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Application of High Performance Thin Layer Chromatography-Densitometry and UV- Visible Spectrophotometry for the Simultaneous Determination of Thiamine in Green Beans

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Abstract : Two methods are described for the simultaneous determination of thiamine in green beans. The green beans were grinded and thiamine was extracted as bases into distilled water, separated by the first method, HPTLC silica gel 60 F₂₅₄ plate using methanol: water: acetic acid: ammonia (5:4.5:0.5:0.75) as mobile phase followed by densitometry measurement of its spot. The second method, a highly sensitive colour reaction has been developed. Thiamine was reacted with bromothymol blue to form an ion association complex in a weak base aqueous solution in the presence of some solubilization agents such as polyvinyl alcohol and analyzed by using UV- visible Spectrophotometry. The solution was measured at a maximum absorbance length of 430.5 nm. The first method showed that the detector response was linear for concentrations between 100-500 µg/ml (r=0.998). The limits of detection and quantitation were 33.7 µg/ml and 113.1 µg/ml, respectively. The second method was found to offer good linearity (18-26 µg/ml, r=0.998) with 0.6 µg/ml limit of detection and 1.9 µg/ml limit of quantitation. Thiamine contents from both methods were analyzed. The result showed that average contents of thiamine from both methods were 0.0396% and 1.0009%, respectively. The two proposed methods were successfully applied to the determination of thiamine in green beans.

Keywords : Green beans, HPTLC-Densitometry, Spectrophotometry UV/Vis, Thiamine.

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