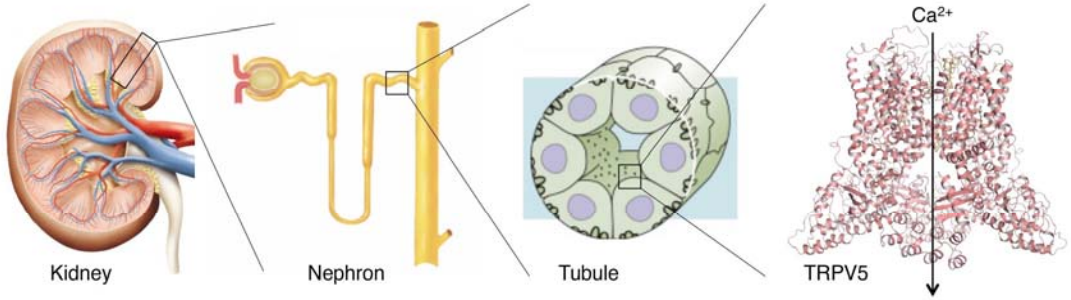
More than half a million people visit emergency rooms every year because of kidney stones. Most urinary stones contain calcium, and too much of this mineral in the urine predisposes to this painful condition. A joint study by the University of Pennsylvania, Rutgers New Jersey Medical School and Temple University, determined the three-dimensional structure of the TRPV5 ion channel, the major transporter protein that removes calcium from the urine (see illustration) (Nature Communications, 9:4198, 2018; https://www.nature.com/articles/s41467-018-06753-6). Overactive variants of this the TRPV5 protein increase calcium reabsorption of calcium, and have been shown to reduce the incidence of kidney stones.



The University of Pennsylvania group lead by Dr. Vera Moiseenkova-Bell used the Nobel Prize winning technique cryo-electron microscopy (CryoEM) to capture the TRPV5 protein both in the closed state, and in the open state, when it conducts calcium. CryoEM uses electron beams to take snapshots of a large number of frozen individual protein molecules. Sophisticated computer algorithms then average hundreds of thousands of these blurry images and sharpen them to the level that allows building of atomistic models, comparable to the resolution of X-ray crystallography.

The Rutgers group led by Dr. Tibor Rohacs experimentally verified the functional predictions of the cryoEM structure by changing individual amino acids and measuring ionic currents through these mutant channels. The open structure of the channel was determined in the presence of the membrane phospholipid PIP_2, which is a known physiological activator of TRPV5. When amino acids in contact with the lipid in the structure were changed, the experiments showed altered effects of the lipid. The closed structure was determined in the presence of the endogenous inhibitor calmodulin, which essentially plugged the pore of the channel protein. When a contact point amino acid was mutated in the channel, the inhibitory effect to calmodulin disappeared. These functional measurements demonstrated that the “frozen” snapshot of the 3D structure of the TRPV5 protein indeed reflects its physiological conformation. Aysenur Yazici a 4th year doctoral student at the School of Graduate Studies performed the electrophysiology experiments; she is shared first author (contributed equally) on the article.

The importance of these findings is twofold. On the practical level, once we know the structure of this protein in an open and closed state, novel molecules can be

designed to increase or decrease the activity of TRPV5, which could eventually lead to developing novel medications treating conditions such as hypercalcemia and kidney stones. On the conceptual level, biophysicists have long been wondering what conformational changes take place in ion channel proteins when they open and close. Both calmodulin and PIP_2 regulate the opening and closing of many ion channels, and the findings may be generalized to other important ion channels.