Abstract: *Tricholoma equestre* (L.:Fr) Kummer, an edible wild mushroom, is consumed in many countries in the world for its particular good taste. From 1992 to 2000, twelve cases of delayed rhabdomyolysis have occurred in France with 25% lethality (Bedry et al, 2001) and two cases in Poland (Chodorowski et al, 2002). These were the consequences of consumption of large quantities (>500g of fresh mushroom/person) in several consecutive meals (>3). The symptoms are characterised by extreme muscle weakness with cramps, sweating and nausea followed by destruction of skeletal muscles and consequent increase of creatin-phospho-kinase in blood. The substance triggering the toxicity in humans is being investigated. Flavomannin-6,6'-dimethylether has been purified as following: removal of fats in petroleum ether, extraction by refluxing in CHCl₃:CH₃OH, (2:1); purification step1, by column chromatography on silica gel, elution by (CHCl₃:MeOH, 9:1 and CHCl₃-CH₃OH, (7:3); step2, preparative HPLC on reversed phase C18 (6µm), elution with a gradient of water, TFA and acetonitrile. 700g of fresh mushroom yielded 160 mg of material at the first step of purification and finally 60 mg of a pure compound characterised as Flavomannin-6,6'-dimethylether by mass spectrum in FAB⁺ mode, (597 [M+Na]⁺, 575 [M+H]⁺) and ¹H NMR plus ¹³C NMR spectra using a 500 MHzs Brücker equipment. Concentrations causing 50% growth inhibition (IC₅₀) on human Caco-2 cells and Hep-G₂ cells were found to be 96 ± 3 μg/mL and 100 ± 8 μg/mL respectively after 24 h of incubation using the MTT test. Furthermore, the compound does not induce apoptosis as measured by DNA fragmentation in agarose gel electrophoresis or determined by flow cytometry. It rather arrests the cell cycle in G0/G1 phase and increases p27 protein levels, suggesting that cells death occurs mainly by necrosis in vivo. This is congruent with muscles cells lyses and inflammation observed in patients.

**Key words:** Flavomannin-6,6'-dimethylether, *Tricholoma equestre*, human CaCo-2 and Hep-G₂ cells, cell cycle arrest, mechanisms of cell death