There are three main pathways of apoptosis (or programmed cell death): the extrinsic death receptor pathway, the intrinsic mitochondrial pathway, and the endoplasmic reticulum (ER) pathway. The pathway correlates with the cause of apoptosis. The BCL-2 family of proteins regulates the intrinsic mitochondrial pathway. The BCL-2 proteins are divided into three subgroups: prosurvival, BH3-only proapoptotic, and multidomain proapoptotic. The balance of the three subgroups determines whether or not a cell will undergo apoptosis.

The multidomain proapoptotic proteins, BAX and BAK, are activated by the BH3-only proapoptotic proteins. The “hit and run” model shows that the BH3-only proapoptotic proteins “hit” the multidomain proapoptotic proteins, displacing the transmembrane domain, the “run” when the formation of the rest of the multidomain protein changes. BAX, a cytoplasmic protein, relocates to the outer mitochondrial membrane (OMM) once activated, while BAK is found on the membrane before it is activated. Once the proteins are activated, the transmembrane domain anchors into the OMM and the formation of the proteins changes so the inner domains shift to the outside. From there, BAX and BAK form pores in the OMM that intermembrane proteins are released into the cytoplasm through.

In some forms of cancer, the balance between the prosurvival and proapoptotic proteins isn’t correct and the cell has either too many prosurvival or too little proapoptotic proteins. Since the cells no longer able to undergo apoptosis at the same rate, a cancerous tumor may be formed. Various treatments targeting these proteins are in different stages of clinical trials. These treatments aim to inhibit the prosurvival proteins or the genes that code for them. The limit of these treatments are mostly of them have negative side effects. One promising treatment is Venetoclax, which in clinical trials has proven to be effective, but the most significant side effect of the treatment was tumor lysis syndrome. Despite these challenges in this field, scientists are still unsure how the multidomain proapoptotic proteins create the pores in the OMM. Many researchers have predicted that the answer to this question will pave the way to future drug developments without the side effects of the current treatments.

During BAX and BAK activation, BH3-only proteins receive a cellular stress signal, and bind to the BAX or BAK proteins. This occurs at the BH3 domain, or the “back pocket” at helix 1. Once activated, BAX relocates to the OMM and its TM domain anchors into the membrane. From there, BAX and BAK form symmetrical homodimers at the BH3 domain. These dimers go on to form oligomers (the exact structure of which are unknown) that create the pores in the OMM.

Various models for pore formation have been suggested. The clamp model suggests that helices 2 and 3 are perpendicular to the OMM, with helices 6-8 parallel to the membrane on both the inside and outside of the TM domain. In the clamp model, helices 6 and 7 insert into the membrane. The in-plane model suggests that helices 2 and 8 insert into the membrane. The clamp model is similar to how bacterial toxins, like diphtheria toxin, create pores in cell membranes. The in-plane model suggests that helices 2-8 insert into the membrane, the exact structure of which are unknown.

Current research theories that the pores formed in the OMM will be toroidal from both proteins and ligands and that the pores created vary in size and shape. This leads to the theory that the pores in the OMM might not be all formed in the same way.