Abstract

Background: - Vitamin A is one of the crucial nutrients recognised with its essence for good vision and regulation of growth and differentiation of cells. Accordingly, deficiency of vitamin A would lead to improper stem cell differentiation; abnormal bone growth and tooth development; diminution in the cells in inborn and attained immune function; diminishing the barrier to infection and suffering in the development of epithelial cellular; changing and reforming the growth and development of embryos, and many other impacts. The present study aims to develop a sensitive robust, and easy LC-MS/MS-based assay for measuring vitamin A in human plasma, and measure plasma vitamin A in healthy and diseased Emirati population.

Methods:-

A positive ion electrospray ionisation (ESI) LC-MS/MS method was used in the Multiple Reaction Monitoring (MRM) mode for quantification. It involved i) liquid-liquid extraction, ii) a guard column together with C18 Ascentis Express F5 column iii) Internal standard, 25-Hydroxyvitamin D3 (6, 19, 19-d3), and iv) identification via ESI and monitoring of three fragmentation of the parent ion. To demonstrate the practical usefulness of our method, blood samples were collected from 277 Emirati baseline and follow-up volunteers and 175 healthy control volunteers; mixed-gender with 73 males in the age group of (18–82 years) and 204 females in the age group of (18–65 years) and healthy volunteers female 167 and male only 8 participants where vitamin A was quantified. The method was validated according to FDA-US guidelines.

Results and Discussion:-

The new method allowed chromatographic separation and quantification of vitamin A. The new assay could detect 0.48 ng/mL of vitamin A in serum. The calibration curve ranges from 7.8 to 1000 ng/ml. The method has been applied on a sample of 277 Emirati individuals including 277 baselines, 277 follow-ups, and 175 health samples. The characteristics of the sample as follows: mixed-gender with 73 males in the age group of (18–82 years) and 204 females in the age group of (18–65 years). The level of vitamin A deficient appears in the baseline volunteers in the female sample which is higher than the lower acceptable value of (600 ng/mL >372 ng/ mL > 200 ng/ mL). This perceived result has increased upon supplementation in the follow-up samples to reach 440 ng/mL. While the male results are within the acceptable limits upper and lower limits for the (600 ng/mL> 438 ng/mL > 200 ng/mL, 200 ng/mL< 540 ng/mL < 600 ng/mL). The healthy sample showed 718 ng/mL combining the female and male. The results clearly show that the concentrations of vitamin A for both females and males increased after supplementation (372 ng/mL to 440 ng/mL for female sample, and 438 ng/mL to 540 ng/mL for male sample). The result shows a sufficient level of vitamin A in the participants.

Conclusions:-

To our knowledge, this is the first specific, reliable, reproducible and robust LC-MS/MS method developed for the accurate detection of Vitamin A. The method can detect low levels of vitamin A. This analytical method does not require time-consuming derivatisation and complex extraction techniques and could prove very useful in clinical studies.