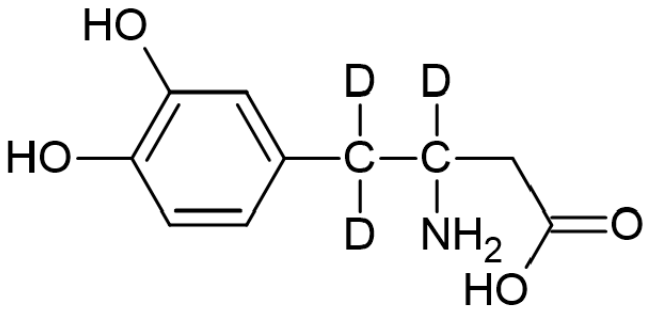
Dramadol- Deuterated form



Deuterated rofecoxib



12-D3NVP- Deuterated nevirapine



Deuterated levodopa

Figure 3. Deuterated analogue drugs to enhance metabolism.

substitution on active metabolite synthesis. Aside from this, metabolic switching at 400μM incubations with mouse primary hepatocytes was also detected. NVP-induced primary mouse hepatocyte cell death was decreased, but not reversed by 12-D3NVP. Taking this into consideration, tri-deuterated NVP is a possible approach towards reducing toxicity[66].

3.10 Levodopa

Parkinson's disease is typically treated with Levodopa, a precursor to Dopamine (DA), along with a peripheral DOPA Decarboxylase (DDC) inhibitor[67]. Combining L-DOPA with a DDC inhibitor over an extended period results in treatment-induced motor problems including wearing off, ON/OFF phenomena, freezing and dyskinesia[68]. There are some mechanisms proposed for this activity. One among them short half-life of Levodopa[69]. So based on the kinetic isotopic effect, selective deuteration of Levodopa was performed to slow down the metabolic rate[70]. It is seen that particular deuteration at the α and β positions (Figure 3) leads to reduced breaking of dopamine by Monoamine Oxidase. It also suggests that the Deuterated Levodopa exhibits prolonged action in comparison with the parent Levodopa by increased availability of DA in the brain[71].

4. Other Strategies to Improve Metabolism

4.1 Metabolic Soft Spot

When the functional groups of drugs are not sterically inhibited such as the benzylic C-H bond, allylic methyl and O-, N-, S-methyl groups are the most desired metabolic soft spot for P450 mediated metabolism. The intrinsic activity of drugs metabolised by cytochrome enzymes is largely determined by the nature of hydroxylation reactions catalysed by enzymes, the chemo and regioselectivity of substrate oxidation, as well as the rate of metabolism[72,73]. Thus, the potential to be a metabolic soft spot in metabolising enzyme systems will be determined by the intrinsic reactivity of the functional groups and the drug specificity of the specific molecule having this particular soft spot[74].

4.2 Prodrugs

Prodrugs frequently have functional groups that are easily broken enzymatically or chemically in the body, such as esters, amides, phosphates, carbonates, or carbamates. They are commonly used to conceal the polar or ionizable functional groups of active compounds and to enhance metabolism[75]. For example, active metabolites have been used to produce novel medications in some circumstances. Amitriptyline is a widely used medication for the treatment of mental illnesses such as depression and anxiety. Amitriptyline is demethylated by CYP2D6,3A4, and 2C19. This metabolite is a more effective and selective norepinephrine reuptake inhibitor than Amitriptyline against the norepinephrine transporter. As a result, the metabolite was developed and eventually became a commercial medicine known as Nortriptyline[76,77].

5. Conclusion

Advancement in research and development allows the development of novel drug candidates to overcome various problems associated with natural and other drug molecules. One among them is deuterated medicinal compounds which is a wide and novel approach to improve the metabolism of compounds. There are various deuterated molecules under study to improve their character. This approach seems to be possible but there are also challenges including differences in human and rat liver microsomes, metabolic pathways, mechanism of action, and half-life. To obtain a novel drug, there is a need to overcome these challenges so that deuterated compounds can come into clinical practice on a large scale.

6. References

1. Veljkovic V, Veljkovic N, Este JA, Huther A, Dietrich U. Application of the EIIP/ISM bioinformatics concept in development of new drugs. Current Medicinal Chemistry. 2007; 14(4):441-53. https://doi.org/10.2174/092986707779941014 PMid:17305545.
2. Meanwell NA. Synopsis of some recent tactical application of bioisosteres in drug design. Journal of Medicinal Chemistry. 2011; 54(8):2529-91. https://doi.org/10.1021/jm1013693 PMid:21413808.
3. Gant TG. Using deuterium in drug discovery: leaving the label in the drug. Journal of Medicinal Chemistry. 2014; 57(9):3595-611. https://doi.org/10.1021/jm4007998 PMid:24294889.
4. Mouli HC, Vinod A, Kumari S, Tiwari AK, Kathiravan MK, Peraman R. Deuterated driven new chemical entities: An

optimistic way to improve therapeutic efficacy. Bioorganic Chemistry. 2023. p. 106490. https://doi.org/10.1016/j.bioorg.2023.106490
5. Buteau KC. Deuterated drugs: unexpectedly nonobvious. J High Tech. L. 2009; 10:22.
6. Bell RP. Liversidge lecture. Recent advances in the study of kinetic hydrogen isotope effects. Chemical Society Reviews. 1974; 3(4):513-44. https://doi.org/10.1039/cs9740300513
7. Guengerich FP. Kinetic deuterium isotope effects in cytochrome P450 oxidation reactions. Journal of Labelled Compounds and Radiopharmaceuticals. 2013; 56(9-10):428-31. https://doi.org/10.1002/jlcr.3031 PMid:24285515 PMCid: PMC4861049.
8. Panda N, Zakki Uddin SK, Anjana GV, Ramalingam P, Palaniappan S, Mohan Maruga Raja MK, Kathiravan MK. Deuterium - A natural isotope to combat microbial resistance. In Journal of Natural Remedies. 2023; 23(2).
9. Pirali T, Serafini M, Cargnin S, Genazzani AA. Applications of deuterium in medicinal chemistry. Journal of Medicinal Chemistry. 2019; 62(11):5276-97. https://doi.org/10.1021/acs.jmedchem.8b01808 PMid:30640460.
10. Hok L, Mavri J, Vianello R. The effect of deuteration on the H2 receptor histamine binding profile: a computational insight into modified hydrogen bonding interactions. Molecules. 2020; 25(24):6017. https://doi.org/10.3390/molecules25246017 PMid:33353215 PMCid: PMC7766521.
11. Kashyap D, Tuli HS, Yerer MB, Sharma A, Sak K, Srivastava S, Pandey A, Garg VK, Sethi G, Bishayee A. Natural product-based nanoformulations for cancer therapy: Opportunities and challenges. In Seminars in Cancer Biology. 2021; 69:5-23. https://doi.org/10.1016/j.semcancer.2019.08.014 PMid:31421264.
12. Siviero A, Gallo E, Maggini V, Gori L, Mugelli A, Firenzuoli F, Vannacci A. Curcumin, a golden spice with a low bioavailability. Journal of Herbal Medicine. 2015; 5(2):57-70. https://doi.org/10.1016/j.hermed.2015.03.001
13. Gupta SC, Patchva S, Aggarwal BB. Therapeutic roles of curcumin: Lessons learned from clinical trials. The AAPS Journal. 2013; 15:195-218. https://doi.org/10.1208/s12248-012-9432-8 PMid:23143785 PMCid: PMC3535097.
14. Stanić Z. Curcumin, a compound from natural sources, a true scientific challenge-A review. Plant Foods for Human Nutrition. 2017; 72:1-12. https://doi.org/10.1007/s11130-016-0590-1 PMid:27995378.
15. Talib WH, Alsalahat I, Daoud S, Abutayeh RF, Mahmod AI. Plant-derived natural products in cancer research: Extraction, mechanism of action, and drug formulation. Molecules. 2020; 25(22):5319. https://doi.org/10.3390/molecules25225319 PMid:33202681 PMCid: PMC7696819.
16. Dhyani P, Quispe C, Sharma E, Bahukhandi A, Sati P, Attri DC, Szopa A, Sharifi-Rad J, Docea AO, Mardare I, Calina D. Anticancer potential of alkaloids: a key emphasis to colchicine, vinblastine, vincristine, vindesine, vinorelbine and vincamine. Cancer Cell International. 2022; 22(1):1-20. https://doi.org/10.1186/s12935-022-02624-9 PMid:35655306 PMCid: PMC9161525.
17. Zhao Y, Pan H, Liu W, Liu E, Pang Y, Gao H, He Q, Liao W, Yao Y, Zeng J, Guo J. Menthol: An underestimated anticancer agent. Frontiers in Pharmacology. 2023; 14:1148790. https://doi.org/10.3389/fphar.2023.1148790 PMid:37007039 PMCid: PMC10063798.
18. Garcia-Oliveira P, Otero P, Pereira AG, Chamorro F, Carpena M, Echave J, Fraga-Corral M, Simal-Gandara J, Prieto MA. Status and challenges of plant-anticancer compounds in cancer treatment. Pharmaceuticals. 2021; 14(2):157. https://doi.org/10.3390/ph14020157 PMid:33673021 PMCid: PMC7918405.
19. Dutta S, Mahalanobish S, Saha S, Ghosh S, Sil PC. Natural products: An upcoming therapeutic approach to cancer. Food and Chemical Toxicology. 2019; 128:240-55. https://doi.org/10.1016/j.fct.2019.04.012 PMid:30991130.
20. Belete TM. Recent updates on the development of deuterium-containing drugs for the treatment of cancer. Drug Design, Development and Therapy. 2022; 16:3465-72. https://doi.org/10.2147/DDDT.S379496 PMid:36217450 PMCid: PMC9547620.
21. Anjana GV, Kathiravan MK. Antimicrobial activity of curcumin and deuterated curcumin. JNR [Internet]. 2022; 22(3):424-31. https://doi.org/10.18311/jnr/2022/29686
22. Girgis S, Lee LR. Treatment of dry age-related macular degeneration: A review. Clinical Exper Ophthalmology [Internet]. 2023; 51(8):835-52. https://doi.org/10.1111/ceo.14294 PMid:37737509.
23. Miller JF, Andrews CW, Brieger M, Furfine ES, Hale MR, Hanlon MH, Hazen RJ, Kaldor I, McLean EW, Reynolds D, Sammond DM. Ultra-potent P1 modified arylsulfonamide HIV protease inhibitors: The discovery of GW0385. Bioorganic and Medicinal Chemistry Letters. 2006; 16(7):1788-94. https://doi.org/10.1016/j.bmcl.2006.01.035 PMid:16458505.
24. Hazen R, Harvey R, Ferris R, Craig C, Yates P, Griffin P, Miller J, Kaldor I, Ray J, Samano V, Furfine E. *In vitro* antiviral activity of the novel, tyrosyl-based Human Immunodeficiency Virus (HIV) type 1 protease inhibitor brecanavir (GW640385) in combination with other antiretrovirals and against a panel of protease inhibitor-resistant HIV. Antimicrobial Agents and Chemotherapy. 2007; 51(9):3147-54. https://doi.org/10.1128/AAC.00401-07 PMid:17620375 PMCid: PMC2043237.
25. Mutlib AE. Application of stable isotope-labeled compounds in metabolism and in metabolism-mediated toxicity studies. Chemical Research in Toxicology. 2008; 21(9):1672-89. https://doi.org/10.1021/tx800139z PMid:18702535.

26. Velthuisen EJ, Baughman TM, Johns BA, Temelkoff DP, Weatherhead JG. Synthesis and pharmacokinetic profile of highly deuterated brecanavir analogues. European Journal of Medicinal Chemistry. 2013; 63:202-12. https://doi.org/10.1016/j.ejmech.2013.02.001 PMid:23474906.
27. Savi P, Herbert JM. Clopidogrel and ticlopidine: P2Y12 adenosine diphosphate-receptor antagonists for the prevention of atherothrombosis. In Seminars in Thrombosis and Hemostasis. 2005; 31(2):174-83. https://doi.org/10.1055/s-2005-869523 PMid:15852221.
28. Savi P, Herbert JM, Pflieger AM, Dol F, Delebassee D, Combalbert J, Defreyn G, Maffrand JP. Importance of hepatic metabolism in the antiaggregating activity of the thienopyridine clopidogrel. Biochemical Pharmacology. 1992; 44(3):527-32. https://doi.org/10.1016/0006-2952(92)90445-O PMid:1510701.
29. Savi P, Combalbert J, Gaich C, Rouchon MC, Maffrand JP, Berger Y, Herbert JM. The antiaggregating activity of clopidogrel is due to a metabolic activation by the hepatic cytochrome P450-1A. Thrombosis and Haemostasis. 1994;72(08):313-7. https://doi.org/10.1055/s-0038-1648859 PMid:7831671.
30. Savi P, Pereillo JM, Uzabiaga MF, Combalbert J, Picard C, Maffrand JP, Pascal M, Herbert JM. Identification and biological activity of the active metabolite of clopidogrel. Thrombosis and Haemostasis. 2000; 84(5):891-6. https://doi.org/10.1055/s-0037-1614133 PMid:11127873.
31. Pereillo JM, Maftouh M, Andrieu A, Uzabiaga MF, Fedeli O, Savi P, Pascal M, Herbert JM, Maffrand JP, Picard C. Structure and stereochemistry of the active metabolite of clopidogrel. Drug Metabolism and Disposition. 2002; 30(11):1288-95. https://doi.org/10.1124/dmd.30.11.1288 PMid:12386137.
32. Kazui M, Nishiya Y, Ishizuka T, Hagihara K, Farid NA, Okazaki O, Ikeda T, Kurihara A. Identification of the human cytochrome P450 enzymes involved in the two oxidative steps in the bioactivation of clopidogrel to its pharmacologically active metabolite. Drug Metabolism and Disposition. 2010; 38(1):92-9. https://doi.org/10.1124/dmd.109.029132 PMid:19812348.
33. Mega JL, Close SL, Wiviott SD, Shen L, Hockett RD, Brandt JT, Walker JR, Antman EM, Macias W, Braunwald E, Sabatine MS. Cytochrome p-450 polymorphisms and response to clopidogrel. New England Journal of Medicine. 2009; 360(4):354-62. https://doi.org/10.1056/NEJMoa0809171 PMid:19106084.
34. Mega JL, Simon T, Collet JP, Anderson JL, Antman EM, Bliden K, Cannon CP, Danchin N, Giusti B, Gurbel P, Horne BD. Reduced-function CYP2C19 genotype and risk of adverse clinical outcomes among patients treated with clopidogrel predominantly for PCI: A meta-analysis. Jama. 2010; 304(16):1821-30. https://doi.org/10.1001/jama.2010.1543 PMid:20978260 PMCid: PMC3048820.
35. Xu X, Zhao X, Yang Z, Wang H, Meng X, Su C, Liu M, Fawcett JP, Yang Y, Gu J. Significant improvement of metabolic characteristics and bioactivities of clopidogrel and analogues by selective deuteration. Molecules. 2016; 21(6):704. https://doi.org/10.3390/molecules21060704 PMid:27248988 PMCid: PMC6274316.
36. Tran C, Ouk S, Clegg NJ, Chen Y, Watson PA, Arora V, Wongvipat J, Smith-Jones PM, Yoo D, Kwon A, Wasielewska T. Development of a second-generation antiandrogen for treatment of advanced prostate cancer. Science. 2009; 324(5928):787-90. https://doi.org/10.1126/science.1168175 PMid:19359544 PMCid: PMC2981508.
37. Scher HI, Fizazi K, Saad F, Taplin ME, Sternberg CN, Miller K, De Wit R, Mulders P, Chi KN, Shore ND, Armstrong AJ. Increased survival with enzalutamide in prostate cancer after chemotherapy. New England Journal of Medicine. 2012; 367(13):1187-97. https://doi.org/10.1056/NEJMoa1207506 PMid:22894553.
38. Ning YM, Pierce W, Maher VE, Karuri S, Tang SH, Chiu HJ, Palmby T, Zirkelbach JF, Marathe D, Mehrotra N, Liu Q. Enzalutamide for treatment of patients with metastatic castration-resistant prostate cancer who have previously received docetaxel: US Food and Drug Administration drug approval summary. Clinical Cancer Research. 2013; 19(22):6067-73. https://doi.org/10.1158/1078-0432.CCR-13-1763 PMid:24141628.
39. Jiang J, Pang X, Li L, Dai X, Diao X, Chen X, Zhong D, Wang Y, Chen Y. Effect of N-methyl deuteration on metabolism and pharmacokinetics of enzalutamide. Drug Design, Development and Therapy. 2016; 10:2181-91. https://doi.org/10.2147/DDDT.S111352 PMid:27462143 PMCid: PMC4939996.
40. Clegg NJ, Wongvipat J, Joseph JD, Tran C, Ouk S, Dilhas A, Chen Y, Grillot K, Bischoff ED, Cai L, Aparicio A. ARN-509: A novel antiandrogen for prostate cancer treatment. Cancer Research. 2012; 72(6):1494-503. https://doi.org/10.1158/0008-5472.CAN-11-3948 PMid:22266222 PMCid: PMC3306502.
41. Jung ME, Ouk S, Yoo D, Sawyers CL, Chen C, Tran C, Wongvipat J. Structure-activity relationship for thiohydantoin androgen receptor antagonists for Castration-Resistant Prostate Cancer (CRPC). Journal of Medicinal Chemistry. 2010; 53(7):2779-96. https://doi.org/10.1021/jm901488g PMid:20218717 PMCid: PMC3180999.
42. Tran C, Ouk S, Clegg NJ, Chen Y, Watson PA, Arora V, Wongvipat J, Smith-Jones PM, Yoo D, Kwon A, Wasielewska T. Development of a second-generation antiandrogen for treatment of advanced

prostate cancer. Science. 2009; 324(5928):787-90. https://doi.org/10.1126/science.1168175 PMid:19359544 PMCid: PMC2981508.
43. Pang X, Wang Y, Chen Y. Design, synthesis, and biological evaluation of deuterated apalutamide with improved pharmacokinetic profiles. Bioorganic and Medicinal Chemistry Letters. 2017; 27(12):2803-6. https://doi.org/10.1016/j.bmcl.2017.04.071 PMid:28478926.
44. LoRusso PM, Rudin CM, Borad MJ, Vernillet L, Darbonne WC, Mackey H, DiMartino JF, De Sauvage F, Low JA, Von Hoff DD. A first-in-human, first-in-class, phase (ph) I study of systemic Hedgehog (Hh) pathway antagonist, GDC-0449, in patients (pts) with advanced solid tumours. Journal of Clinical Oncology. 2008; 26(15_suppl):3516. https://doi.org/10.1200/jco.2008.26.15_suppl.3516
45. Geeraert P, Williams JS, Brownell I. Targeting the hedgehog pathway to treat basal cell carcinoma. Journal of Drugs in Dermatology: JDD. 2013; 12(5):519-23.
46. Robarge KD, Brunton SA, Castanedo GM, Cui Y, Dina MS, Goldsmith R, Gould SE, Guichert O, Gunzner JL, Halladay J, Jia W. GDC-0449-A potent inhibitor of the hedgehog pathway. Bioorganic and Medicinal Chemistry Letters. 2009; 19(19):5576-81. https://doi.org/10.1016/j.bmcl.2009.08.049 PMid:19716296.
47. Wong H, Chen JZ, Chou B, Halladay JS, Kenny JR, La H, Marsters Jr JC, Plise E, Rudewicz PJ, Robarge K, Shin Y. Preclinical assessment of the absorption, distribution, metabolism and excretion of GDC-0449 (2-chloro-N-(4-chloro-3-(pyridin-2-yl) phenyl)-4-(methylsulfonyl) benzamide), an orally bioavailable systemic hedgehog signalling pathway inhibitor. Xenobiotica. 2009; 39(11):850-61. https://doi.org/10.3109/00498250903180289 PMid:19845436.
48. Wong H, Theil FP, Cui Y, Marsters JC, Khojasteh SC, Vernillet L, La H, Song X, Wang H, Morinello EJ, Deng Y. Interplay of dissolution, solubility, and nonsink permeation determines the oral absorption of the hedgehog pathway inhibitor GDC-0449 in dogs: An investigation using preclinical studies and physiologically based pharmacokinetic modelling. Drug Metabolism and Disposition. 2010; 38(7):1029-38. https://doi.org/10.1124/dmd.110.032680 PMid: 20406853.
49. Wang F, Jiang H, Deng Y, Yu J, Zhan M, Zhao L, Chen Y. Design, synthesis and biological evaluation of deuterated Vismodegib for improving pharmacokinetic properties. Bioorganic and Medicinal Chemistry Letters. 2018; 28(14):2399-402. https://doi.org/10.1016/j.bmcl.2018.06.025 PMid: 29929879.
50. Kowanetz M, Ferrara N. Vascular endothelial growth factor signalling pathways: therapeutic perspective. Clinical Cancer Research. 2006; 12(17):5018-22. https://doi.org/10.1158/1078-0432.CCR-06-1520 PMid: 16951216.
51. Nakamura K, Taguchi E, Miura T, Yamamoto A, Takahashi K, Bichat F, Guilbaud N, Hasegawa K, Kubo K, Fujiwara Y, Suzuki R. KRN951, a highly potent inhibitor of vascular endothelial growth factor receptor tyrosine kinases, has antitumor activities and affects functional vascular properties. Cancer Research. 2006; 66(18):9134-42. https://doi.org/10.1158/0008-5472.CAN-05-4290 PMid:16982756.
52. Taguchi E, Nakamura K, Miura T, Shibuya M, Isoe T. Anti-tumor activity and tumour vessel normalisation by the vascular endothelial growth factor receptor tyrosine kinase inhibitor KRN951 in a rat peritoneal disseminated tumour model. Cancer Science. 2008 ; 99(3):623-30. https://doi.org/10.1111/j.1349-7006.2007.00724.x PMid: 18201272.
53. Berge EM, Bowles DW, Flaig TW, Lam ET, Jimeno A. Tivozanib: Practical implications for renal cell carcinoma and other solid tumours. Drugs of Today (Barcelona, Spain: 1998). 2013; 49(5):303-15. https://doi.org/10.1358/dot.2013.49.5.1960218 PMid: 23724410.
54. Guo S, Pang X, Peng L, Zhan M, Fan L, Gong Y, Kang F, Chen Y. Design, synthesis and biological evaluation of deuterated Tivozanib for improving pharmacokinetic properties. Bioorganic and Medicinal Chemistry Letters. 2015; 25(11):2425-8. https://doi.org/10.1016/j.bmcl.2015.03.088 PMid: 25908516.
55. Raffa RB, Friderichs EL, Reimann WO, Shank RP, Codd EE, Vaught JL. Opioid and nonopioid components independently contribute to the mechanism of action of tramadol, an 'atypical' opioid analgesic. Journal of Pharmacology and Experimental Therapeutics. 1992; 260(1):275-85. PMid: 1309873.
56. Shao L, Abolin C, Hewitt MC, Koch P, Varney M. Derivatives of tramadol for increased duration of effect. Bioorganic and Medicinal Chemistry Letters. 2006; 16(3):691-4. https://doi.org/10.1016/j.bmcl.2005.10.024 PMid:16257206.
57. Crofford LJ. COX-1 and COX-2 tissue expression: Implications and predictions. The Journal of Rheumatology. Supplement. 1997; 49:15-9.
58. Hawkey CJ. COX-2 inhibitors. The Lancet. 1999; 353(9149):307-14. https://doi.org/10.1016/S0140-6736(98)12154-2 PMid:9929039.
59. Schneider F, Hillgenberg M, Koytchev R, Alken RG. Enhanced plasma concentration by selective deuteration of rofecoxib in rats. Arzneimittelforschung. 2006; 56(04):295-300. https://doi.org/10.1055/s-0031-1296724 PMid:16724516.
60. World Health Organization. The selection and use of essential medicines: Report of the WHO expert committee, 2017 (including the 20th WHO model list of essential medicines and the 6th model list of essential medicines for children). World Health Organization. 2017.
61. Usach I, Melis V, Peris JE. Non-nucleoside reverse transcriptase inhibitors: A review on pharmacokinetics, pharmacodynamics, safety and tolerability. Journal of the International AIDS Society. 2013; 16(1):18567. https://doi.org/10.7448/IAS.16.1.18567 PMid:24008177 PMCid: PMC3764307.

62. Pollard RB, Robinson P, Dransfield K. Safety profile of nevirapine, a nonnucleoside reverse transcriptase inhibitor for the treatment of human immunodeficiency virus infection. Clinical Therapeutics. 1998; 20(6):1071-92. https://doi.org/10.1016/S0149-2918(98)80105-7 PMid:9916603.
63. Zhang C, Wang W, Zhou M, Han Y, Xie J, Qiu Z, Guo F, Li Y, Wang H, Ghanem KG, Li T. The interaction of CD4 T-cell count and nevirapine hepatotoxicity in China: A change in national treatment guidelines may be warranted. Journal of Acquired Immune Deficiency Syndromes Journal of Acquired Immune Deficiency Syndromes. 2013; 62(5):540-5. https://doi.org/10.1097/QAI.0b013e3182845cc7 PMid:23288032.
64. Sharma AM, Klarskov K, Uetrecht J. Nevirapine bioactivation and covalent binding in the skin. Chemical Research in Toxicology. 2013; 26(3):410-21. https://doi.org/10.1021/tx3004938 PMid:23387501.
65. Chen J, Mannargudi BM, Xu L, Uetrecht J. Demonstration of the metabolic pathway responsible for nevirapine-induced skin rash. Chemical Research in Toxicology. 2008; 21(9):1862-70. https://doi.org/10.1021/tx800177k PMid:18729332.
66. Sharma AM, Li Y, Novalen M, Hayes MA, Uetrecht J. Bioactivation of nevirapine to a reactive quinone methide: implications for liver injury. Chemical Research in Toxicology. 2012; 25(8):1708-19. https://doi.org/10.1021/tx300172s PMid:22793666 PMCid: PMC3475366.
67. Huebert ND, Palfreyman MG, Haegele KD. A comparison of the effects of reversible and irreversible inhibitors of aromatic L-amino acid decarboxylase on the half-life and other pharmacokinetic parameters of oral L-3, 4-dihydroxyphenylalanine. Drug Metabolism and Disposition. 1983; 11(3):195-200.
68. Olanow CW, Stern MB, Sethi K. The scientific and clinical basis for the treatment of Parkinson's disease (2009). Neurology. 2009; 72(21 Supplement 4):S1-36. https://doi.org/10.1212/WNL.0b013e3181a1d44c
69. Salat D, Tolosa E. Levodopa in the treatment of Parkinson's disease: Current status and new developments. Journal of Parkinson's disease. 2013; 3(3):255-69. https://doi.org/10.3233/JPD-130186 PMid:23948989.
70. Wiberg KB. The deuterium isotope effect. Chemical Reviews. 1955; 55(4):713-43. https://doi.org/10.1021/cr50004a004
71. Schneider F, Erisson L, Beygi H, Bradbury M, Cohen-Barak O, Grachev ID, Guzy S, Loupe PS, Levi M, McDonald M, Savola JM. Pharmacokinetics, metabolism and safety of deuterated L-DOPA (SD-1077)/carbidopa compared to L-DOPA/carbidopa following single oral dose administration in healthy subjects. British Journal of Clinical Pharmacology. 2018; 84(10):2422-32. https://doi.org/10.1111/bcp.13702 PMid:29959802 PMCid: PMC6138493.
72. Trunzer M, Faller B, Zimmerlin A. Metabolic soft spot identification and compound optimization in early discovery phases using metasite and LC-MS/MS validation. Journal of Medicinal Chemistry. 2009; 52(2):329-35. https://doi.org/10.1021/jm8008663 PMid:19108654.
73. De Voss JJ, Sibbesen O, Zhang Z, Ortiz de Montellano PR. Substrate docking algorithms and prediction of the substrate specificity of cytochrome P450cam and its L244A mutant. Journal of the American Chemical Society. 1997; 119(24):5489-98. https://doi.org/10.1021/ja970349v
74. Zhang Z, Sibbesen O, Johnson RA, De Montellano PR. The substrate specificity of cytochrome P450cam. Bioorganic and Medicinal Chemistry. 1998; 6(9):1501-8. https://doi.org/10.1016/S0968-0896(98)00091-1 PMid:9801821.
75. Gallant JE, Deresinski S. Tenofovir disoproxil fumarate. Clinical Infectious Diseases. 2003; 37(7):944-50. https://doi.org/10.1086/378068 PMid:13130407.
76. Venkatakrishnan K, Greenblatt DJ, Von Moltke LL, Schmider J, Harmatz JS, Shader RI. Five distinct human cytochromes mediate amitriptyline N-demethylation *in vitro*: Dominance of CYP 2C19 and 3A4. The Journal of Clinical Pharmacology. 1998; 38(2):112-21. https://doi.org/10.1002/j.1552-4604.1998.tb04399.x PMid:9549641.
77. Ulrich S, Läuter J. Comprehensive survey of the relationship between serum concentration and therapeutic effect of amitriptyline in depression. Clinical Pharmacokinetics. 2002; 41:853-76. https://doi.org/10.2165/00003088-200241110-00004 PMid:12190332.