In 1861, the Chesapeake and Ohio Canal became the borderline between the Union and Confederacy being located adjacent to the Potomac River from Cumberland, Maryland, to Georgetown, a distance of 184.5 miles. During the following four years, the C&O Canal was subjected to movements of contending armies, Confederate raids, and government interference resulting in much damage to masonry structures, canal boats and the Canal prism. Consequently, Canal service became very irregular with trade greatly reduced.

The C&O Canal and B&O Railroad during the war became a major line of supply for the Army of Potomac and the civilian population of Washington, D.C. The Confederacy recognized this strategic importance and directed numerous actions against the Canal throughout the war to eliminate this supply line.

The first year of war, 1861, the Canal was initiated into the havoc and destruction the war would bring to the transportation system. The drastic decline in traffic and revenue was due to Federal use of the Canal as well as Confederate raids. After Union troops occupied the Virginia shore opposite Washington, D.C., the Alexandria Aqueduct was drained and used as a double track wagon road with an overhead bridge expanding to the top of the aqueduct. With this structure closed, Alexandria was cut from Canal trade.

In June the Confederates attempted their first effort to destroy the C&O Canal. Brigadier General Joseph E. Johnston, commander of Rebel forces at Harpers Ferry, was forced to retreat to Winchester by Union movements.

Before his retirement, Johnston sent teams out from Harpers Ferry to Dam 5 above Williamsport to create havoc on the waterway by burning lockgates, boats and with a failed attempt to destroy the log-cribbed Dam 5. Hereinafter, Union troops were assigned to protect the Canal not only from Great Falls to Williamsport, but along the entire towpath to Cumberland. For the remainder of 1861, the Canal was placed under military jurisdiction which resulted in Federal troops confiscating Canal equipment, mules, boats and to transport soldiers, undetected, artillery and supplies for the army. Many times the troops were very beneficial by providing labor to make repairs caused by raids and floods.

On November 5, President Davis ordered "Stonewall" Jackson to command the Valley District. Jackson initiated the greatest threat to the Canal during the entire war; Jackson learned the Canal was in winter operation and recognized the continued use of the Canal would be "of great service to the Federal army in Washington" and decided "to cut off western supplies by breaking Dam 5". After a failed attempt to destroy Dam 5 by Captain McLaughis with cannon and infantry, Jackson personally led a second attempt on December 17. That night a party of men crept halfway across the dam and slid into icy water. Undetected until morning, the Rebels succeeded in building a breastwork which protected them from Federal fire. After four days and nights of work in frigid water and running a deadly gauntlet across the dam, the dam was breached and Jackson pulled back. The canal resumed navigation around Dam 5 by the 21st, but the dam was not totally repaired until 1866.
Under the direction of military authority, the waterway was opened during the winter of 1861-1862 with the use of ice breakers. With a report of Canal operation in winter on January 1, Jackson made his final move to destroy the Canal, to counter a move by reinforced Federals on his supply base at Winchester. On January 5, the Confederates moved on Hancock and subjected the town to a two-day bombardment with very little damage to the Canal. After Union reinforcements arrived, Jackson returned to Romney for winter camp.

The year 1862 almost brought the Canal to the brink of collapse due to invasion, government confiscation and major floods. Receipts from all sources amounted to $72,642. while expenditures were $231,711.

On March 8, 1862, C.S.S. Merrimac sunk the U.S. warships Cumberland and Angress which seriously affected the operation of the Canal. Fearing the Merrimac would attack Washington, Secretary of War Stanton ordered the seizure of 60 canal boats to be filled with stone and sunk in a shallow channel in the Potomac to prevent the Merrimac from ascending the Potomac. By March 10, 103 canal boats were confiscated from the Canal to be readied for sinking, but fortunately Secretary of Navy Gideon Wells, upon hearing of the victory of the Monitor over the Merrimac, with Lincoln's consent, ordered no boats shall be sunk. Several weeks later President Lincoln, on board a steamer to Fortress Monroe, noticed the numerous boats and exclaimed, "That is Stanton's navy and is as useless as the pups of a man is to a suckling child." By the end of April the Canal authorities clamoured for the return of the boats or the Canal would cease operating as a supply line. The boats were desperately needed to transport coals and supplies to the army and to enervate funds to meet the cost competition of the Pennsylvania Railroads. Most of the boats were returned immediately except for forty boats used by the Federals to bridge the Rappahannock River.

With Lee's invasion of the North in September, 1862, the Monocacy Creek Aqueduct became the receiver of Rebel efforts to destroy the Canal. The Confederate lead division under Major General D.H. Hill crossed the Potomac at White's Ford and dispersed two Union regiments. Hill succeeded in draining the Seven Mile Level and leveling the banks to allow movement of the army. Damage was done to Lock 27, but the aqueduct proved too enormous to attempt destruction with little tools. On September 9, Lee ordered Major General John G. Walker to destroy the Monocacy Aqueduct and move onto occupy Loudoun Heights to capture Harpers Ferry. Walker tried to destroy the aqueduct, "but not a seam or crevice could be discovered in which to insert the point of a crow bar and the only resource was in blasting. But the drills were too dull and the granite too hard and after several hours the attempt was abandoned".

The spring of 1863 brought new life with increased trade as the war moved away from Maryland and deeper into Virginia. By June, Lee initiated his second invasion of the North that climaxed at the Battle of Gettysburg. As Rebel troops passed through the C&O Canal, raids were carried out along the entire Canal length resulting in minor damage. From his retreat from Gettysburg, Lee was pinned against high water of the Potomac River at Williamsport. Canal boats were used and framing from houses to construct pontoon bridges. To deny Federal use of the bridge, the tie-line was cut and the bridge destroyed. By August 4, the Canal was back in operation.
From August, 1863, to November, 1864, the Canal received numerous Confederate raids, all with the same objective to end Canal operations. The raids were a major hindrance to boating operations since the raids resulted in destruction of Canal boats, burned lockgates, stealing of mules and cargo and attempted destruction of structures. Many boatmen refused to operate fearing loss of life and their boats. The raids instigated by Stuart, Rosser, Mosby, White and Early were performed by small groups of calvary and with great speed. The damage sustained by these raids were repaired in a week period.

After the war restoration and improvement to the Canal began immediately, but was not completed until 1869. During the Civil War, the Canal was of great importance to the Union serving as a line of supply and communications for the Army of Potomac, Washington, D.C., and the eastern seaboard. Coal became the major cargo for the canal during the war that provided fuel for smelting iron and fire for steamships. Additional cargos of wheat, flour, corn, agricultural products moved up and down the waterway to feed Union armies and the Washington population. Despite the destruction and loss of trade, the C&O Canal played an active and contributing role during the War Between the States.