UV radiation can damage DNA to mainly form cyclobutane pyrimidine dimer or (6-4) photoproduct. Such lesion may eventually lead to skin cancer. Photolyase, a flavin photoenzyme, can revert such damage with high repair efficiency. Here, we combined femtosecond spectroscopy and molecular biology and have completely mapped out the entire repair evolution at the most fundamental level by following the dynamics from the initial reactants, to the fleeting intermediates and to the final repaired products. By resolving more than nine elementary steps in the complex enzymatic reaction, we captured five electron-transfer reactions and also bond breaking and forming processes. These dynamics are in synergy to achieve a maximum repair efficiency. Various mutations were also carried out to identify the critical residues in the active site for function. We carefully examined the various photolyases and observed a unified electron transfer mechanism with determination of the critical role of the unique folded cofactor structure.