SUMMARY

The objective of this study was to enhance and accelerate the ability of the green alga *Chlorella vulgaris* for the production of high energetic compounds mainly the production of biodiesel with high quantitative and qualitative value using different a biotic stress factors for the over production of lipid.

The following is a summary of the main experimental results obtained:

1. The obtained results revealed that the growth of *Chlorella vulgaris* was markedly increased with increasing the concentration of sodium nitrate up to 5 mM of sodium nitrate more than its corresponding control concentration in BBM (3mM). On the other hand, the biosynthesis and accumulation of lipids were markedly increased as nitrate deprived up to 0.1 mM. The previous results indicate that nitrogen deprivation is a feasible tool for the over production of lipid contents and the relation between lipids and growth was found to be inversely.

2. The mean growth rates and cell productivity of *Chlorella vulgaris* was gradually inhibited with increasing the salinity levels (NaCl salt) up to 0.45 mM more than the corresponding control but, the over production of lipids was directly increased with increasing the level of salinity up to 0.45 mM of sodium chloride. The previous results indicate that salinity is a feasible tool for the over production of lipid contents and the relation between lipids and growth was found to be inversely.

3. The mean growth rates of Chlorella *vulgaris* was gradually increased ferrous sulphate concentrations up to 35.25 µM. On the other hand, 44.06 µM of ferrous sulphate resulted in a noticeable inhibition in the mean growth rate of *Chlorella vulgaris* less than its corresponding control. Results also, revealed that the over production of lipids was subsequently increased slightly with increasing the level of ferrous sulphate up to 35.25 µM. But, any further excess more than 35.25 µM resulted in an inhibition in the lipid accumulation.

4. The results revealed that the growth of *Chlorella vulgaris* was directly proportional with increasing the concentrations of manganese chloride salt more than its concentration in control cultures (7.3 µM). The biosynthesis and accumulation of lipids in *Chlorella vulgaris* was slightly increased with increasing concentrations of manganese chloride salt (µM).

5. Treating the alga *Chlorella vulgaris* under different concentrations of cobalt nitrate salt revealed that the growth was slightly affected under different cobalt nitrate concentrations. The growth was more or less similar than its concentration in control cultures (1.68 µM). Results also manifested that the biosynthesis and accumulation of lipids in *Chlorella vulgaris* was slightly affected with the variable concentrations of cobalt nitrate (µM).

6. The mean growth rates and cell productivity of *Chlorella vulgaris* grown under different concentrations of H_2O_2 (mM) indicated that the growth of *Chlorella vulgaris* was subsequently decreased as it

treated with different concentrations of H_2O_2 up to 4mM. While treating the alga with 6mM H_2O_2 causes a great suppression in the growth and the alga enter the death phase after 2 days of incubation period, so this concentration is consider the lethal one. The impact of different concentrations of H_2O_2 on the lipid contents of *Chlorella vulgaris* revealed that the biosynthesis and accumulation of lipid was subsequently increased with increasing concentrations of H_2O_2. And the maximum production of lipid was recorded in cultures treated with 4 mM of H_2O_2.

7. The different stress factors (specially the concentrations that give the highest lipid content) revealed that all treatments resulted in an obvious decrease in photosynthetic pigments of *Chlorella vulgaris* (Chlorophyll a, Chlorophyll b, carotenoids and total pigments) compared with the corresponding control. On the other hand the other hand results indicated that the only increases in photosynthetic pigments were recorded when the alga treated with 35.25 µM ferrous sulphate.

8. The results showed that there were great variations in the major biochemical contents of *Chlorella vulgaris* treated with the best concentrations of different nutrients for lipid production. The resulted data indicated that under the previous stress factors the biosynthesis and accumulation of lipids in *Chlorella vulgaris* varies inversely with the accumulation of carbohydrates and protein contents. The data also, showed that the lipid content determined gravimetrically don't differ so much from the lipid determined spectrofluorometrically using Nile red stain.

9. Gas chromatographic analysis of fatty acid methyl esters % of *Chlorella vulgaris* treated with the best concentrations of different nutrients for lipid production revealed that thirteen fatty acids were identified from Capric (C12:0) to Lignoceric fatty acids (C24:0). It was found that treating *Chlorella vulgaris* with 0.1mM of $NaNO_3$ and 0.45mM of NaCl resulted in an obvious increase in total saturated fatty acids (SFA) from their corresponding control. On the other hand, treating the alga with 35.25 µM $FeSO_4$, 12 µM $MnCl_2$, 2 µM $Co(NO_3)_2$ and 4 mM H_2O_2 resulted in an obvious decreases in total SFA from their corresponding control. And the resulted decreases in total SFA was found to be shifted towards the biosynthesis of unsaturated fatty acids (USFA) and vice versa. The maximum yield of SFA was found in the lipid extract of cultures treated with 0.1 mM of $NaNO_3$. Data also showed that the major constituents of the lipid fraction of *Chlorella vulgaris* were found to be C14, C16, C18, C18:1, C18:2 and C18:3 fatty acids. Also, it was found that all of the treatments in the present work resulted in formation of C24 fatty acid which wasn't recorded in the lipid profile of untreated alga. The results of methyl ester profiles for all treatments suggested that 0.1mM of $NaNO_3$ and 0.45mM of NaCl considered suitable conditions for a lipid production with highly SFA in *Chlorella vulgaris*.

10. The obtained results were discussed in the light of these recorded by other investigators.