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## Analysis and Estimation of Eugenol Content in Microemulsion Formulation Containing Clove Oil (*Syzigium aromaticum*)

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### ABSTRACT

Oil in water based Microemulsion formulation (ME) containing Clove oil was analyzed and quantitated for its Active Ingredient (A.I.) by Gas chromatography Mass Spectrometry (GC-MS). Clove oil based microemulsion formulation (6%) was developed in-house and tested for its active ingredients. Standard sample preparation methods for testing the Emulsifiable Concentrate (EC) formulations were found ineffective for the estimation of A.I. (i.e. Eugenol) in ME formulation. A sonication based extraction method was developed and optimized to extract and quantify the Eugenol (Clove oil) and its other components from the micro-emulsion (6%, ME). The main five components analyzed in clove oil technical by GC-MS were: Eugenol (92.11%), Eugenol acetate (0.43%), alpha-Caryophyllene (0.90%), Caryophyllene (5.47%) and Caryophyllene oxide(1.09%). Probe based Sonication was found to be the effective approach in breaking the micelle and extracting the A.I. into solution in less time as compare to the bath sonication. Amongst five different extracting solvents (viz. Acetonitrile, Acetone, n-Hexane, Ethyl acetate and Dichloromethane) Ethyl acetate showed better extraction. The same solvent also showed less time for complete A.I. extraction. GC-MS instrument showed linear response to A.I., with  $r^2 = 0.988$ , measured at 0.1, 0.5, 1.0, 10 and 50 µg/ml. concentrations. Mean recovery of method is more than 95 % with R.S.D below 20%.

### **Graphical Abstract**



Extraction of clove oil from its ME formulation and GC-MS, TIC of clove oil technical

Keywords: Ultra-Sonication, Microemulsion, Clove-Oil, Gas chromatography mass spectrometry.