

## Impacts of Algal Cells and Humic acid on the Formation of Disinfection By-Products during chlorination of drinking water

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### ABSTRACT

The production of high quality drinking water requires disinfection process to control the pathogenic microorganisms and algae. The algae and natural organic matter represented by humic acid (HA) are considered important precursors for the formation of disinfection by-products (DBPs). In the present work, the contribution of algae and HA in trihalomethanes (THMs) formation was demonstrated. The effects of Chl (a) concentration, HA concentration, Cl<sub>2</sub> dose and pH of water on formation potential of THMs were investigated. The results showed that the yield of all DBPs were higher upon the chlorination of *Scenedesmus obliquus* and HA than in case of *Scenedesmus obliquus*. Also the concentrations of TTHMs were increased as the reaction time was extended. However the kinetics of TTHMs formation was very rapid, where most of TTHMs were formed after 15 min of reaction time. It was concluded that the THMs formation highly dependent on the algal content rather than HA indicating that the control of algae in water is highly recommended in order to avoid the formation of high levels of harmful THMs.

**KEYWORDS:** Algae; THMs; HA; Chl(a); disinfection by-products.

### INTRODUCTION

Disinfection has been applied in drinking water treatment process for removal of waterborne pathogens and preserves human health. Application of chlorine as disinfectant started in the early 1900s (Richardson *et al.*, 2000). However, this step of treatment is usually accompanied with a formation of by-products (disinfection by-products; DBPs). The formation of DBPs in drinking water has proved to cause serious public health concerns (Rook, 1974 and Singer, 1994). Therefore, restricted regulations have been stated for the presence of these DBPs in drinking water. The United States Environmental Protection Agency regulates THMs in drinking water at 80 µg L<sup>-1</sup> (EPA 2014). While the permissible limits of four total trihalomethanes (THM) species are 100 µg L<sup>-1</sup> in the European Union and Egypt.

From the beginning of seventies of the last century, it has been known that natural organic matter (NOM) is the major organic precursor to the formation of DBPs during drinking water treatment. Aquatic NOM is a complex mixture of many chemical groups, including humic substances, simple carboxylic acids, amino acids, proteins, and carbohydrates (Croue *et al.*, 2000). But, humic acid (HA) is the major fraction of natural organic matter in water; it can interfere with several water treatment processes (Chen *et al.*, 1977). It is known that HA can react with oxidants to produce small molecular organic substances, which contribute to producing DBPs. In particular, HA is the principal precursor of trihalomethanes (THMs), which are potentially carcinogenic (Nie *et al.*, 2010; Xue *et al.*, 2011).

Algae are considered also as active precursors for the organic matters that present in freshwater. Algal cells have an extensive variety of organic nitrogen compounds, for instance; proteins, peptides, amino sugars, traces of other organic acids and polysaccharides. They are found in rivers, reservoirs and lakes. The increase of algal cells and their excreted metabolic substances may cause a series of problems for water treatment such as: (1) undesirable taste and odour; (2) potential toxicity concerns, particularly with blue-green algae which may produce algal toxins; (3) interference by both algal cells and their metabolic substances with the coagulation process (Plummer and Edzwald, 2002; Henderson *et al.*, 2008 and Takaara *et al.*, 2007) and (4) Algal cells can also penetrate filters and lead to DBPs formation from chlorination within the water treatment plant (Ma and Liu, 2002 and Rosario-Ortiz, 2013). The formation of DBPs from algae varies according to algal species growth phase and chlorination conditions e.g. contact time, chlorine dosages, pH, temperature (Fang *et al.*, 2010).

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Algal-derived organic materials generally were small molecules, exhibiting very different properties (Henderson *et al.*, 2008), while humic substances generally were large. In water treatment, large molecules could be easily removed by coagulation (Amy *et al.*, 1992), while small hydrophilic molecules were difficult (Boyer *et al.*, 2008). It was reported that with an increase in the percentage of low-molecular-weight dissolved organic carbon (DOC, <5 kDa) the removal efficiency of the total DOC decreased (Cheng and Chi, 2003). Eventually, these hydrophilic low molecular-weight molecules can serve as effective precursors of DBPs especially dihaloacetic acids (Hua and Reckhow, 2007). As compared to humic substances, algae can contribute significantly to the DBP precursors. Experiments with four algal species in different stages of growth showed that maximum chloroform yields from algal cultures exceeded yields from humates (Chen *et al.*, 1977).

The objective of this study was to investigate the formation of THMs during chlorination in the presence of different organic precursors, including algal organic matter (AOM) and humic acid (HA) under different reaction conditions. THMs were selected as a representative to chlorinated DBPs in this study because they usually the major DBPs formed during chlorination step.

## METHODS AND MATERIALS

### *Algae cultivation procedure*

One of the dominant green algal species, *Scenedesmus obliquus* (*S. obliquus*) were isolated from phytoplankton community of Nile River water, purified and recultivated using BG11 media. *S. obliquus* was in the logarithmic phase of growth when introduced to the standard algal culture medium. Algal cultures were grown and incubated at  $24 \pm 2^\circ\text{C}$  under continuous illumination (2,500 Lux) without aeration. The cultures were shaken once daily to prevent clumping and adherence of the algal cells to the containers. Cultures were harvested during the log growth phase for use in the experiments. Stock cultures of *S. obliquus* cells were collected by centrifugation at 4000 rpm for 5 min. The amount of algae inoculated in each experiment was determined at the beginning of the experiments using chlorophyll (a) measurement according to APHA (1998).

### *Chlorination*

The stock chlorine solution was prepared by bubbling of a pure chlorine gas (> 98 %) through de-ionized water and its concentration was determined by iodometric method whereas the chlorine residues after experiments were measured by the DPD colorimetric method according to (APHA, 1998). The chlorination experiments were conducted in a series of 1 L screw cap glass bottles with no head-space. The bottles were pre-soaked in 10% sulfuric acid for 24 h and rinsed with de-ionized water.

To prepare algal samples for experiment, chlorophyll "a" contents were determined. The volume of algal solutions were inoculated to the glass bottles and diluted by de-ionized water with media to produce 10, 20 and 30 µg/L of chlorophyll "a". On the other hand, the HA was purchased from Sigma Aldrich Company to act as NOM precursor. The stock solution of HA was prepared by dissolving 1000 mg of HA in 1 L purified water and diluted to obtain different concentrations (4, 8 and 12 mg/L). 5 mg/L Chlorine dosage was chosen to determine the formation of THMs. The formation of THMs was evaluated after reaction contact time of 5, 15, 30 and 60 minutes. A blank sample bottle was prepared with each batch of samples by the same manner but in absence of algal cells and humic acid. The potential impact of the growth medium in terms of DBPs formation was subtracted from that produced by algae and HA as it very low value.

### *THMs extraction and analysis*

Reference standard of the four trihalomethanes (THMs), including chloroform ( $\text{CHCl}_3$ ), bromodichloromethane ( $\text{CHCl}_2\text{Br}$ ), chlorodibromomethane ( $\text{CHClBr}_2$ ), and bromoform ( $\text{CHBr}_3$ ) were processed using the Standard Method (APHA, 1998). Briefly, 12 ml sample was pipetted into a 25 ml bottle with a round Teflon lined stopper, and 3 ml of pentane was then added to the bottle. The bottles were tightly sealed and shaken well and inverted up down for 4 min. The pentane extract was then transferred to a 1.5 ml vial. A GC-ECD system consisting of an Agilent 6890 gas chromatograph with a GC column (HP-5- 30 m long, 0.25 µm internal diameters, 0.25 µm coating) and Electron capture detector were used for THM determination. The oven temperature was programmed as follows: an initial temperature of  $50^\circ\text{C}$  was held for 2 min, and then increased at a rate of  $7.5^\circ\text{C}/\text{min}$  to  $105^\circ\text{C}$ . The temperature of the injector was set at  $200^\circ\text{C}$ . Nitrogen was used as the carrier gas. The detector temperature was set at  $300^\circ\text{C}$ .

## RESULT AND DISCUSSION

To study the effect of algal content on the total trihalomethanes (TTHMs) formation, several experiments of chlorination for water containing various amounts of algae (expressed as Chlorophyll (a) content) ranged from 10-30  $\mu\text{g/L}$  were conducted. Figure 1 revealed that TTHMs concentrations produced by algal cells increases with increasing chlorination time and reached maximum within 60 min. Also, it was worthy noted that the formation of TTHMs is highly affected by increasing the concentration of the algal cells in water samples. These results are in agreement with previous study by (Bukaveckas *et al.*, 2007 ) who suggests that a high level of DBP formation can be expected during the algae blooming season.

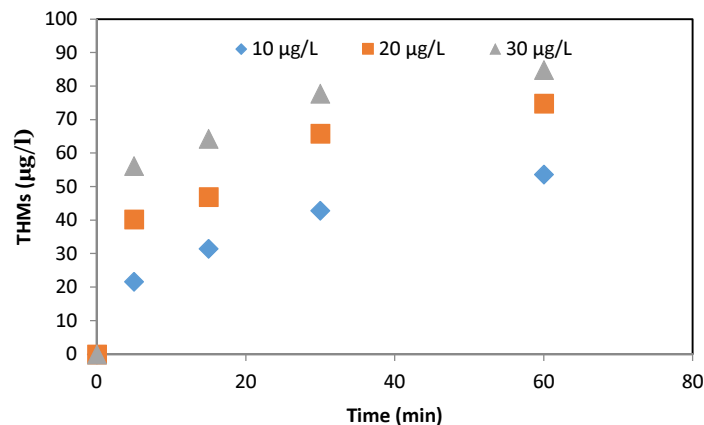


Figure 1. TTHMs formation for *S. obliquus* upon chlorination at pH =7

Figure 2 displays the average values of specific THMs yields during chlorination of different concentration of algal cells. The specific yield of TTHMs produced from chlorination of algal cells (10  $\mu\text{g/L}$  Chl (a)) was increased from 2.16 to 5.39  $\mu\text{g}/\mu\text{g}$  Chl (a) by increasing the contact time, while slight increase of yield of THMs produced from algal cell chlorination (20, 30  $\mu\text{g/L}$  Chl (a)) was observed. The obtained results showed that the THMs formation was very rapid, where most of THMs were formed after 15 min of reaction time. (Hong *et al.*, 2008) studied the formation of trihalomethanes upon chlorinating of algal cells, they found that green algae showed higher yield of chloroform (34  $\mu\text{g}/\text{mg}$  C). Also they add that algal cell is considered as important precursors of THMs formation. Thus, it is mandatory to remove algae physically without rupture the cells from water treatment to reduce THMs formation.

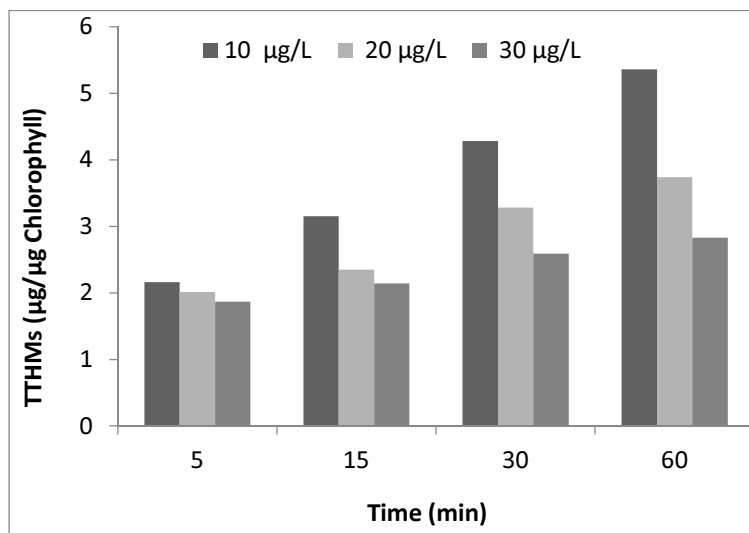
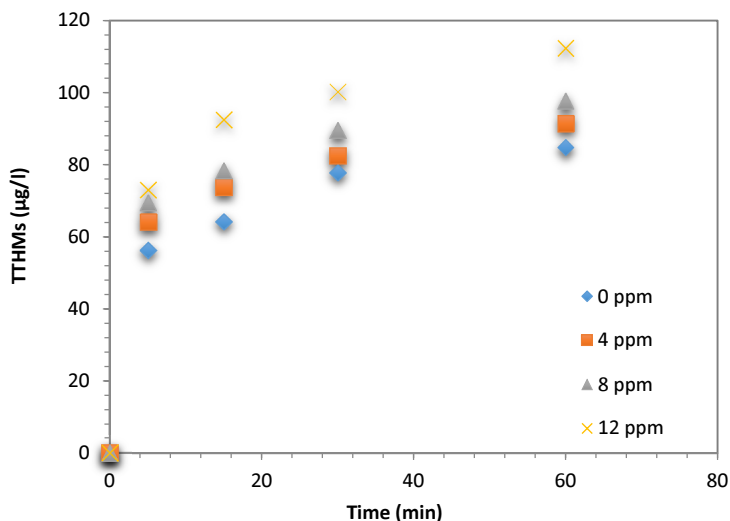


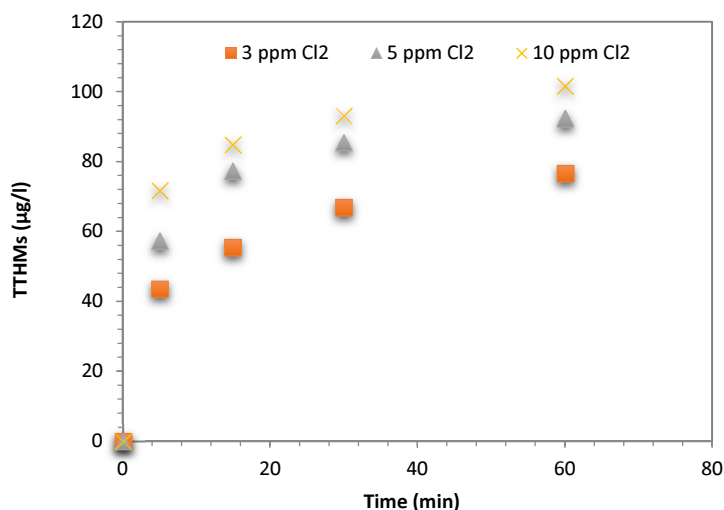
Figure 2. Specific total THMs after chlorination of water containing different concentrations of *S. obliquus*.

TTHMs formation in presence of algal cells (10  $\mu\text{g/L}$  as Chlorophyll (a) concentration) and various amounts of HA (4-12 mg/L) was assessed to explore the effect of co-presence of the two DBPs precursors. The obtained results of TTHMs concentration at different concentrations of HA are presented in Figure 3. TTHMs formation levels were slightly increased with the concentration of HA. As, the increase in HA concentration from 4 to 12 mg/L results in an increment of TTHMs level from 91.4 to 112  $\mu\text{g/L}$ . Therefore, it can be concluded that the TTHMs formation is more dependent on the algal cell amount rather than HA.



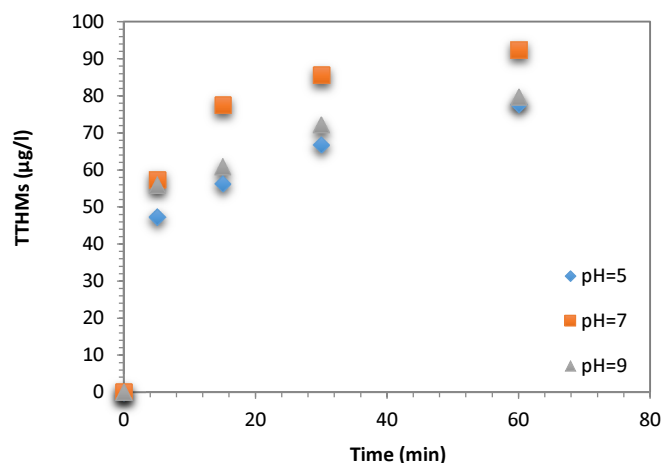
**Figure 3. Effect of humic acid concentration on TTHMs formation in presence of *S. obliquus* cells (10  $\mu\text{g/L}$ ).**

To estimate the dependence of THMs formation on chlorine dose used in chlorination, one liter of distilled water, containing 8 mg/L of HA and 10  $\mu\text{g/L}$  of algal cells, was chlorinated with different dosages of  $\text{Cl}_2$ . The profile of TTHMs formation with time is depicted in Figure 4. It was observed that there is an increase in produced TTHMs with the increment of chlorine dose. This might be due to higher probability of chlorine interaction with the available NOM.



**Figure 4. Effect of different chlorine doses on TTHMs formation as a function of algal cells and HA.**

pH of water has an influence on the formation of THMs, thus the effect of pH on the production of THMs was investigated in the pH range from 5 to 9 using constant initial concentration of chlorine. It can be seen from Figure 5 that TTHMs formation levels relatively increase as the pH values increased from 5 to 7. Significantly higher total concentration of THMs was obtained at pH = 7 (THMs = 92.5  $\mu\text{g/L}$ ) than that at pH = 5 (THMs = 77.8  $\mu\text{g/L}$ ). A slight decrease in total THMs concentration for pH = 9 (14 %) was recorded compared with that at pH = 7 due to the fact that the produced halogenated THMs hydrolyze and decrease at high pH values (Krasner *et al.*, 1989).



**Figure 5. Effect of pH on disinfection by-products (DBPs) as a function of algal cells and HA.**

### Conclusion

The influence of co-presence of humic acid and algae on the formation of THMs has been investigated under several conditions. The formation of THMs was proved to be positively affected by chlorination dose and time and concentrations of both algae and humic acid. It was also noticed that the effect of algae on the THMs formation is more pronouncing than that of HA. Therefore, it is recommended to spend greater efforts in removing algae from water rather than using sophisticated techniques to remove humic acid from raw water.

### REFERENCES

- Amy G.L., J. Sierka, D.P. Bedessem, L. Tan, 1992. Molecular size distribution of dissolved organic matter, *J. Am. Water Works Assoc.*, 84:67-75.
- APHA (American Puplic Health Association), AWWA, WEF, 1998. *Standard Methods for the Examination of Water and Wastewater*, Twentieth ed., Washington, DC.
- Boyer, T.H., P.C. Singer, G.R. Aiken, 2008. Removal of dissolved organic matter by anion exchange: effect of dissolved organic matter properties, *Environ. Sci. Technol.* 42:7431-7437.
- Bukaveckas, P. A., D. McGaha, J.M. Shostell, R. Schultz, J.D. Jack., 2007. Internal and external sources of THM precursors in a midwestern reservoir, *J. Am. Water Works Assoc.*, 127–136.
- Chen, Y., N. Senesi, M. Schnitzer, 1977. Information provided on humic substances by E4/E6 ratios. *Soil Science Society of America Journal*, 352-358.
- Cheng, W.P., F.H. Chi, 2003. Influence of eutrophication on the coagulation efficiency in reservoir water, *Chemosphere*, 53: 773-778.
- Croue, J.P., J.F. DeBroux, G.L. Amy, G. Aiken, J. Leenheer, 2000. Natural organic matter-structural characteristics and reactive properties, *J. Am. Wks. Assoc.*, 65-93.
- EPA 2014. Basic Information about Disinfection Byproducts in Drinking Water: Total Trihalomethanes, Haloacetic Acids, Bromate, and Chlorite. Environmental Protection Agency.
- Fang, J., X. Ma, C. Yang, C. Shang, 2010. Formation of carbonaceous and nitrogenous disinfection by-products from the chlorination of *Microcystis aeruginosa*, *Water Res.*, 44:1934-1940.
- Henderson, R.K., A. Baker, S.A. Parsons, B. Jefferson, 2008. Characterisation of algogenic organic matter extracted from cyanobacteria, green algae and diatoms. *Water Research*, 3435–3445.

- Hong, H.C., A. Mazumder, M.H. Wong, Y. Liang, 2008. Yield of trihalomethanes and haloacetic acids upon chlorinating algal cells, and its prediction via algal cellular biochemical composition, *Water Research*, 4941–4948.
- Hua, G.H., D.A. Reckhow, 2007. Characterization of disinfection byproduct precursors based on hydrophobicity and molecular size, *Environ. Sci. Technol.*, 41:3309 -3315.
- Krasner, S. W., M.J. McGuire, J.J. Jacangelo, 1989. The occurrence of disinfection by-products in U.S. drinking water. *J. Am. Water Works Assoc.*, 41-53.
- Ma, J., W. Liu, 2002. Effectiveness and mechanism of potassium ferrate(VI) preoxidation for algae removal by coagulation, *Water Res.*, 871–878.
- Nie, Y.L., C. Hu, L. Zhou, J.H. Qu, Q.S. Wei, D.S. Wang, 2010. Degradation characteristics of humic acid over iron oxides/FeO core-shell nanoparticles with UVA/H<sub>2</sub>O<sub>2</sub>, *Journal of Hazardous Materials*, 474-479.
- Plummer, J.D., J.K. Edzwald, 2002. Effects of chlorine and ozone on algal cell properties and removal of algae by coagulation, *Journal of Water Supply Research and Technology-Aqua*, 307–318.
- Richardson, S., A. Thruston Jr, T. Caughran, P. Chen, T. Collette, K. Schenck, B. Lykins Jr, C. Rav-Acha, V. Glezer, 2000. Identification of new drinking water disinfection by-products from ozone, chlorine dioxide, chloramine, and chlorine, *Water Air Soil Pollut.*, 95-102.
- Rook, J.J., 1974. Formation of haloforms during chlorination of natural waters, *Water Treat. Exam.*, 23: 234-242.
- Rosario-Ortiz, F.L., 2013. Singlet oxygen formation from wastewater organic matter, *Environmental Science and Technology*, 8179-8186.
- Singer, P.C., 1994. Control of disinfection by-products in drinking water, *J. Environ. Eng.*, 727-774.
- Takaara, T., D. Sano, H. Konno, T. Omura, 2007. Cellular proteins of *Microcystis aeruginosa* inhibiting coagulation with polyaluminum chloride, *Water Research*, 1653–1658.
- Xue, G., H.H. Liu, Q.Y. Chen, C. Hills, M. Tyrer, F. Innocent, 2011. Synergy between surface adsorption and photocatalysis during degradation of humic acid on TiO<sub>2</sub>/activated carbon composites, *Journal of Hazardous Materials*, 765-772.