A certain receptor is selective to specific drugs and largely unknown. Does this selectivity originate from the binding pocket or are there other significant selectivity sites that preselect the drug? In this regard, another fundamental question is the binding dynamics and the path that the drug travels from the extracellular to the binding pocket. We still do not know how drug diffuses through a highly tortuous cavity to arrive at the binding pocket.

To address the above questions, we used molecular dynamic (MD) simulations and the state of the art post-processing software MSMBuilder and machine learning algorithm tICA (Time-Structure Based Independent Component Analysis) developed in the Pande Lab. First, we found an allosteric site responsible for the selectivity of µOR through monitoring binding affinity of different ligands. Second, we tried to understand the pathway from the allosteric to orthosteric site. Since ligands need to undertake specific orientations to permeate through the complex and highly-energetic barrier cavity of the receptor, conventional long-trajectory MD simulations starting from allosteric site can’t help us to unravel the binding pathway.

METHODS & RESULTS
To tackle the challenges mentioned in the introduction, we randomly generated 560 initial ligand positions and orientations as the starting points of the simulations. Each simulation was run for 200 ns to allow enough time for the relaxation of the ligand to equilibrium position/orientation, as well as to traverse metastable potential wells. These seeds were created in, as well as between, the allosteric and orthosteric sites. We analyzed the trajectories using tICA to find the most important reaction coordinates (Fig. 1). MSMs were also built on these trajectories to find the most populated states and their connectivity (Fig. 2).

WHY BLUE WATERS
Blue Waters is an extremely powerful and versatile computational resource. In addition to powerful CPU and GPU hardware, the fast interconnect allows us to do types of calculations (rapid adaptive sampling, Markov State Model construction, force field optimization, etc.) that we could not do on other platforms, such as distributed resources like Folding@home. Also, the availability of NAMD simulation package on Blue Waters has particular advantages for adaptive sampling and restrained equilibrations.

NEXT GENERATION WORK
Our future work will focus on the activation/deactivation mechanisms of µOR. Since the nature of conformational changes in the activation pathway of these receptors is very subtle, and delicate and encompasses microsecond to millisecond timescales, we will take advantage of Blue Waters to run thousands of MD simulations to be able to shed light into this drug attractive receptor. Our ultimate goal is to create a unified model of receptor activation, from µOR initial interaction with opiates to receptor activation to G protein interaction.

PUBLICATIONS AND DATA SETS