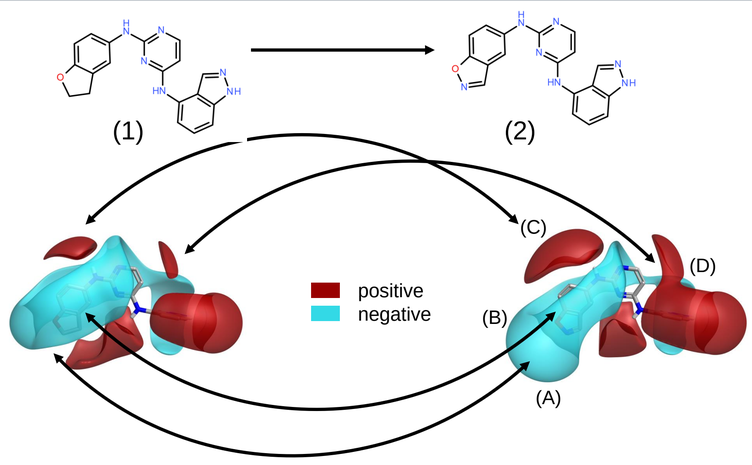
Deciphering Kinase SAR using Electrostatics

Martin J. Slater, Tim Cheeseright, Giovanna Tedesco, Sylvie Sciammetta, Mark D. Mackey
Cresset, Cambridgeshire, UK martin@cresset-group.com cresset-group.com

Electrostatics to drive design

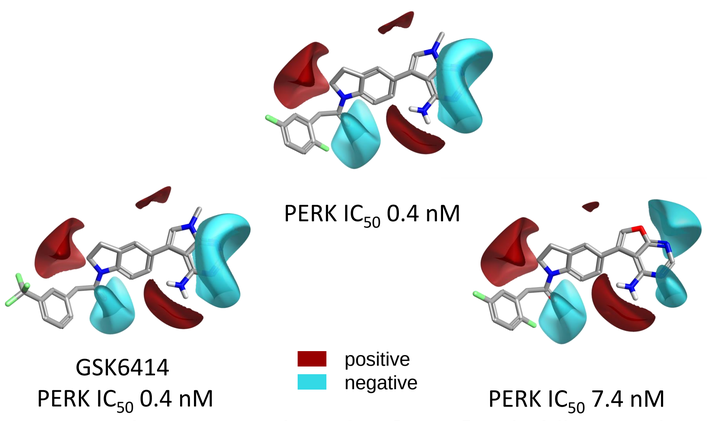
When designing a new molecule, interactive visual feedback on changes in electrostatic interactions is highly beneficial. However, changes are rarely simple. A change on one side of the molecule can often influence a distal region, especially if the systems are electronically linked through *π*-systems. Let's consider the transformation of a dihydrobenzofuran (1) into a benzoisoxazole (2).

This simple change has multiple effects due to the increased electron-withdrawing character of the new heteroatom and the addition of an aromatic ring: (A) the boundary of negative electrostatic potential extends further; (B) the shape and size of the negative *π*-cloud is significantly altered; (C) the size and extents of the positively charged aromatic edge are increased; and (D) there is a small increase in the positive potential associated with the aromatic hydrogens of the pyrimidine nucleus at the other end of the molecule.



Electrostatics to drive activity

In a study of PERK inhibitors,[1] Axten *et al*. show that changes in electrostatics of the hinge-binding heteroaryl group can be directly related to activity.



Heteroaryl groups such as furo[2,3-*d*]pyrimidin-4-amine (right), which are associated to a less negative electrostatic field with respect to pyrrolo[2,3-*d*]pyrimidin-4-amine (left and center), are also less potent on PERK.

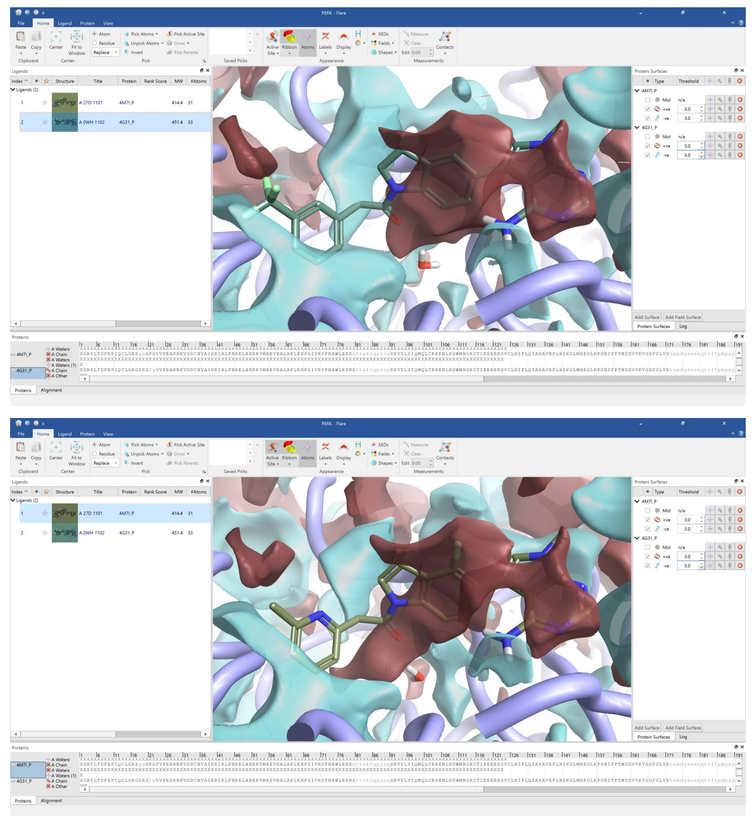
Complement to protein

A similar approach to that used for ligand electrostatics[2,3] was used to generate the PERK protein electrostatic environment in Flare,[4] starting from the PDB entry 4G31 and including the water molecule bridging the interaction between the carboxamide carbonyl and Val952, Val651 (top right).

The protein interaction potentials for PERK nicely complement those of GSK6414. The electron-rich pyrrolo[2,3-*d*]pyrimidine heteroaryl system and the carboxamide carbonyl sit in the middle of an area of positive potential in the PERK active site. At the same time, the areas of positive electrostatic potential from the ligand corresponding to the 4-amino group and the CH_2 bridge sit in an area of negative potential in the protein.

The protein interaction potential of the PERK active site also provides the most likely rationale for the lower potency of the furo[2,3-*d*]pyrimidin-4-amine analogue, as the lower electron density on the aromatic nucleus reduces the complementarity of the ligand with the PERK active site.

Interestingly, the main features of the electrostatic potential in the PERK active site are largely conserved across different structures, as can be appreciated comparing the structures of 4G31 (top) and 4M7I (bottom). Both protein structures were prepared with Build Model, which is Flare's built-in protein preparation tool, using a similar atom selection.



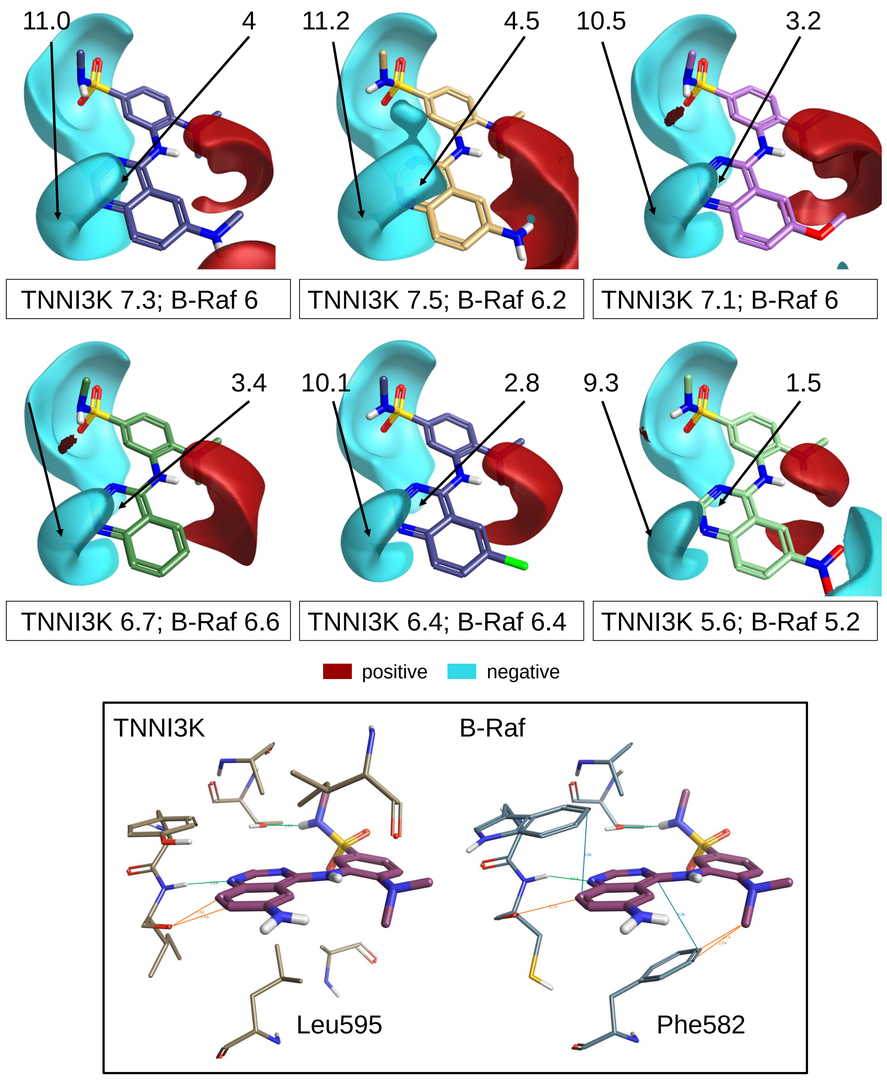
Electrostatics to drive selectivity

Modulation of electrostatic properties can be used to drive selectivity as well as activity. In an excellent study of TNNI3K inhibitors, Lawhorn *et al.*[5] showed how the H-bonding acceptor strength of a range of substituted quinazolines could be directly correlated to activity through the Hammett s_P of the 6-substituent. At the same time, the effect on the activity against the related B-Raf kinase was analyzed.

In B-Raf, the quinazoline ring engages in *π*-stacking interactions with a Phe residue which is absent in TNNI3K. While electron-donating 6-substituents enhance the H-bond acceptor character on both kinases, they are detrimental to B-Raf-specific *π*-stacking interactions, thus increasing selectivity for TNNI3K along with potency.

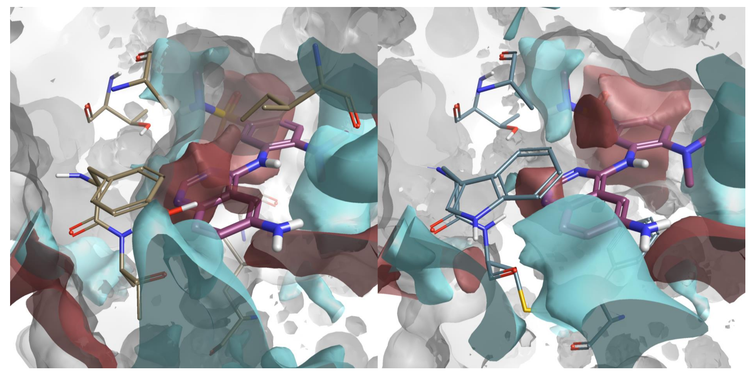
Top right: Six TNNI3K inhibitors taken from Lawhorn *et al*. showing negative electrostatic potential intensity at the edge (responsible for H-bond acceptor properties) and face (influencing B-Raf-specific *π*-stacking interactions) of the quinazoline system, along with activity data against TNNI3K and B-Raf.

Bottom right: Comparison between active sites of TNNI3K (PDB 4YFF) and B-Raf (PDB 4YHT). All compounds were modeled by in-place editing of the co-crystallized ligand from 4YFF.



Complement to protein

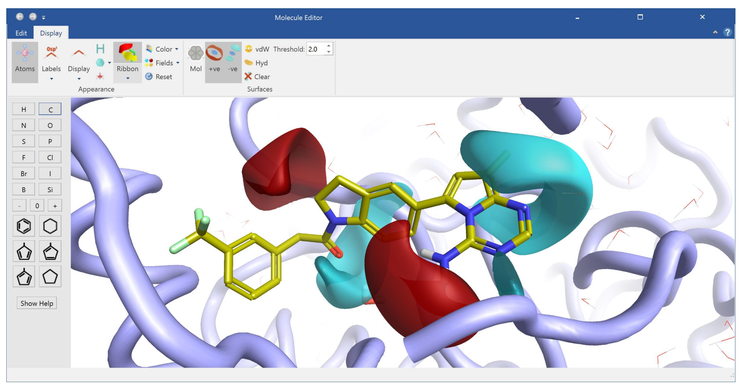
The protein interaction potentials for TNNI3K and B-Raf nicely illustrate the selectivity trend. In TNNI3K, the negative electrostatic region at the edge and face of quinazoline sits nicely in a region of positive electrostatics. Accordingly, electron-donating substituents which increase the electron density on the quinazoline ring favor TNNI3K activity.



On the contrary, in B-Raf the quinazoline sits in a region which is mainly negative, due to the aromatic residues lining its active site. This makes the effect of electron-donating substituents detrimental for B-Raf activity, and favorable for TNNI3K *vs* B-Raf selectivity.

Electrostatics on the fly

The Molecule Editor in Flare enables interactive editing of a ligand in the context of a protein, immediately visualizing the impact on the ligand fields. The screenshot shows the effect of changing the hinge-binding group to a pyrrolotriazine.



Conclusion

Protein interaction potentials and ligand fields are a powerful way of understanding the electrostatics of ligand-protein interactions. The knowledge gained is invaluable for informing ligand design to optimize activity and selectivity.

References

[1] *J. Med. Chem.* **2012**, *55*, 7193-7207
[2] *J. Comp. Aided. Mol. Des*. **1994**, *8*, 653-668
[3] *J. Chem. Inf. Model.* **2006**, *46*, 665-676
[4] http://www.cresset-group.com/flare
[5] *J. Med. Chem.* **2015**, *58*, 7431-7448