

The impact of high alkali media on the biochemical compositions of two hydrophytes; *Lemna minor* L. and *Ceratophyllum demersum* L. from Shatt Al-Arab River, Basrah-Iraq

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Abstract - The present work aims at understanding the effect of high alkali media on the biochemical composition of two hydrophytes; *Lemna minor* L. and *Ceratophyllum demersum* L. Each of these two hydrophytes was planted in 4-one liter glass containers of pH 7.5, 8.5, 9.5 and 10.5 under laboratory conditions. Samples were taken after two weeks for physico-chemical parameters and tissue (protein, fat and ash) analysis. The results show that the highest protein contents (34.81 %, 34.66 %) were found in the control medium of *L. minor*, in *C. demersum* at pH 7.5. The least protein content was 21.45% in *L.minor* at pH 9.5. As to the fat contents the highest value (7.50 %) was recorded in *L. minor* at pH 9.5, while the least one was 1.181% in *L. minor* at pH 10.5. The highest ash content of *C. demersum* was 19.88 % at pH 10.5 and the least was 11.93% at the control. High pH showed negative impact on protein content of the two aquatic plants. The protein was reduced as the pH value elevates, but at pH 10.5 the protein content rised again. As for fat content, increasing pH caused increasing of fat in *L. minor* and its decreasing in *C. demersum* except pH 10.5 at which the fat comes up again in reverse to the former plant. The ash content of *L. minor* was fluctuating, whereas in *C. denersum*, the increasing of pH value caused increasing of ash content.

Keywords: High pH, biochemical compositions, *Lemna minor*, *Ceratophyllum demersum*, Shatt Al-Arab River.

Introduction

The term "pH" is a mathematical transformation of the hydrogen ion [H⁺] concentration; it conveniently expresses the acidity or alkalinity of water, the lower case letter "p" refers to "power" or exponent, and pH is defined as the negative logarithm of the hydrogen ion concentration, each change of one pH unit represents a ten-fold change in hydrogen ion concentration (Tucker and D'Abramo, 2008). The pH scale is usually ranging from 0 to 14, but pH can extend beyond those values, so at 25°C, pH 7.0 describes the neutral point of water at which the concentrations of hydrogen and hydroxyl ions (OH⁻) are equal (each at 10⁻⁷ moles/l) so conditions become more acidic as pH decreases and more basic as pH increases (Tucker and D'Abramo, 2008). The pH of freshwater ecosystems can fluctuate considerably within daily and seasonal timeframes, and most freshwater

animals have evolved to tolerate a relatively wide environmental pH range but animals can, however, become stressed or die when exposed to pH extremes or when pH changes rapidly, even if the change occurs within a pH range that is normally tolerated (Tucker and D'Abramo, 2008). In addition to the direct effects of pH on aquatic animals, the hydrogen ion concentration affects aqueous equilibria involving ammonia, hydrogen sulfide, chlorine and dissolved metals and the interactions of pH with these variables are often more important than the direct effects of pH on aquatic animals, so direct "pH toxicity" is relatively rare in aquaculture ponds because farm sites and water supplies are selected to provide a desirable environment for culture, which should include a pH of approximately 6 to 9 (Tucker and D'Abramo, 2008). Aquatic plants are essential source of energy and nutrient for other trophic levels (Mann, 1988) some vertebrates feed on living plants others on dead and/or decomposed. Ali and Abdullah (1997) evaluate the energy values, organic and ash contents of 14 species of aquatic plants from Shatt Al-Arab habitat. Talal (2009) studied the impact of low acidity media on two hydrophytes; *Salvinia natans* L. and *Ceratophyllum demersum* L., and found that acidic media could cause retardation of growth of the hydrophytes and pH 7 is quite fit for the hydrophytes. Alabaster and Lloyd (1980) stated that chronic exposure to pH values above 10 was harmful to all species studied, while salmonids and some other species were harmed at pH values above 9.

The USEPA (1976, 1986) has concluded that a pH range of 6.5 to 9.0 provides adequate protection for the life of freshwater fish and bottom-dwelling macro invertebrates, outside this range, fish suffer adverse physiological effects that increase in severity as the degree of deviation increases until lethal levels are reached. One of the primary goals of aquatic weed management in public and private waters is to control growth of invasive plant species while maintaining a diversity of native submersed and emergent species. Native aquatic plants can improve water clarity and quality, provide valuable fish and wildlife habitat, reduce sediment re-suspension, and help prevent the spread of invasive plants (Savino and Stein, 1982; Heitmeyer and Vohs, 1984; Smart, 1995; Dibble *et al.*, 1996). The aim of the present work is to demonstrate the effects of various pH media on the chemical composition of the two hydrophytes *Lemna minor* and in *Ceratophyllum demersum* from Shatt Al-Arab River, which was not conducted before here in Basrah.

Materials and Methods

The experiment was carried out to understand the effect of high alkali media on the tissue compositions of two hydrophytes; *L. minor* and *C. demersum*. Each of these two hydrophytes was planted in 4 glass containers of 1 L capacity each and with pH of 7.5, 8.5, 9.5 and 10.5 under laboratory conditions. Samples were taken after two week from the water (physico-chemical) and tissue (protein, fat and ash) for analysis. T-test was used for analyses of the data.

A- Experiment preparations:

- 1- Hydrophytes species: Two local species of aquatic plants were selected from Shatt Al-Arab River these are *L. minor* and *C. demersum*.

- 2- Containers: glass containers of one liter each were used for culturing each species. These containers contained cleaned dechlorinated tap water.
- 3- Culturing process: Each species of the two plants were cultivated in a set of four duplicated containers adjusted for pH 7.5, 8.5, 9.5 and 10.5 treatments with one as a control. The measurements were taken after fifteen days of the starting experiment.
- 4- Sampling analysis: Sampling was carried out for physical, chemical analysis before the experiment while tissue analyses were carried out at the end of the experiment.

B- Water Analysis:

The physical-chemical analyses of water; salinity, pH, and EC were determined using a digital laboratory salinometer (type Kent Lie 5005, USA instrument) and multimeter YSI-Incorporated model-556MPS apparatus. Temperatures were measured *in situ*, using a mercury thermometer, to the nearest 0.1°C.

C- Plant Tissue Analyses:

At the end of the experiment, the two treated plants were taken out from the containers, air or oven dried for overnight then grained and stored in cool and dried place for further tissue analyses.

- 1- Ash contents: The ash content was estimated by burning the plant tissues by Muffle furnace at 550°C (A.O.A.C., 1984).
- 2- Protein contents: Nitrogen was estimated by semi-micro Kjeldahl method (Pearson, 1970), and multiplied by 6.25 to get the protein contents.
- 3- Fat contents: The fat content was determined by (A.O.A.C., 1984) by using Soxhelt instrument.

Results

The physico-chemical characteristics of water used for maintaining *Lemna minor* L. and *Ceratophyllum demersum* L. were listed in Tables (1 and 2). The percentages of protein, fat and ash contents for both aquatic plants were represented in Figures (1 - 5).

The T-test indicates that there was a significant difference among ash contents and so were the difference among fat and protein contents at 0.05 level.

Table 1. The physico-chemical characteristics of water media used for rearing of *L. minor*.

Treatment	DO (mg/l)	Salinity (‰)	C°
<i>Lemna minor</i> before the experiment	6.5	3.7	27.0
<i>Lemna minor</i> in control medium	6.0	3.0	27.0
pH 7.5	6.5	4.5	27.2
pH 8.5	4.5	4.4	27.0
pH 9.5	5.0	4.0	27.0
pH 10.5	5.0	4.3	27.0

Table 2. The physico-chemical characteristics of water media used for rearing *C. demersum*.

Treatment	DO (mg/l)	Salinity (‰)	C°
<i>C. demersum</i> before the experiment	6.5	3.0	27.0
<i>C. demersum</i> in control medium	5.0	3.0	27.0
pH 7.5	6.5	4.4	27.2
pH 8.5	4.5	4.5	27.0
pH 9.5	5.0	3.8	27.0
pH 10.5	5.0	4.5	27.0

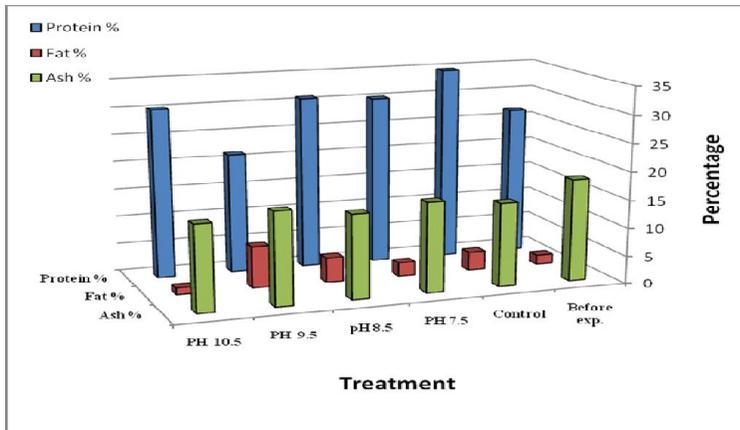


Figure 1. Percentages of protein, fat and ash of *L. minor*.

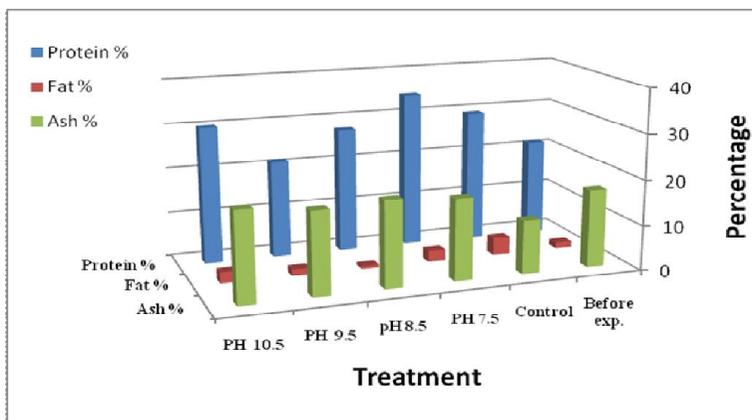


Figure 2. Percentages of protein, fat and ash of *C. demersum*.

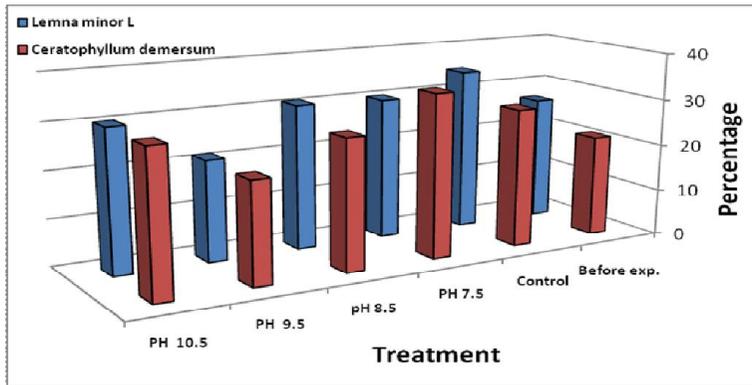


Figure 3. Comparison of protein contents of *L. minor* and *C. demersum*.

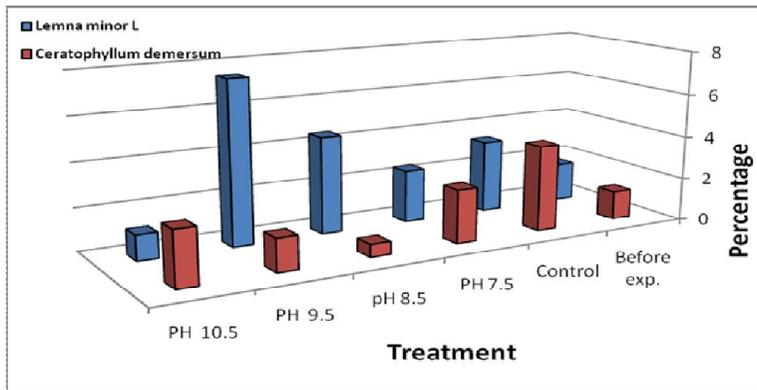


Figure 4. Comparison of fat contents of *L. minor* and *C. demersum*.

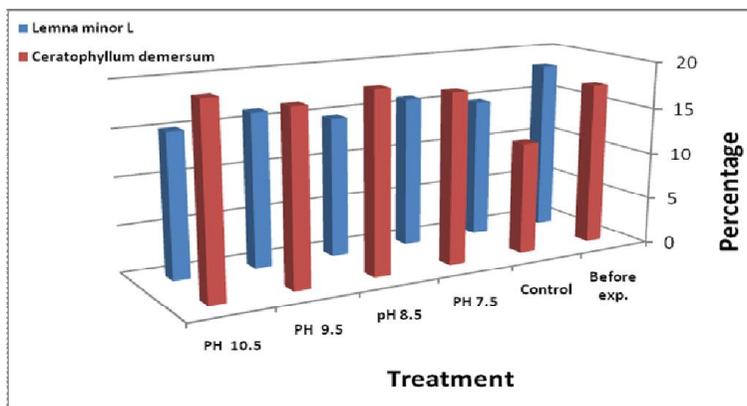


Figure 5. Comparison of ash contents of *L. minor* and *C. demersum*.

The results indicate that the highest protein contents were in the control of *L. minor* 34.81% and 34.66% in *C. demersum* at pH 7.5 aquarium. The least protein content was 21.45% at pH 9.5 in *L. minor*. As to the fat content the highest value 7.50% was at pH 9.5 in *L. minor*, the least was 1.1808% at pH 10.5 in *L. minor*. While the highest ash content was 19.88% at pH 10.5 in *C. demersum* and the least was 11.93% at the control of *C. demersum*. High pH showed negative impact on protein content of the two aquatic plants. The protein was reduced as pH value elevates, but at pH 10.5 the protein contents rose up again. As for fat content, increasing pH caused increasing of fat in *L. minor* except at pH 10.5 but it was decreased in *C. demersum* except at pH 10.5 at which the fat comes up again. The ash content of *L. minor* was inconsistent, while in case of *C. demersum*, increasing of pH value caused increasing of ash contents.

Discussion

High pH values caused low fluctuation in DO concentration (Tables 1 and 2). Although the pH had no apparent effect on the ash contents of *L. minor*, yet increasing pH value caused an increase of ash contents in *C. demersum*, possibly increasing of pH value could cause more degradation of tissue components. A marked decrease of protein contents in both plants with increasing pH had been detected, since it is a growth limiting factor (Guo *et al.*, 2011). Increasing of fat contents with increasing pH was observed in *L. minor* whereas in *C. demersum* the fat contents decreased with increasing pH, the fat content difference between the two plants could be due to that *L. minor* was already found in an environment of nutrients was richer than that of *C. demersum*.

The photosynthetic activity of 15 species of microalgae and macrophytes was measured by continuous recording of oxygen concentration but the microalgae had considerably greater apparent affinities for HCO_3^- and slightly greater apparent affinity for CO_2 than the macrophytes, while the macrophytes had larger apparent affinities for CO_2 than for HCO_3^- , assimilation rate in a solution of constant alkalinity showed a distinct reduction when the CO_2 concentration decreased to a value typical of each species (Allen and Spence, 1981). Saline stress generally involves osmotic stress and ion injury, but alkaline stress not only included osmotic stress and ion injury, but also has high pH effect (Yang *et al.*, 2009). The high pH environment surrounding the roots can directly cause mineral elements availability significantly decreased and Ca^{+2} , Mg^{+2} and HPO_4^{-3} to precipitate, which possibly inhibit ion uptake and disrupt the ion homeostasis of plant cells (Shi and Wang, 2005; Xue and Liu, 2008). Growth of giant salvinia (*Salvinia molesta* Mitchell) under different pH ranges was examined at the Lewisville Aquatic Ecosystem Research Facility (LAERF) in Lewisville, Texas, this giant salvinia grew to completely cover a research pond over 15 week period when pH was less than 7.5, but the growth was reduced in a second pond maintained at a higher pH ranging from 8.5-10.0 (Owens *et al.*, 2005). In the absence of processes that add or remove carbon dioxide, the initial pH of water in contact with air depends on its alkalinity, waters with low alkalinities have an initial pH at the low end of that range, while waters of higher alkalinities have higher pH and during daylight, algae and

under water plants remove carbon dioxide from the water as part of the sunlight driven process of photosynthesis, and relative respiration rates are affected by water temperature and the biomass of plants, animals and microorganisms in the water and bottom sediment and rates of photosynthesis are controlled primarily by sunlight intensity, plant biomass and water temperature so the daily interplay of respiration and photosynthesis causes pH to cycle up and down during a 24 hour period so however, when plants or algae are growing rapidly, more carbon dioxide is removed each day by photosynthesis than is added each night by respirations, as result, pH may rise to abnormally high levels during the afternoon and may even remain high through the night, this condition may last for many days, until photosynthesis decreases or respiration increases (Guo *et al.*, 2011).

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تأثير الأوساط القاعدية المرتفعة على التركيب الكيميائي الحيوي للنباتين المائيين عدس الماء *Lemna minor* L. والشمبلان *Ceratophyllum demersum* L. من نهر شط العرب ، البصرة ، العراق

خديجة كاظم حريب

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المستخلص - هدفت التجربة الى معرفة تأثير الأوساط مرتفعة القاعدية على التركيب الكيميائي لأنسجة النباتين المائيين عدس الماء *Lemna minor* L. والشمبلان *Ceratophyllum demersum* L. إستزرع هذان النباتان في الأوساط القاعدية 7.5 و 8.5 و 9.5 و 10.5 تحت ظروف مختبرية. أخذت العينات بعد خمسة عشر يوماً وأجريت التحاليل الفيزيائية والكيميائية على الأوساط القاعدية والأنسجة (لتقدير محتوى البروتين والدهن والرماد). أظهرت النتائج بأن أعلى نسبة مئوية للبروتين كانت 34.81% في حوض السيطرة العائد لعدس الماء ثم تليها 34.66% في حوض pH 7.5 العائد للشمبلان وأن أدنى نسبة كانت 21.45% في حوض pH 9.5 العائد لعدس الماء. فيما يتعلق بالنسبة المئوية للدهن فأن أعلى نسبة له كانت 7.50% في حوض pH 9.5 العائد لعدس الماء وأن أدنى نسبة كانت 1.181 في حوض pH 10.5 العائد لعدس الماء بينما أعلى نسب مئوية للرماد كانت 19.88% في حوض pH 10.5 العائد للشمبلان وأن أدنى نسبة كانت 11.93 في حوض معاملة السيطرة العائد للشمبلان، أظهرت النتائج بأن pH المرتفع له تأثير سلبي على محتوى البروتين لكلا النباتين، حيث إنخفض بإرتفاع قيمة pH ولكنه إرتفع ثانية عند pH 10.5. وفيما يتعلق بمحتوى الدهن فأن إزدياد pH سبب إزدياد الدهن في عدس الماء وانخفاضه في الشمبلان ماعدا في pH 10.5 فأن الدهن عاد للإرتفاع ثانية وعلى عكس النبات الأول. أما الرماد فقد تنذبذب محتواه في عدس الماء في جميع الأحواض في حين أن إزدياد pH سبب ارتفاع محتوى الرماد في الشمبلان.