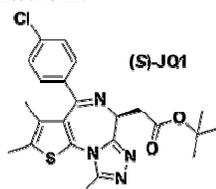




Compound JQ1 user guide:

I. Structural information:



Compound name: (S)-JQ1, Bromodomain inhibitor (for BRD2, 3, 4, and T)

Chemical name: tert-butyl 2-((6S)-4-(4-chlorophenyl)-2,3,9-trimethyl-6H-thieno[3,2-f][1,2,4]triazolo[4,3-a][1,4]diazepin-6-yl)acetate

Chemical Formula: C₂₃HClN₄O₂S

CAS #: 1268524-70-4

Exact Mass: 456.1387

Molecular Weight: 456.9882

SMILES: C1C=CC=C(C2=N[C@@H](CC(OC(C)(C)C)=O)C3=NN=C(C)N3C4=C2C(C)=C(C)S4)C=C1

Provider: James Bradner Laboratory at Dana-Farber Cancer Institute.

II. Storage:

1. The compound is more stable for long-term storage as a solid, which should be kept at 4 °C minimum although lower temperature storage is highly recommended.
2. The compound can also be converted to 10 mM DMSO stock solution, and kept at -20 °C. However, it has been observed that multiple freeze-thaws can cause the decomposition of the compound.

III. In vitro study condition:

1. For biological assay:
 Recommended concentration range from 0.5 nM to 50 μM
2. For cell assay:
 Recommended working concentration with cell assay is 250 nM.

IV. In vivo study condition:

Formulation: 10%HP-β-CD solution was used as a vehicle which improves the solubility of the compound. The Hydroxypropyl beta cyclodextrin is available at sigma-aldrich (catalog # is C0926-5G at sigmaaldrich.com). The 10% cyclodextrin is prepared in sterile water, which was filtered with 0.22 micron filter.

1. Detailed procedure for dosing the animal:
 - a. We make a concentrated stock of JQ1 in DMSO at a concentration of 50 mg/mL. This is frozen in aliquots sufficient for a single day's dosing (in our case for 20 adult mice).
 - b. Each day, a single vial is thawed at 37 °C.

- c. 9 parts of 10% cyclodextran is added dropwise while gently vortexing.
- d. The result is indeed a fine milky solution, but there should not be any visible particles.
- e. We dose 10 uL/gm of body weight to achieve a final dose of 50 mg/kg.
- f. We observed that the JQ1 remains active in frozen aliquots. However, repeated cycles of freeze-thaws was not tested and not recommended.
- g. The animal can also be dosed 100 mg/kg/day or two doses with either 30 or 50 mg/kg.

We sincerely hope this information is useful for your study with JQ1. More detailed experimental information can be found in the supporting information of our recent *manuscript*, (Nature, 2010, 468, 1067-1073). Please feel free to contact us if you have any questions.

Sincerely



James Bradner, M.D.