ABSTRACT

An accurate, simple and distinctive HPLC-DAD analytical method is developed for the simultaneous determination and validation of illicit substances such as stimulants, anti-depressants, diuretics, dyes and vitamins in weight loss formulations. To gain popularity, for the sake of money and increase effectiveness of product, different manufacturers add illicit drugs in weight loss formulations. Caffeine, Ephedrine, Nicotinamide, Phenolphthalein and Fruosemide were analyzed during this process. The developed HPLC-DAD method is very simple, specific and has gradient mode .This validated method is suitable for routine control analysis and recommended for the determination of any illicit, impurity and adulterant component. Good chromatographic separations between caffeine, ephedrine, nicotinamide, phenolphthalein fruosemide and stress induced degradations products were accomplished within 10 minutes using C-18 column with acetonitrile (organic modifier) and 50mM sodium dihydrogen phosphate buffer and pH adjusted to 3.8 by using ortho-phosphoric acid. The ratio of organic modifier during the gradient was 30:60:80:80:30.The flow rate of mobile was 1 ml/min and detection was done by DIODE ARRAY DETECTOR at 254nm.Developed method was validated according to ICH guidelines. Linearity was from (12.5-225µg/ml) for caffeine, ephedrine, nicotinamide, phenolphthalein and fruosemide. The LOD values were found to be 0.5µg/ml for caffeine,0.8µg/ml for nicotinamide,1.1µg/ml for ephedrine,0.5µg/ml for phenolphthalein and 1.2µg/ml for fruosemide. The LOQ values were found to be 2.6µg/ml for caffeine, 3.6µg/ml for ephedrine, 2.9µg/ml for nicotinamide,3.4µg/ml for phenolphthalein and 2.7µg/ml for fruosemide. All the analytes including the degradation products were separated with acceptable peak tailing and resolution. This established method can successfully be used for the simultaneous determination of any illicit drug in weight loss formulations or any refine dosage form. This method was also able to separate other impurities and illicit component from the suspected samples.

Key Words: Caffeine, Nicotinamide, Ephedrine, Phenolphthalein, Fruosemide and HPLC-DAD.