Unsaturated fatty acids, which are the essential components required for normal cellular function, play a role in controlling the fluidity of membrane and are precursors of regulatory molecules. Recently, there has been considerable interest in microbial fatty acid production. The oleaginous filamentous fungus, *Mucor rouxii*, is one of the promising sources of essential oil production. However, there is little information on fatty acid synthesis in this fungus. Therefore, the *M. rouxii* mutant defective in fatty acid desaturation would be a useful tool for understanding the fatty acid-synthesizing process. In this study, two approaches including ultraviolet light (UV light) and electropolation combined with chemical mutagenesis (MNNG) were used to induce mutation of *M. rouxii*. Combining method resulted in the survival rate of *M. rouxii* less than UV light. The mutants having low unsaturated fatty acid content were isolated using filtration enrichment technique and cultivation at low temperature. The results of fatty acid analysis showed that unsaturated fatty acids contents of these mutants were reduced. In addition, the changes in morphology of *M. rouxii* mutants such as sporulation and mycelial color were found. The study of the expression of fatty acid desaturase genes of these mutants is in progress.