

**ABSTRACT****Comparison Validation of TLC-Densitometry and TLC-Bioautography Method for Determination of Oxytetracycline in Shrimp**

A simple Thin Layer Chromatography (TLC) Densitometry and TLC-Bioautography has been used for the identification and quantification of oxytetracycline. Validation both of the methods has been developed for determination oxytetracycline in shrimp, included parameter linearity, accuracy (recovery), precision (CV), limit of detection, and limit of quantification, as a purpose to verifying that the parameters is appropriate with literature. Thin Layer Chromatography of oxytetracycline has been carried out by using silica gel GF<sub>254</sub>, which has been treated by EDTA 1% as a stationary phase, chloroform: methanol: sodium EDTA 1% (55:25:10 v/v) as mobile phase. Retardation factor (Rf) of Oxytetracycline was 0,44. *Staphylococcus aureus* was used as a bacterial test. The analyte spots were quantified by using densitometry method at wavelength 365 nm. The response was found to be linear at the amount of oxytetracycline between 100 ppm - 500 ppm. Oxytetracycline's regression equation line was  $Y = 9992,1091X - 1861,706$ ,  $r$  value = 0,9916, and  $V_{x_0} = 7,9332\%$  for densitometry and  $Y = 0,8573X - 7,5901 \cdot 10^{-3}$ ,  $r$  value = 0,9942, and  $V_{x_0} = 1,6743\%$  for bioautography. Recovery and CV were  $61,7450\% \pm 2,4737$  and  $6,8402\%$  for densitometry, and  $59,7287\% \pm 2,9133$  and  $4,8775\%$  for bioautography. Limit of detection value was 12,10 ppm and 20,04 ppm for densitometry and bioautography respectively. Limit of quantitation value was 40,35 for densitometry, and 50,10 ppm for bioautography. Conclusion, all of the validation parameters were appropriate with literature, except accuracy and precision, but the methods can be applied for determining oxytetracycline in shrimp, and there's no meaning difference between both of them for determining oxytetracycline in shrimp for accuration parameter; for precision, there's difference between TLC-Densitometry and TLC-Bioautography.

Keyword: validation, oxytetracycline, TLC-Densitometry, bioautography, shrimp.