KalVista Pharmaceuticals

The discovery of sebetralstat, an oral, small molecule plasma kallikrein inhibitor to treat attacks of hereditary angioedema

RSC/SCI 22nd Med Chem Symposium 2023

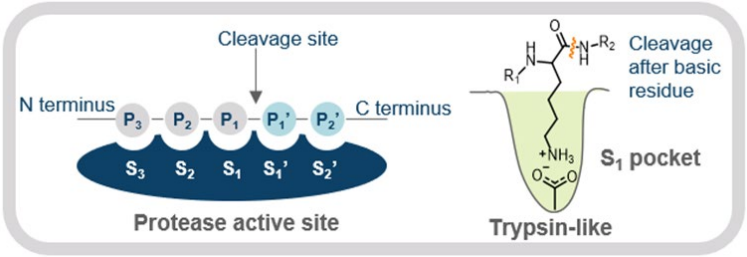
Presented by Rebecca Davie

Conflict of Interest

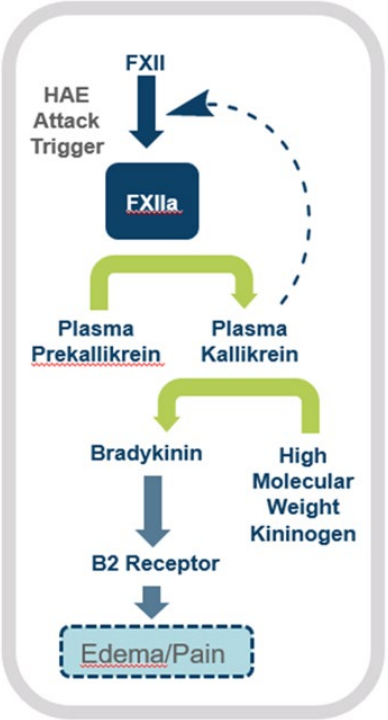
- Employee of KalVista Pharmaceuticals

Plasma Kallikrein and the Kallikrein - Kinin System

- Plasma kallikrein (PKa) is a trypsin-like serine protease



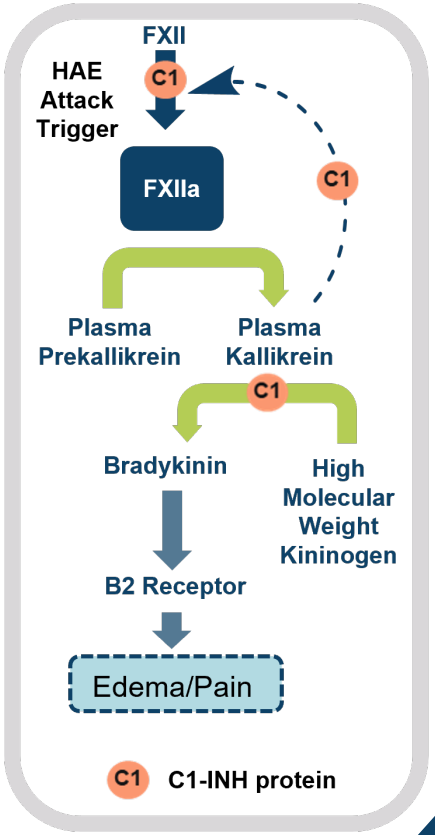
- PKa is derived from its zymogen plasma prekallikrein which circulates in the blood
- Plasma prekallikrein is activated by FXIIa during contact system activation
- PKa cleaves high molecular weight kininogen to generate bradykinin; a potent mediator of vascular permeability and inflammation



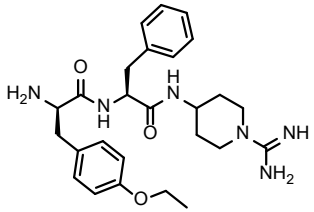
HAE, hereditary angioedema;

Hereditary Angioedema (HAE)

- HAE is a rare genetic disease causing painful and potentially life-threatening swelling in various parts of the body
- It is associated with uncontrolled PKa activity and generation of bradykinin
- C1-INH protein is the primary physiological inhibitor of PKa
- Reduced expression or function of C1-INH in HAE facilitate unrestricted PKa activity that drives attacks of angioedema

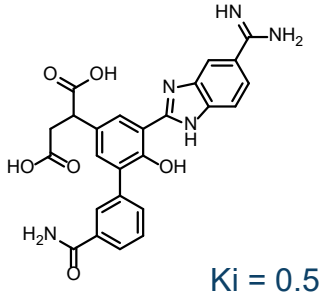


Reported Inhibitors at Project Initiation



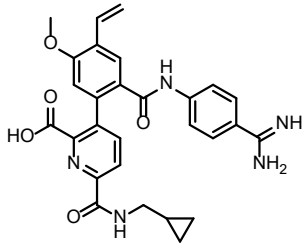
KV999272 IC_{50} = 3.6 nM

KalVista WO2003076458
Clermont, A et al. Investigative Opthalmology & Visual Science. 2016;57(6):2390-2399.



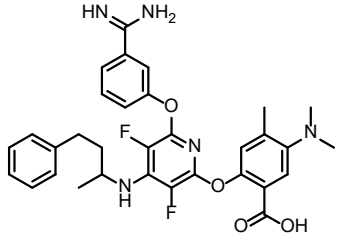
Ki = 0.5 nM

Young WB, et al. *Bioorg Med Chem Lett.* 2006;16(7):2034-2036.



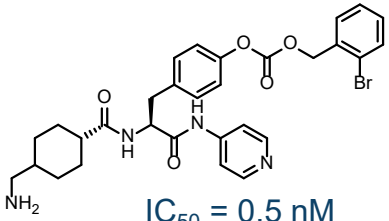
Ki (initial) = 1.3 nM
Ki (final) = 0.26 nM

Zhang J, et al. *Med Chem.* 2006;2(6):545-553.



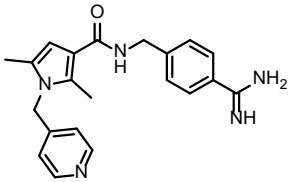
Ki = 9 nM

Kolte D, et al. *Br J Pharmacol.* 2011;162(7):1639-1649.



IC_{50} = 0.5 nM

Okada Y, et al. *Chem. Pharm. Bull (Tokyo). 2000;48(2),184-193.*

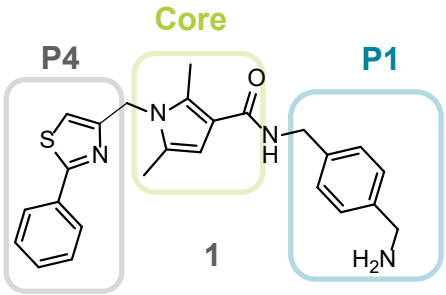


ASP-465, Ki = 30 nM

ActiveSite WO2008016883

All examples contained functionality challenging for oral drug delivery

Oral PKa Hit Identified



IC_{50} 72 nM (LE = 0.32)

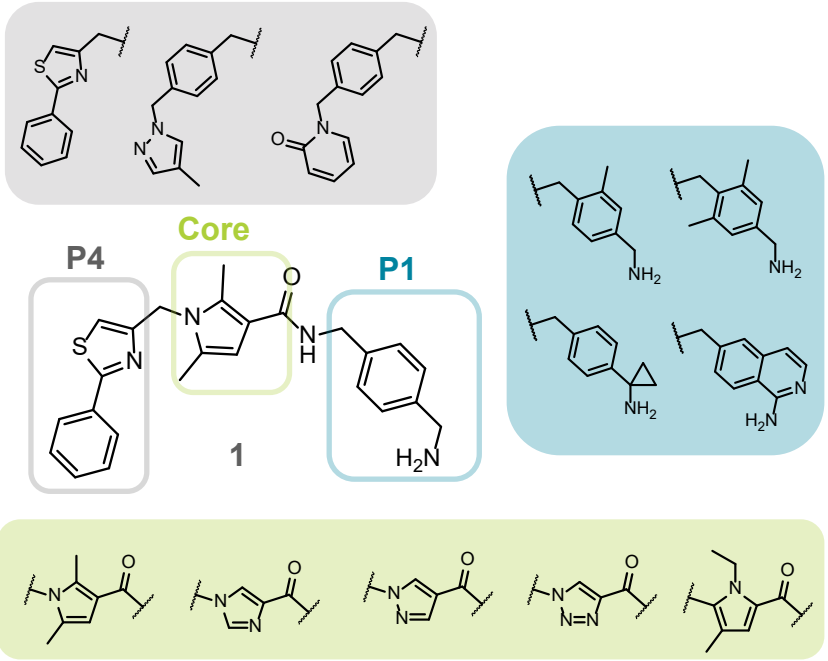
cLogP *ACD/percepta*	**3.7**
cLogD$_{7.4}$ *ACD/percepta*	**2.1**

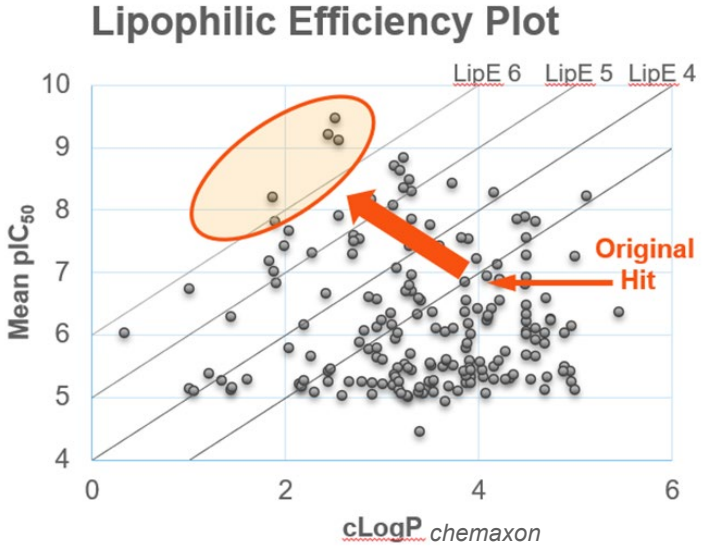
- **Potency without a benzamidine P1 group**
- **Selective**
  - >10μM for plasmin, thrombin, trypsin and KLK1
- **Oral exposure**
  - F = 52%
  - High CL
- **Good starting point**
  - Ligand efficient
  - Robust chemistry

Aim: to further reduce the basic nature of P1 and maintain potency

KLK1 = tissue kallikrein 1

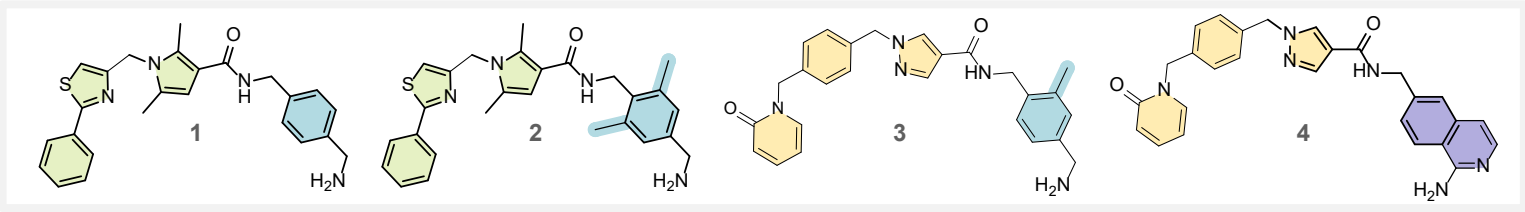
Hit-to-Lead





Motifs selected for potency, modulation of basicity and clearance ➡ matrix array

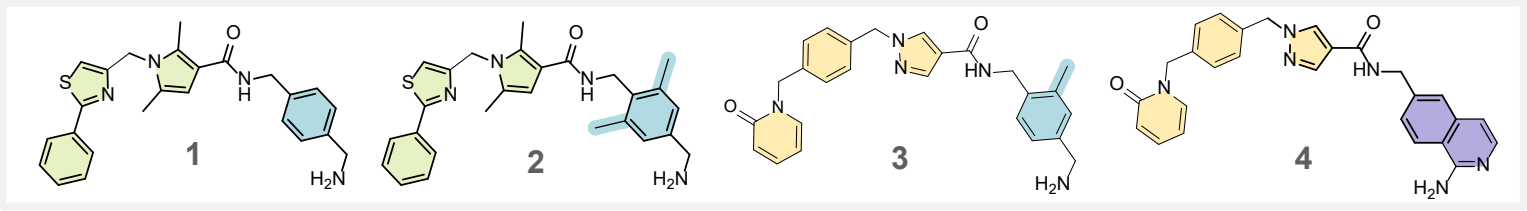
Matrix Highlights



	1	2	3	4
Predicted pK_a *ACD/percepta*	**9.1**	**9.2**	**9.1**	**7.5**
cLogP / $cLogD_{7.4}$ *ACD/percepta*	**3.7 / 2.1**	**4.8 / 3.1**	**1.6 / -0.1**	**2.0 / 1.7**
PKa IC_{50} (nM)	**72**	**7**	**0.7**	**0.6**

- 100-fold improvement in potency with pyrazole core in combination with benzyl-pyridone
- Reduced basicity with aminoisoquinoline **4** - matched the potency of benzylamine analogue **3**

Matrix Highlights

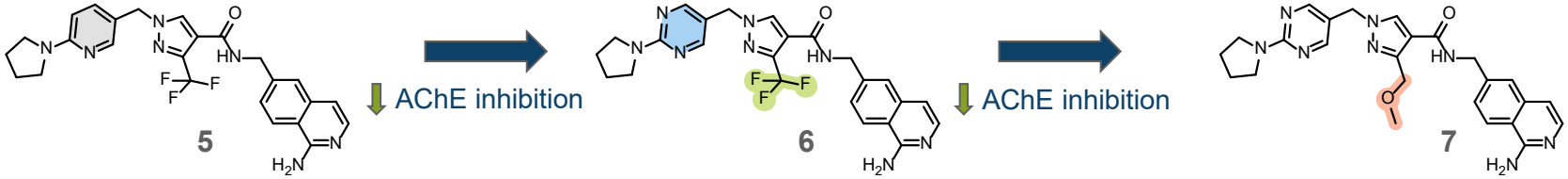


		1	2	3	4
PKa IC_{50} (nM)		**72**	**7**	**0.7**	**0.6**
Predicted CL RLM / RHEP (mL/min/kg)		**2 / *nd***	**<4 / <3**	**2 / <14**	**25 / 10**
Rat PK	**CL** (mL/min/kg)	**107**	**28**	**223**	**21**
	V_{ss} (L/kg)	**19.5**	**9.5**	**2.2**	**0.9**
	F %	**52**	**57**	**0**	**0**

- Extra hepatic CL component for benzylamines
- New lead compound 4 ➡ away from highly basic lipophilic compounds

nd, not determined

Aminoisoquinoline P1 Series Highlights

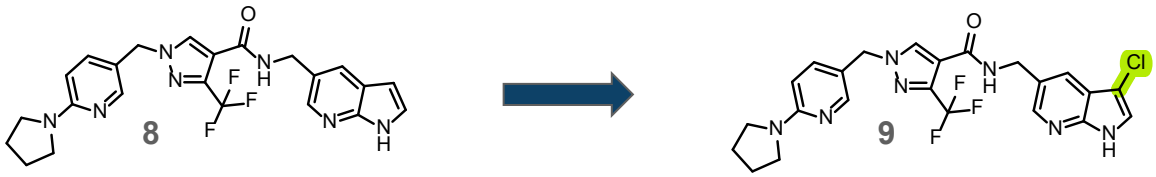


		5	6	7
cLogP / cLogD$_{7.4}$ *ACD/percepta*		**3.1 / 2.7**	**2.7 / 2.3**	**2.0 / 1.6**
PKa IC$_{50}$ (nM)		**8**	**7**	**9**
AChE IC$_{50}$ (nM)		**24**	**160**	**671**
Rat PK	**CL** (mL/min/kg)	**20**	**12**	**27**
	V$_{ss}$ (L/kg)	**5.6**	**1.3**	**2.8**
	F (%)	**31**	**34**	**35**

- F 20% in dog and monkey
- >300-fold selectivity over plasmin, thrombin, trypsin, KLK1
- hERG IC$_{50}$ 13µM
- Ames negative
- Kidney pathology in monkey toxicology study

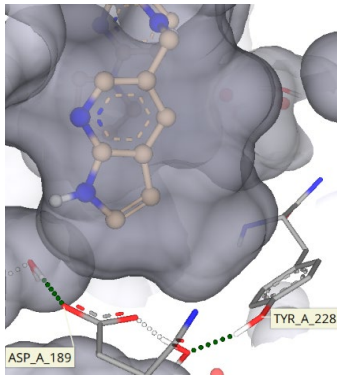
KLK1 = tissue kallikrein 1. AChE = acetylcholinesterase.

Modification to P1 – Chloroazaindole Discovery

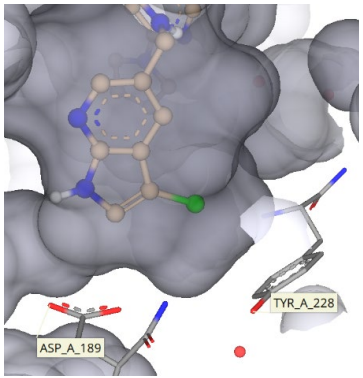


	8	9
PKa IC_{50} (nM)	360	12
AChE IC_{50} (nM)	2317	1970

Crystal structures bound into the active site of human PKa. View of the S1 pocket



- No Asp189 interaction
- Cl addition designed to form an interaction with Tyr228
- Cl sits 4-5Å above Tyr 228 in the S1 pocket

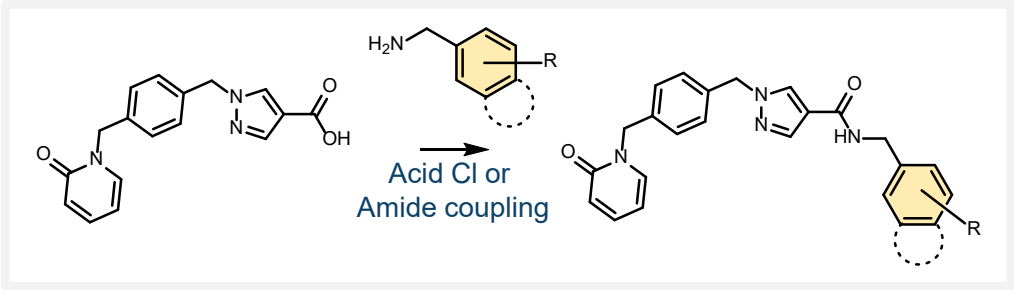


PKa potency demonstrated with a non-ionised P1

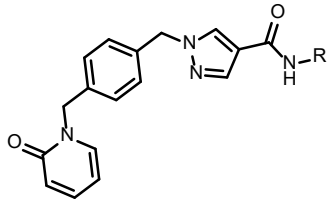
Project Turning Point

- Interaction with Asp189 in S1 pocket not required for potency
- Confidence for a bolder scope of S1 pocket binders
- Library of primary amines using ZINC fragment-like database
  - Filtered on physical chemical properties, shape-based overlays of known P1s and molecular docking
- Target
  - Neutral (non-ionised) groups
  - Variety of ring substituents
  - ~90% focus monoaryl rings

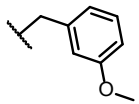
Library with potent scaffold

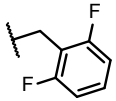


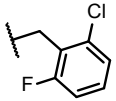
Neutral P1 Library

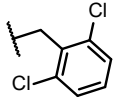


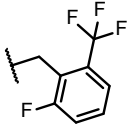
R =



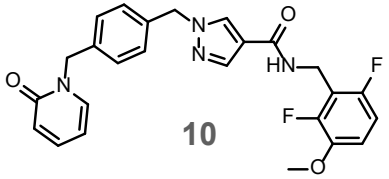








PKa IC_{50} = 90nM | 62nM | 31nM | 40nM | 8nM



10

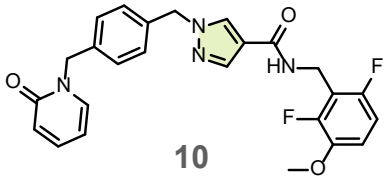
Key Findings

- *meta*-Methoxy group identified as a mono-substituted analogue
- Di-*ortho* substitution favoured for potency
- SAR additive

	10
cLogP *ACD/percepta*	**2.4**
IC_{50} (nM)	**3.0**
3-day Caco2 A-B P_{app} (nm/s)	**32**
HLM / RLM CL_{int} (µL/min/mg of protein)	**36 / 63**

- Novel potent substituted phenyl P1 identified
- Permeability and CL_{int} tuning required

Lead Optimisation



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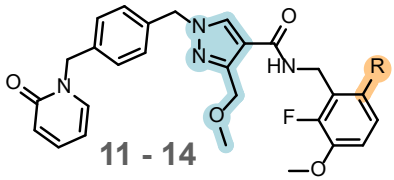


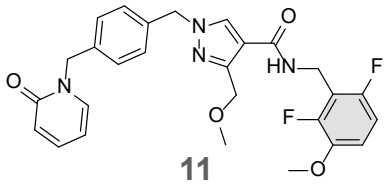
11 - 14

		10	11	12	13	14
	R =	-	F	H	Cl	CN
PKa IC_{50} (nM)		3.0	6.0	17	5.5	1.0
3-day Caco2 A-B P_{app} (nm/s)		32	180	250	93	140
Kinetic Solubility (mg/mL)	0.1N HCl	*nd*	>0.12	>0.12	>0.12	>0.12
	pH 7.4 buffer	*nd*	>0.12	>0.12	>0.12	>0.12
Thermodynamic Solubility* (mg/mL)	FaSSGF	*nd*	0.009#	*nd*	*nd*	0.021
	FaSSIF	*nd*	0.009#	*nd*	*nd*	0.031

- Substitution on the pyrazole core essential for permeability
- Thermodynamic solubility gave a more informed picture

nd, not determined
**Performed with amorphous material at a nominal 1mg/ml, #Crystalline material*

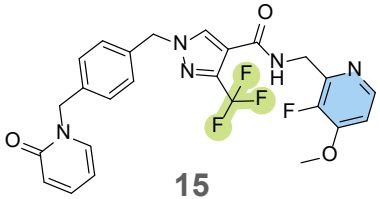
Lead Optimisation



11

↓ Lipophilicity
CL

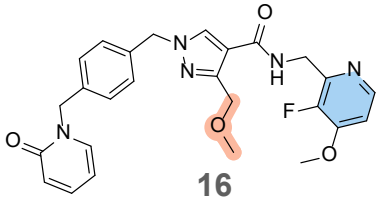
↑ Solubility



15

↓ Lipophilicity
CL

↑ Solubility



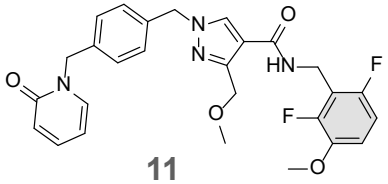
16

		11	15	16
cLogP *ACD/percepta*		2.9	2.2	1.8
PKa IC_{50} (nM)		6.0	3.2	6.0
3-day Caco2 A-B P_{app} (nm/s)		180	170	90
HLM / RLM CL_{int} (µL/min/mg of protein)		92 / 101	49 / 35	14 / 23
Thermodynamic Solubility* (mg/mL)	**FaSSGF**	0.009#	0.69	>1.0
	FaSSIF	0.009#	0.022	0.22

Tuned ADME properties whilst maintaining potency and a level of permeability

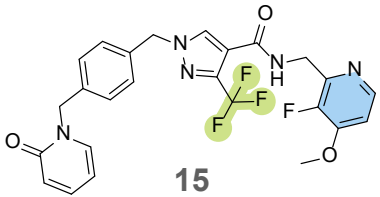
*Performed with amorphous material at a nominal 1mg/ml, #Crystalline material

Lead Optimisation



11

Solubility



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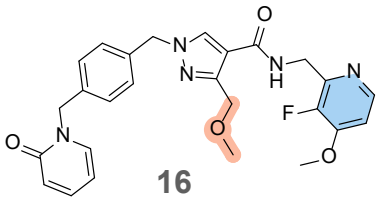
Solubility



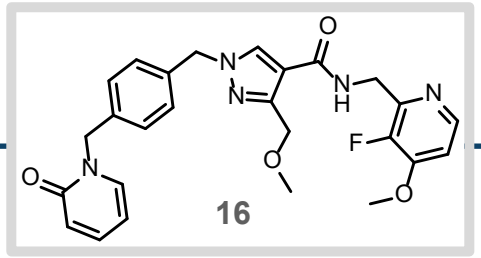
16

		11	15	16
PKa IC_{50} (nM)		**6.0**	**3.2**	**6.0**
Predicted CL RLM / RHEP (mL/min/kg)		**2.6 / 19**	**5.1 / 19**	**1.8 / 9.8**
Rat PK	**CL** (mL/min/kg)	**13**	**7**	**12**
	V_{ss} (L/kg)	**0.4**	**0.3**	**0.5**
	$t_{1/2}$ (min)	**51**	**66**	**62**
	C_{max} (ng/mL) at 10mg/kg	**2892**	**2487**	**2803**
	F %	**64**	**97**	**82**

- Compounds distinguished in second species PKs
- Monkey PK for 16 maintained a C_{max} of 2242 ng/mL and 48% F

Results supported continued progression

Sebetralstat (KVD900)



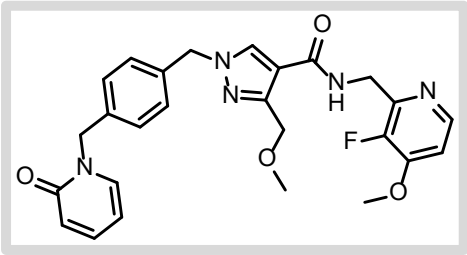
- Potent, competitive and reversible inhibitor of PKa
- Selective across a panel of proteases
- Clean off-target safety panel, IC_{50} >10 µM
- hERG IC_{50} >33 µM
- Ames negative
- CYP_{450} inhibition (human liver mics)
  - >25µM across the 7 main isoforms

Nominated as the pre-clinical lead, KVD900

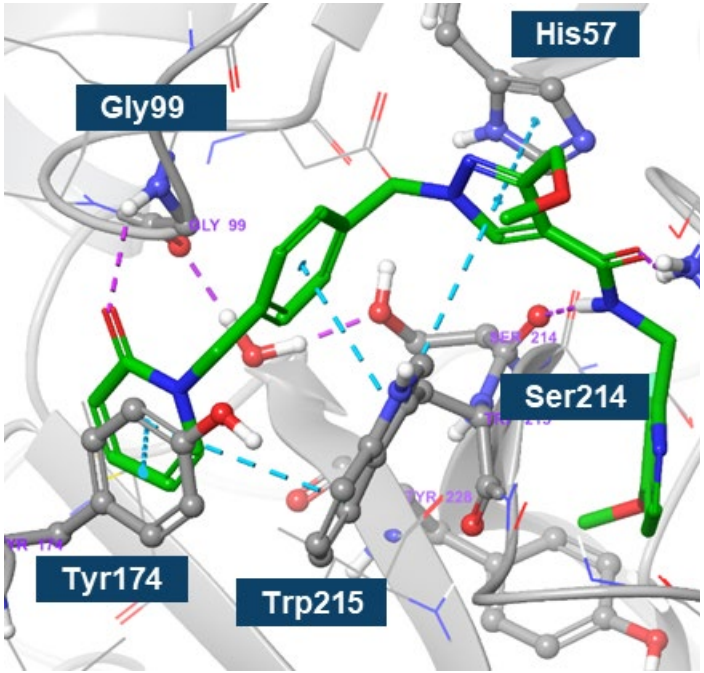
Enzyme	IC_{50}
Plasma Kallikrein	**6.0 nM**
Tissue Kallikrein 1 (KLK1)	>40 µM
FXIIa	>40 µM
FXIa	>40 µM
FXa	>10 µM
FVIIa	>10 µM
Plasmin	>40 µM
Thrombin	>40 µM
Trypsin	>40 µM
Beta-secretase 1	>10 µM
Cathepsin D	>10 µM
Cathepsin G	>10 µM
Renin	>10 µM
Tissue Plasminogen Activator	>10 µM
Tryptase	>10 µM

Davie, RL, et al. Sebetralstat (KVD900): A Potent and Selective Small Molecule Plasma Kallikrein Inhibitor Featuring a Novel P1 Group as a Potential Oral On-Demand Treatment for Hereditary Angioedema *J. Med. Chem., 2022, 65, 13629-13644.*

Sebetralstat – Binding in PKa Active Site



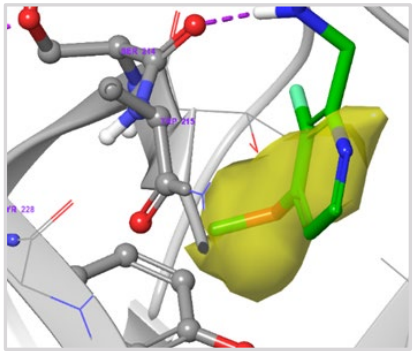
- Characteristic U-shaped conformation stabilised by the ‘Trp flip’ (Trp215)[1]
- The network of π-stack interactions and H-bonding compensates for the loss of the Asp189 salt-bridge in moving to neutral P1s
- Pyrazole core substituent solvent exposed



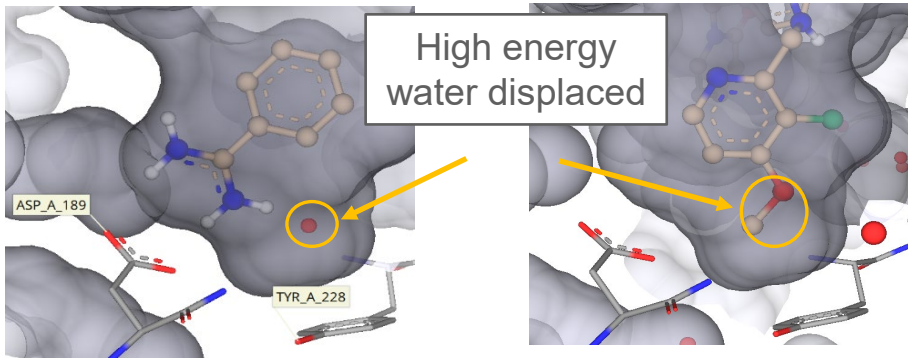
X-ray structure of sebetralstat bound into PKa
PDB Code: 8A3Q (resolution of 2.06Å)

1. Partridge JR, et al. *J Struct Biol.* 2019;*206(2):*170-182.

Sebetralstat – Binding in PKa S1 Pocket



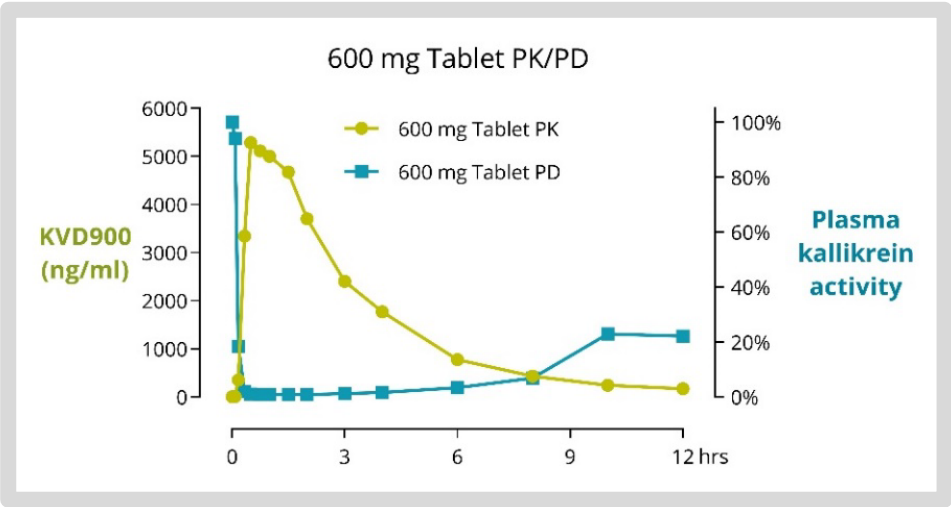
- No specific polar interactions seen for P1 group
- Aryl ring and methoxy substituent nicely occupy a lipophilic area in S1 pocket
- Pyridine N sits outside of the hydrophobic space
- High energy water situated above the face of Tyr228
- Methoxy group displaces the high energy water



Benzamidine (left; PDB: 2ANW) and sebetralstat (right; PDB: 8A3Q) bound in PKa

Sebetralstat Phase 1 - Rapid and Complete Inhibition of PKa

- 68 healthy volunteers received active treatment in Phase 1 single ascending dose trial
- Rapid onset of inhibition determined using PKa assay in whole plasma
- No SAEs reported and no patients withdrew due to adverse events



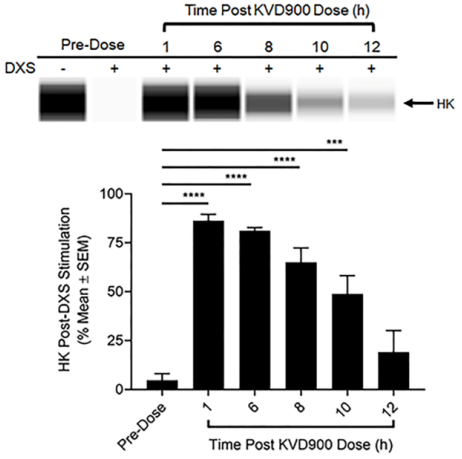
- The PK profile matched the desired on-demand profile
- Rapid absorption and near complete PKa inhibition

Maetzel A, et al. KVD900, an oral on-demand treatment for hereditary angioedema: Phase 1 study results. *J Allergy Clin Immunol* 2022, 149, 2034-2042.

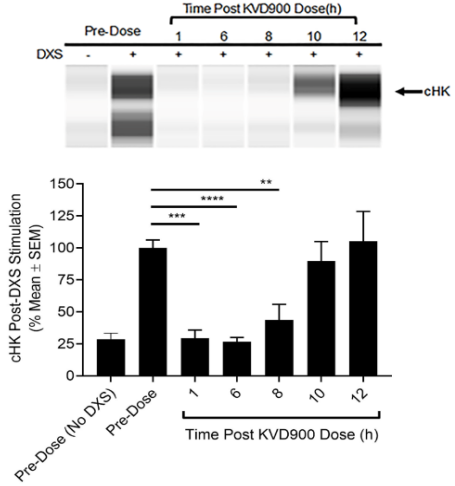
Effect of Orally Administered Sebetralstat on Kallikrein-Kinin System

KalVista Pharmaceuticals

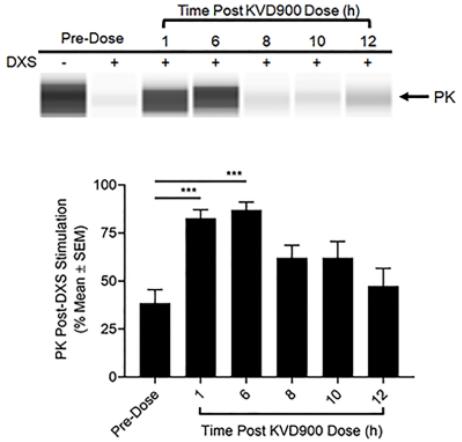
High Molecular Weight Kininogen (HK)

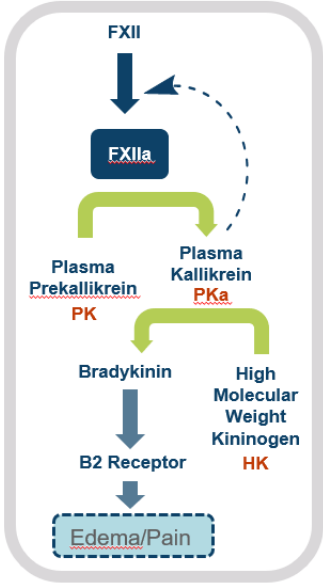


Cleaved HK (cHK)



Plasma Prekallikrein (PK)



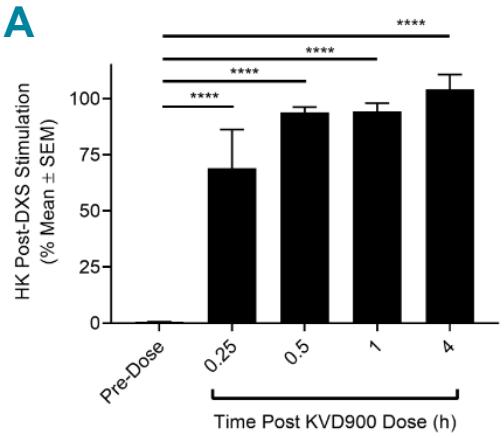


Orally dosed sebetralstat inhibits dextran sulfate-stimulated kallikrein-kinin system *ex vivo* activation and HK cleavage in plasma from 1 to 8 hours post dose

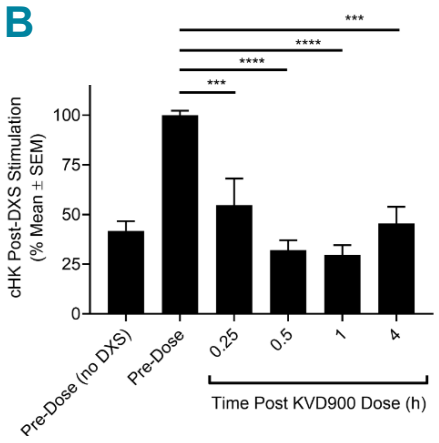
Duckworth EJ, *et al.* Pharmacological suppression of the kallikrein kinin system with KVD900: An orally available plasma kallikrein inhibitor for the on-demand treatment of hereditary angioedema *Clin Exp Allergy 2022, 52, 1059-1070.*

Effect of Sebetralstat on DXS-stimulated PKa activity in HAE Plasma

High Molecular Weight Kininogen (HK)



Cleaved HK (cHK)



A

- DXS stimulation of predose plasma resulted in complete consumption of HK
- Strong protection of HK between 15 min and 4h

B

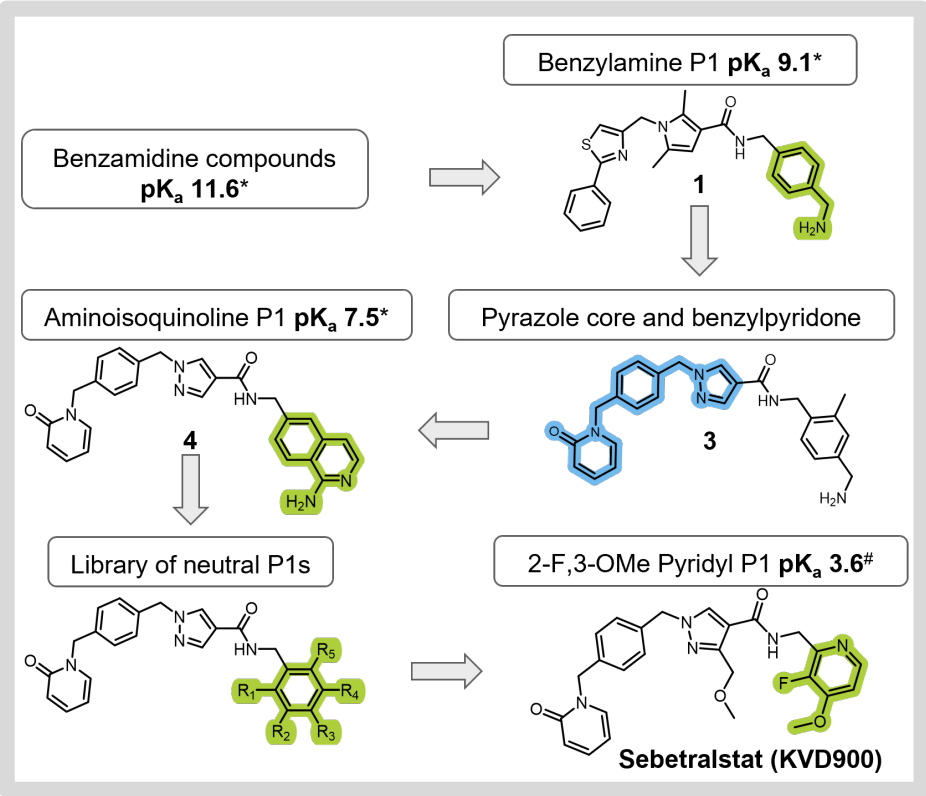
- Cleaved HK (cHK) increases following DXS stimulation of predose plasma
- Sebetralstat blocked the generation of cHK from 15 min to 4 hours post dose

Sebetralstat provides rapid protection of HK, and blocks the generation of cHK

Duckworth EJ, *et al.* Pharmacological suppression of the kallikrein kinin system with KVD900: An orally available plasma kallikrein inhibitor for the on-demand treatment of hereditary angioedema *Clin Exp Allergy 2022, 52, 1059-1070.*

Evolution of PKa Inhibitors

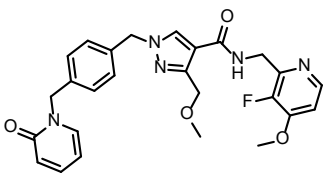
- Structure and property-based approach delivered potent, reversible competitive PKa inhibitors
- Conformational change (“Trp-flip”) in the protein and the neutral P1 group contributed to exceptionally selective PKa inhibitors
- Identified a novel, non-basic P1 group in sebetralstat with suitable PK for oral acute delivery



*pK_a values calculated using ACDlabs percepta software; #KVD900 measured pK_a from PION using a UV-metric method over 1.5-12.5 pH range

Sebetralstat Summary

- Positive Phase 2 clinical trial with sebetralstat
  - Statistically and clinically significant responses as an oral on-demand treatment for HAE attacks
- Enables early intervention and maximises treatment success
- Significantly improved patient reported outcomes of treatment effect and attack severity
- Generally safe and well tolerated
- Phase 3 clinical trial underway (KONFIDENT, NCT05259917)



Sebetralstat

More information on the KONFIDENT trial can be found at: **www.konfidentstudy.com**

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