BioSolveIT
expect actives!

Activity Spotter
Extract Molecular Patterns

The Activity Spotter derives 3D features from aligned active and inactive molecules to capture a compound series' biological activity, providing SAR insights and pharmacophore constraints for screening.

How Does Activity Spotter Work?

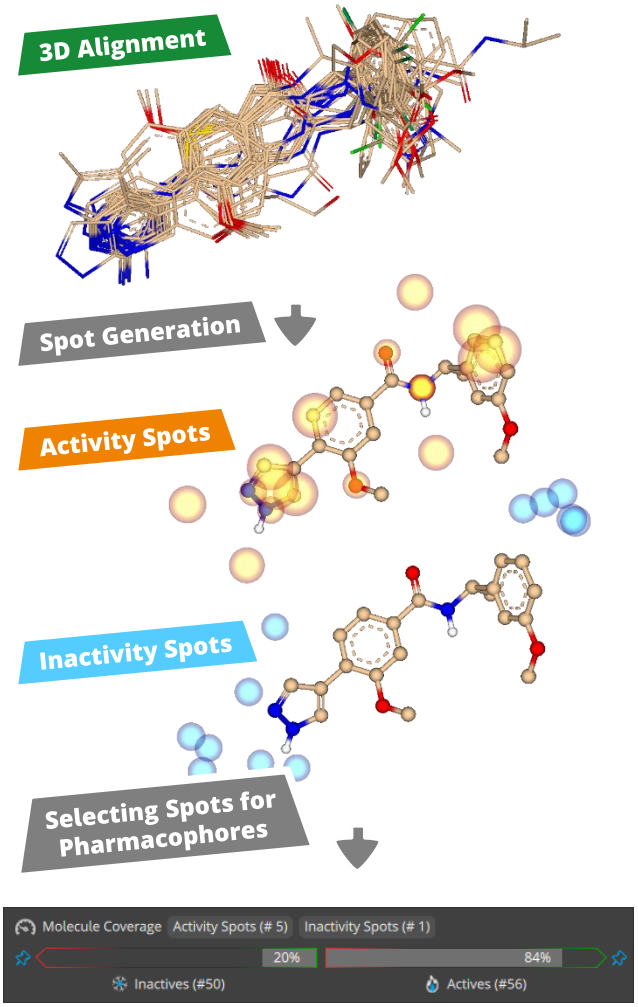
The main functionality of the Activity Spotter is to capture key features of active molecules in a 3D context. In the Activity Spotter Mode, molecules are brought into a common 3D frame (via ligand alignment, docking, or pocket superpositioning) while being classified as Active or Inactive. From recurring interaction patterns across the set, it derives localized 3D clusters ("Spots") that highlight features linked to activity (Activity Spots) or to inactivity (Inactivity Spots). The binary nature of the approach is not limited to activity; it can also be extended to metabolic behavior, toxicity effects, or selectivity profiles, as well as other drug discovery challenges that can be reduced to "preferred" versus "non-preferred" outcomes.
The generated Spots can be used as inclusive or exclusive pharmacophores for subsequent virtual screening campaigns to identify novel chemotypes with key features, or as an SAR foundation to further optimize existing scaffolds.

Setup and Workflow

- **Alignment is foundational:** ligands must be in a consistent 3D frame via docking, pocket alignment, or ligand-based alignment (e.g. FlexS/SeeSAR's Similarity Scanner Mode).
- **Use rigid reference compounds whenever possible:** rigid ligands provide more reliable geometry and cleaner Spot patterns.

Spot Generation

Spots are generated by clustering feature points in 3D space, derived from the ligands' heavy atoms and, potential interaction-point representations for H-bonds. By grouping pharmacophore or interaction points that recur across multiple compounds, the method defines consensus spatial regions, so-called "Spots", that capture common interaction patterns in a shared 3D context.



Covering Activity with Pharmacophores

Use the generated spots to build pharmacophore combinations that capture many actives while excluding inactives. Spots can be set to "include" or "exclude" (inactivity spots are always exclusion) and used as filters in structure- or ligand-based virtual screening to enrich promising candidates. The resulting SAR can also guide further decoration and scaffold optimization.

Additional Considerations

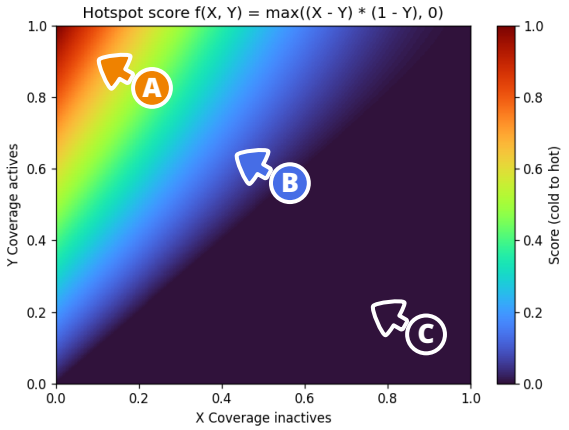
- Decorate compounds: explore substitutions around a reference scaffold to see which chemical modifications expand coverage of active-favoring spots while avoiding inactive-associated regions.
- Not limited to activity: the same approach can be applied to other endpoints (selectivity, toxicity, covalent reactivity, ...).
- During the Spot generation, each compound is treated "as is" in the provided form; the algorithm does not enumerate or normalize tautomers before clustering.

Separation Power of Activity Spots

A particular strength of the Activity Spotter is that it can crystallize the most important features that distinguish actives from inactives. Activity Spots are therefore assigned ranks indicating which spot best describes the actives. For example, a Spot that occurs only in active molecules appears at rank 1, whereas one that also shows up in some inactives is displayed further down the ranking.

Maximizing Active Coverage

- The algorithm computes an internal score that reflects how well a spot pattern covers actives while avoiding inactives with the goal to maximize active coverage and minimize inactive coverage.
- As soon as inactives begin to be covered, the score is penalized. This weighting prefers separation that favors actives, i.e., it prioritizes pattern that keep inactives out even that means not capturing every possible active.



Weight principle: **(A)** Activity spots are those that cover many actives with few/no inactives. **(B)** Activity Spots featuring more inactives rank lower. **(C)** Inactivity Spots are regions that never contain an active. These are strong indicators of inactivity and can be treated as avoidance zones during design.

Spot Types and Feature Tightness

Different features are clustered with different radii, reflecting how "tight" or "relaxed" their spatial consensus should be. The clustering size is also reflected in the size of the displayed Activity Spots.

Spot Type	Clustering Radius (Å)	Considered Clustering Ranks
Acceptor Atom	1.2	2
Donor Atom	1.2	2
Acceptor Atom Interaction	1.6	2
Donor Atom Interaction	1.6	2
Nonpolar Atom	1.2	2
Aromatic Ring	2.0	2
Nonpolar Exposed Atom	2.0	5

- Donors/acceptors are tight (small radius), because their directionality and placement are highly specific.
- Interaction-point variants are more relaxed (medium radius), allowing tolerance in where the interaction is realized.
- Lipophilic / aromatic-like features are broader (larger radius), reflecting fuzzier spatial requirements.
- Nonpolar Exposed Atom uses more ranks (5): this suggests added flexibility/robustness in how exposed nonpolar patterns are detected across varied chemotypes.

Definition: Nonpolar Exposed Atom

- The Spot type "Nonpolar Exposed Atom" must be a heavy atom, not polar (C, halogen), and has only 1 direct heavy-atom neighbor (i.e., terminal or near terminal heavy atom).
- Subsequently, it is tested whether it is "exposed" by proximity to other atoms in the same molecule: The other atoms must be at least 2 bonds removed, not a hydrogen and not in a ring.
- If there are 2 of these atoms within a certain distance of the atom, it is not exposed. (Distance is VdW radius atom + VdW radius other atom -0.3)

Additional Notes

- Inactivity spots always display the same size.