OPTIMIZATION OF A HIGH PERFORMANCE THIN LAYER CHROMATOGRAPHY METHOD TO QUANTIFY SOLANINES IN POTATOES.

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In the frame work of our studies on natural toxins present in food, we have previously carried out experiments on linamarin [1]. Now we report on the analysis of potato, *Solanum tuberosum* L., that contains glucoalkaloids (GAs) generally called "solanines".

GAs are toxic: they inhibit the cholinesterases. Generally undamaged tuber contains about 20-150 mg total GAs/kg depending on the cultivar of potato. Several scientists consider 200 mg GAs/ kg potato as a non-toxic concentration [2].

Usually GAs (α-solanine and α-chaconine) in potatoes are quantitatively determined by high performance liquid chromatography (HPLC). The HPLC methods involve time-consuming sample preparation by solid phase extraction (SPE) and non specific UV detection at 202 nm [3]. GAs do not have strong chromophores and a derivatization step is generally needed to specifically determine them. Therefore high performance thin layer chromatography (HPTLC) can be a choice method.

We thus developed the following method:

Thin layer chromatography is performed on a silica gel $60F_{254}$ HPTLC plate (Merck). Before application of the samples, the layer is prewashed with methanol, dried in air, and activated at 110°C overnight. The samples (5 µl/spot) are applied, in bands of 4 mm, by means of an automated applicator. The plate is heated for 30 min. at 90°C. Vertical development of the plate is performed with a saturated mixture of dichloromethane-methanol-water-ammonium hydroxide (70:30:4:0,4; v/v/v/v) up to a distance of 85 mm. After the development step, the chromatogram is first air-dried for 15 min. and then heated at 90°C for 60 min. Afterwards visualization is directly accomplished by dipping three times the plate into a modified Carr-Price reagent ($SbCl_3$–acetic acid–dichloromethane, 2:2:6, p/v/v) and subsequently heating on a hot plate at 105°C for 5 min. The chromatographed GAs all yield red chromatogram zones on a colorless background. The densitometric quantification is performed at 507 nm by reflectance scanning. Since these GAs zones begin to turn purple after 30 min, it is necessary to carry out quantification within this time frame.

GAs were extracted from potatoes using several methods cited in the literature [4]. Results obtained by our HPTLC technique will be compared and discussed with our modification of the Carr-Price visualization method.

After determination of the appropriate response function, the proposed method was validated. Good results with respect to linearity, accuracy and precision were obtained in concentration range studied..

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