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**Comparative application *in vitro* and *in vivo* of antimicrobial and Nano silver in controlling of some fish diseases pathogens**

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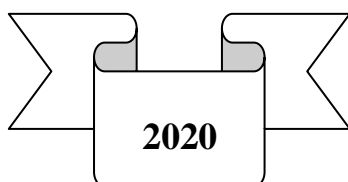
**(Bacteriology and Mycology)**

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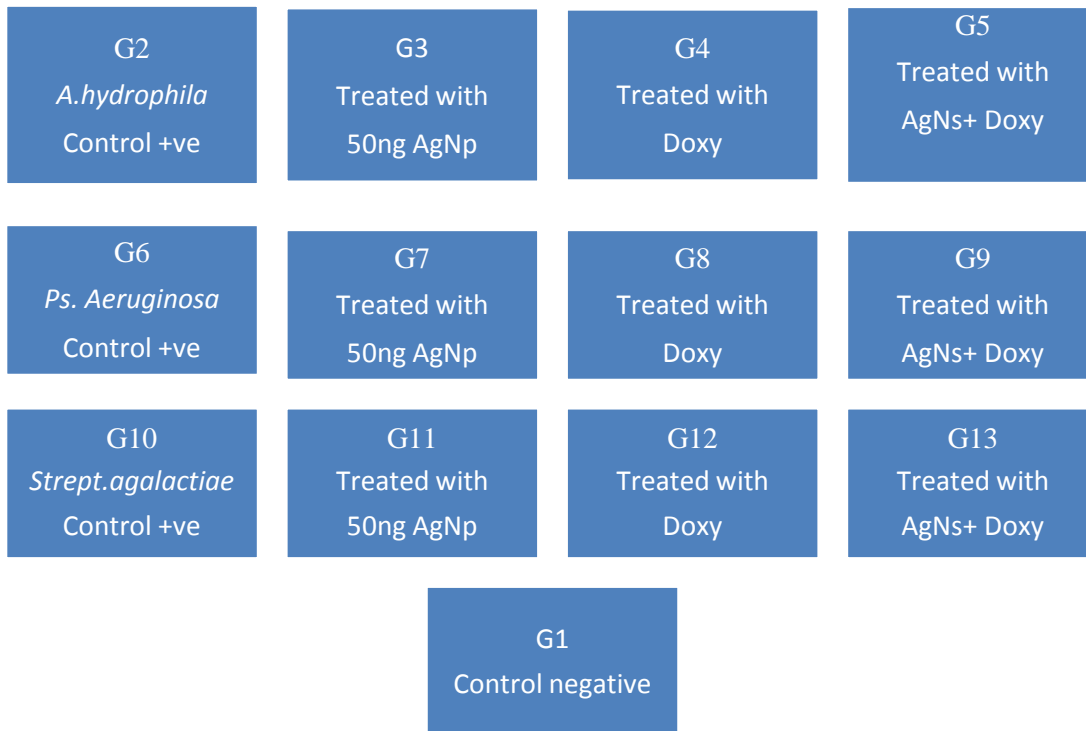
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## 6- SUMMARY

The Current study aims are the comparison between the traditional antibiotics used in treating fish diseases caused by *A. hydrophila*, *P. aeruginosa* and *Strept. agalactiae* and the use of AgNPs as a new trend for combating the diseases. Moreover; the combination between both AgNPs and the most potent and widely used antibiotic (doxycycline) was used. The *in vivo* study was done and the residues of AgNPs in fish flesh were noted.

### **This performed by:**

- 1- Obtaining a reference strain and field strains of some bacteria those causing fish diseases as *A. hydrophila*, *P. aeruginosa* and *Strept. agalactiae*.
- 2- These isolates cultured and confirmed by the different bacteriological methods.
- 3- Antimicrobial sensitivity test was done to determine the most potent antibiotic against these pathogens which revealed the common one as doxycycline.
- 4- Application of different concentrations of AgNPs *in vitro* to determine the best concentrations that achieved the MIC to bacteria. The best concentration was 50 ppm that can give better inhibition zone.
- 5- TEM was used to check the effects of AgNPs on bacterial cells and the extent of cell damage
- 6- *In vivo* experiment was done on a 13 groups of *O. niloticus* for 30 days.
- 7- Histopathological examination of fish gills and liver and muscles to find out the effects of bacteria on fish and the curative effects of antibiotics as compared with AgNPs alone or combined with doxycycline.
- 8- TEM was performed on fish flesh to detect whether AgNPs has residues in muscles after treatment and if found until when?



### The result revealed that:

The results revealed that the use of AgNPs in treatment of fish diseases is very effective at the field with restriction of the withdrawal time.

- At concentration of 50 ppm of AgNPs agar well diffusion disc showed inhibitory zones of bacterial growth of the 3 bacteria species.
- Using of both AgNPs together with doxycycline *in vitro* produced a larger inhibition zone.
- The turbidity test revealed that lower turbidity upon using AgNPs.
- The effect of AgNPs on biofilm of bacteria, biofilm was reduced upon treatment as compared with normal.
- The effect of AgNPs on cell respiration was great as compared to normal groups
- The TEM test confirmed the different modes of actions of AgNPs on bacterial cells.

- The result of Challenge test revealed that the use of AgNPs or AgNPs combined with doxycycline after challenge with *A. hydrophila*, *P. aeruginosa* and *Strept. agalactiae* as compared with the positive control groups and even the groups treated with antibiotic only. Moreover, the groups that treated with both AgNPs combined with doxycycline showed no mortalities and the signs of diseases healed completely.
- The histopathology of liver, muscles and gills showed that treatment with AgNPs lead to minimal tissue damage.

**In conclusion:**

Recently, infections correlated with antibiotic-resistant bacteria have increased in aquaculture industry. AgNPs as antibacterial agent alone or combined with doxycycline gave an effective action against the most threatening bacteria at fish farms; *A. hydrophila*, *P. aeruginosa* and *Strept. agalactiae* whether *in vitro* or *in vivo*.