Many essential cellular proteins use iron-sulfur (Fe-S) clusters as cofactors. These proteins often serve as enzymes, components of the electron-transport chain or as intracellular sensors. Prior to the use of the cluster in a protein, it needs to be formed or created *de novo*. In total, four different mechanisms of Fe-S cluster biogenesis can be used by the eukaryotic cell – ISC, CIA, SUF and NIF. All of these pathways include a specific targeting system for delivering the cluster to its acceptor protein. Errors in biosynthesis of Fe-S clusters are mostly lethal and can lead to failure in development of multicellular organisms. Despite this a better characterization of these mechanisms is needed as research is currently still in progress. This bachelor's thesis provides current information regarding the mechanisms of Fe-S clusters biogenesis in eukaryotes acquired mostly from mammalian cells, including humans, and from well-known model organisms such as *Saccharomyces cerevisiae*, *Arabidopsis thaliana*, and parasitic protist *Giardia intestinalis*.