

PARAMETER EFFECTS ON ISOLATION OF XANTHOHUMOL FROM HOPS EXTRACTS BY SUPERCRITICAL FLUID CHROMATOGRAPHY

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Nowadays, costumers demands functional and healthy products. Natural additives provide the final product with a healthy value instead the use of synthetic chemical compounds in food products. Xanthohumol is a natural compound which is only found inside hops plant (*Humulus lupulus* L.). The hops plant is a dioeciously twining perennial which is cultivated widely throughout the temperate zones of the world. The inflorescences (hop cones or 'hops') are used in the brewing industry to give beer its characteristic flavour and aroma. Xanthohumol is a natural compound which is only found inside hops plant (*Humulus lupulus* L.). It shows toxicity to different cancer cells such as human breast, colon and ovarian cancer cells. Xanthohumol also appears to have a role as a fairly powerful antioxidant, even more than vitamin E.

Chromatography is a technic used to analyse or separate different components from a sample. Fundamentally, chromatographic techniques could divide depending on the state (gas, liquid or supercritical fluid) of their mobile phase. A sample is solved inside the mobile phase and then, this stream goes through a column (packed column or capillary column). That column is called stationary phase. Each component of the sample exhibits more or less affinity to the mobile phase or stationary phase, therefore each component come out from the column in a different time depending on that affinity. In this research, Supercritical fluid chromatograph was utilized to separate xanthohumol from hops extract. A mixture of CO₂ and a modifier was employed as a mobile phase to increase the solvent strength of CO₂. Different columns (Waters Symmetry C18 5µm 4.6x250mm; LiChrospher[®] 100 DiOL 250x4mm 5µm, Waters Symmetry[®] C18 5µm 4.6x250mm plus Zorbax SB-C18 3.5µm 4.6x150mm), three different pressures (220 bar, 250 bar and 270 bar), three different temperatures (30°C; 45°C; 60°C), different flow-mass rates (2,81 g/min; 3,09 g/min; 3,38 g/min) and a flow rate gradient were used. Pure ethanol, a mixture of ethanol and 4% of citric acid, and ethyl acetate were applied as modifiers for the mobile phase. Two different hops extracts, one rich in xanthohumol and another one rich in fatty acids, were mixed and used to develop the experiments. Isolation of xanthohumol from hop extract was achieved. Optimal parameters for that separation will be shown.