



ABSTRACT

A linear, accurate and precise HPLC-DAD method has been developed and validated for the simultaneous determination of betamethasone valerate and dexamethasone in poultry feeds. The chromatographic separation was executed within the run time of 4 minutes using C18 column (250 mm x 4.6 mm, 5- μm particle) and gradient elution system consisting of tetrahydrofuran (THF) pH at 4.2 as mobile phase A and methanol as mobile phase B (70-100%) at the flow rate of 1.0 mL min⁻¹. The DAD detections of the components were carried out at the wavelengths of 254nm and 210nm, and column was kept at ambient temperature. All the analytes were separated with acceptable peak tailing and resolution. Quantitation and linearity was achieved at 254 nm over the concentration range of 20- 200 $\mu\text{g/ml}$ for Dexamethasone and 50-200 $\mu\text{g/ml}$ for Betamethasone valerate after derivatization. The limit of detection (LOD) and limit of quantitation (LOQ) values were found to be 0.55 $\mu\text{g/ml}$ and 1.20 $\mu\text{g/ml}$ and 3.4 $\mu\text{g/ml}$ and 2.8 $\mu\text{g/ml}$ for Dexamethasone and Betamethasone valerate, respectively. The anticipated method was optimized and validated for linearity, accuracy, precision, LOD, LOQ, selectivity and robustness in the light of ICH guidelines and it was successfully employed for the determination of illicit drugs in poultry feeds. 05 commercially available poultry feed samples were collected from the local market and extracted in methanol. The developed HPLC-DAD method was applied on the poultry feed samples for the simultaneous detection of betamethasone valerate and dexamethasone. The results revealed that all the 5 samples contained illicit drugs.

Keywords: HPLC-DAD, illicit drugs, stress induced degradation, steroids, betamethasone valerate and dexamethasone.