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Recent updates on ions and nutrients uptake by halotolerant freshwater and marine microalgae in conditions of high salinity

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ABSTRACT

Algae is an appropriate natural resource to augment the optimal use of undesired ions in water and wastewater. Increasing algal cells, the consumption of particular ions, including chloride, nitrate, phosphate, and ammonium, provides a suitable way to optimize water treatment processes. Different algal species have the capability to survive in extreme salinities by developing resistance against osmotic pressure in saline water. The current study reviews the effect of salinity on algal biomass production, algal growth rate, chlorides, nitrates, phosphates, chemical oxygen demand (COD), total nitrogen, total phosphorus, and ammonium ions. Mainly algae cultivated in freshwater, synthetic brackish water, seawater, and hypersaline water, were studied for this review. Various ion uptake mechanisms used by the algal cell are summarized, focusing on biosorption and bioaccumulation processes. Critical parameters such as light intensity, pH, and temperature variations significantly influence ion and nutrients uptake efficiencies. Analysis performed on collected data indicated that halophytic algae could survive in high salinities at elevated growth rates compared to freshwater. The halotolerant algal species showed an inclining trend of chloride ion removal with an elimination capacity of 7.5 $g.m^{-3}.h^{-1}$. Moreover, the nitrate uptake rate in halophytic algae is 10-folds higher to phosphate, regardless of salinity level. It could be concluded that microalgae will be beneficial for ion and nutrient uptake processes in treating high saline water.

1. Introduction

Many countries have adapted to the lack of freshwater resources in coastal regions by treating and consuming seawater. Available technologies such as thermal desalination, reverse osmosis, and conventional methods have been extensively used for seawater treatment. Many researchers are currently exploring how to develop cost-effective treatment methods to reduce environmental stress and save energy. One novel and practical technique can be the engineered cultivation of algae, which takes up harmful ions from water bodies, such as chlorides, nitrogen, and phosphorus, but these ions are nutritional sources for algae. The engineered algal cultivation technique can supply harmful ions/nutrients to harvest algae, making the cultivation process more economical. Many countries in the Arabian Peninsula (Gulf region), Israel, and the USA use reverse osmosis plants to treat seawater for domestic purposes. This treatment process usually reduces the concentration of chloride ions in the influent. The desalination technique is energy-intensive, and treated water cost becomes high. The World Health Organization guideline for chloride ion concentrations in drinking water is up to 250 $mg.L^{-1}$ [1]. Chloride ion concentration prevailing in seawater requires removal efficiency (RE) of approximately 99.28% to provide drinking water for consumers.

Abbreviations: ATP, adenosine triphosphate; COD, Chemical oxygen demand; EPS, Extracellular polymeric substances; NADP, Nicotinamide adenine phosphate; RE, Removal efficiency; TDS, Total dissolved solids; *S. sp.*, *Scenedesmus species*; *N. salina*, *Nannochloropsis salina*; *C. vulgaris*, *Chlorella vulgaris*; *C. sorokiniana*, *Chlorella sorokiniana*.

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The general definition of salinity is the presence of at least 35 $g.L^{-1}$ or parts per thousand (ppt) of dissolved salts in water [2]. The chloride ion is the major contributor to salinity. Salinity has been further classified into different water categories based on the chloride ion concentration. For example, the chloride ion concentration ranges from 30 to 150 $mg.L^{-1}$ in freshwater, 300 to 1000 $mg.L^{-1}$ in brackish water, and 30,000 to 35,000 $mg.L^{-1}$ in seawater [3]. Algae can be grown in laboratories by providing essential nutrients and saline conditions. The property of algae to consume specific ions under different salinities can be used in reactors to capitalize on their ion and nutrient uptake capability. Moreover, the uptake of ions by algae for growth under the influence of salinity can be utilized for water treatment through, for example, desalination.

Algae grow naturally at various salinities due to their osmotic pressure-tolerating capacity [4]. Both halotolerant freshwater algae and marine algae can survive at high salinities. The most common halotolerant freshwater algae are *Chlorella vulgaris*, *Scenedesmus obliquus*, and *Chlorella sorokiniana*, which are reported to tolerate salinity of up to 11 $g.L^{-1}$ [5–7]. In particular, *C. vulgaris* and *S. obliquus* exhibit even higher tolerance to salinity [8,9]. Gani et al. [10] evaluated the effect of temperature, photoperiod, light intensity, and salinity on *Botryococcus* sp. They found that the freshwater green algae showed optimum growth at salinities up to 35 $g.L^{-1}$ and at 33 °C temperature. Although the growth of these halotolerant freshwater microalgae is inhibited above a certain level of salinity [11,12], these studies show that halotolerant freshwater algae can survive under conditions of salt stress. However, the marine alga *Platymonas subcordiformis* showed higher growth at ~30 $g.L^{-1}$ NaCl [13], and other marine species, such as *Nannochloropsis* sp., *N. salina*, and *Dunaliella tertiolecta*, showed growth at salinities of 35–48 ppt [14–16]. Marine algae require a high-salinity environment for growth [17–19], as reported mainly for *Dunaliella* sp. [4,17]. Algae can grow at higher salinities depending upon the water type and algal species. Along with salinity, algae require a suitable environment for growth. The main requirements are appropriate growth medium, temperature [20], light intensity [21–24], pH range [25–27], and supply of air and CO_2 [28]. Although these conditions do not guarantee perfect growth, the conditions required to cultivate algae at a laboratory scale have been used widely for different applications.

Algae generate valuable products such as biodiesel production, lipids, fatty acids extractions, food supplements, and protein extraction [29]. However, to the authors' best knowledge, there was no previous review published in the ion and nutrient uptake by the algae in the presence of high salinity. Therefore, this study was formulated based on algae consuming up ions and nutrients in high salinity and discussed the effect on algae due to different salinities. The ions and nutrients involved in this review correspond to the chloride, nitrate, phosphate, chemical oxygen demand, total nitrogen, total phosphorus, and ammonium ions. Chloride is an essential parameter studied in detail for desalination treatment plants because it contributes to the salinity [30]. Therefore, there is a great need for or an up-to-date review to compile recent research advances regarding ions and nutrients uptake by microalgae in the influence of salinity.

This review focuses on ions and nutrients uptake by algae cultivated in different salinities using synthetic water brackish water, freshwater, wastewater, and seawater. The review also discusses sewage, which has been reported to have very high salinity, such as produced wastewater. For example, wastewater produced from oil and shale gas sites tends to be very saline. This review also compares halotolerant freshwater and marine algae and analyzes their optimal algal growth rate under different salinities. The salinity range selected in this review has intervals of 5 ppt, starting from 0, 2.5, and 7.5, and continuing to 50 ppt. Generally, the salinity data reviewed herein terminate at 32.5 ppt as few studies have assessed salinity beyond 32.5 ppt. In this review, ion uptake is reported in terms of elimination capacity (units: $g.m^{-3}.h^{-1}$). Elimination capacity is used as the performance indicator and as a tool to design a water treatment plant without validating the microbial quantity and activity [31]. Elimination capacity estimates the load of the microbial community that could consume the contaminant or organic matter in most of the bioreactors. Therefore, elimination capacity becomes an important parameter to measure for water/wastewater treatment plants. The overall contribution of halotolerant freshwater and marine algae to ion uptake has been analyzed. As the number of marine algae studies is limited, it was difficult to compare marine algae with halotolerant freshwater algae in terms of ions and nutrients uptake. However, this review presents a detailed analysis and discussion of how algae take up ions under conditions of high salinity.

2. Methodology and discussion on essential parameters for algae

Data for this review were collected from experimental studies conducted over the past decade, i.e., from January 2010 to August 2021, as

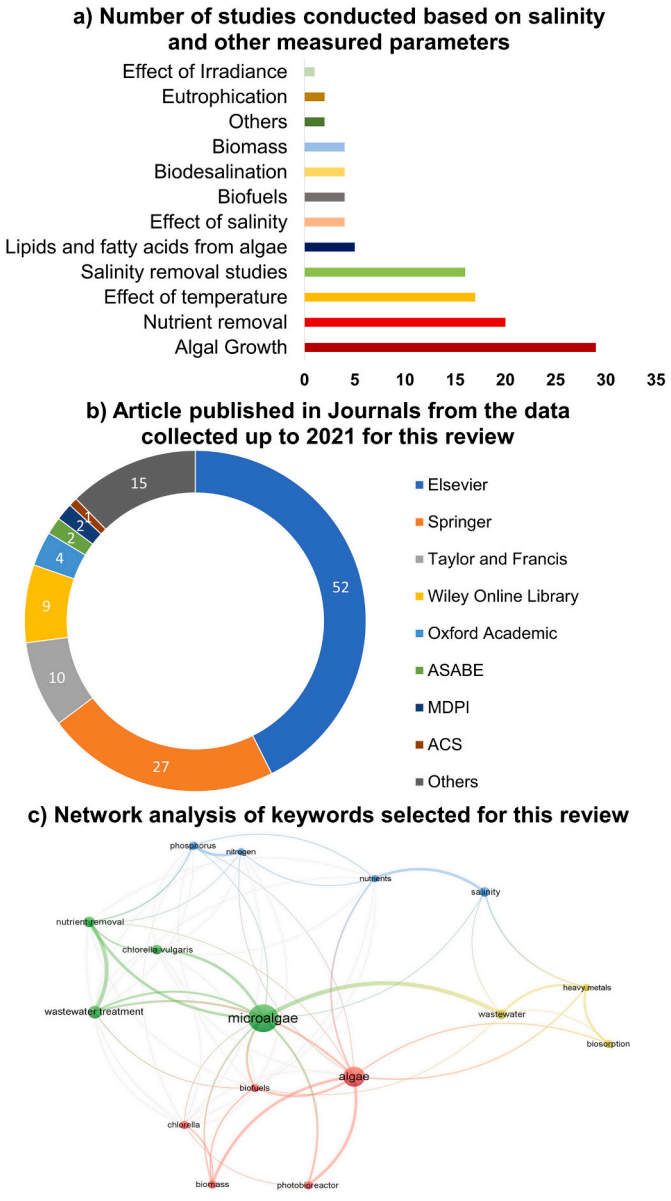

Fig. 1. Article analysis from the literature collected for this review, a) analysis of different studies conducted on the algae in the presence of saline environment, b) overview of articles published in different journals up to the year 2021 focusing on salinity removal by algae, c) network analysis map of keywords selected for this review article.

shown in Fig. 1. We conducted a literature search using the Science Direct, Scopus, Google Scholar, and Web of Science databases by applying the keywords "effect on algal growth in a high saline environment," "salt stress on algae in high salinity," "ion uptake by algae," and "effect of high salinity on ion uptake by algae." Fig. 1a represents the topics collected from the articles obtained from the keywords above. The articles collected were based on the salinity in response to other measurements as well. However, most articles covered the domain of algal growth, nutrient removal, the effect of temperature, lipid, and fatty acids. Fig. 1b represents the statistics of collected articles that were issued under different journal publishers. Most articles collected for this review were published in Elsevier, Springer, Wiley Online Library, Taylor and Francis. Fig. 1c represents the network analysis map of keywords collected for the current review. Many articles cited in this review contain microalgae, algae, wastewater treatment, salinity, and nutrient removal keywords.

The data were collected and separated into algae and species grown in different types of water, such as marine water, freshwater, brackish

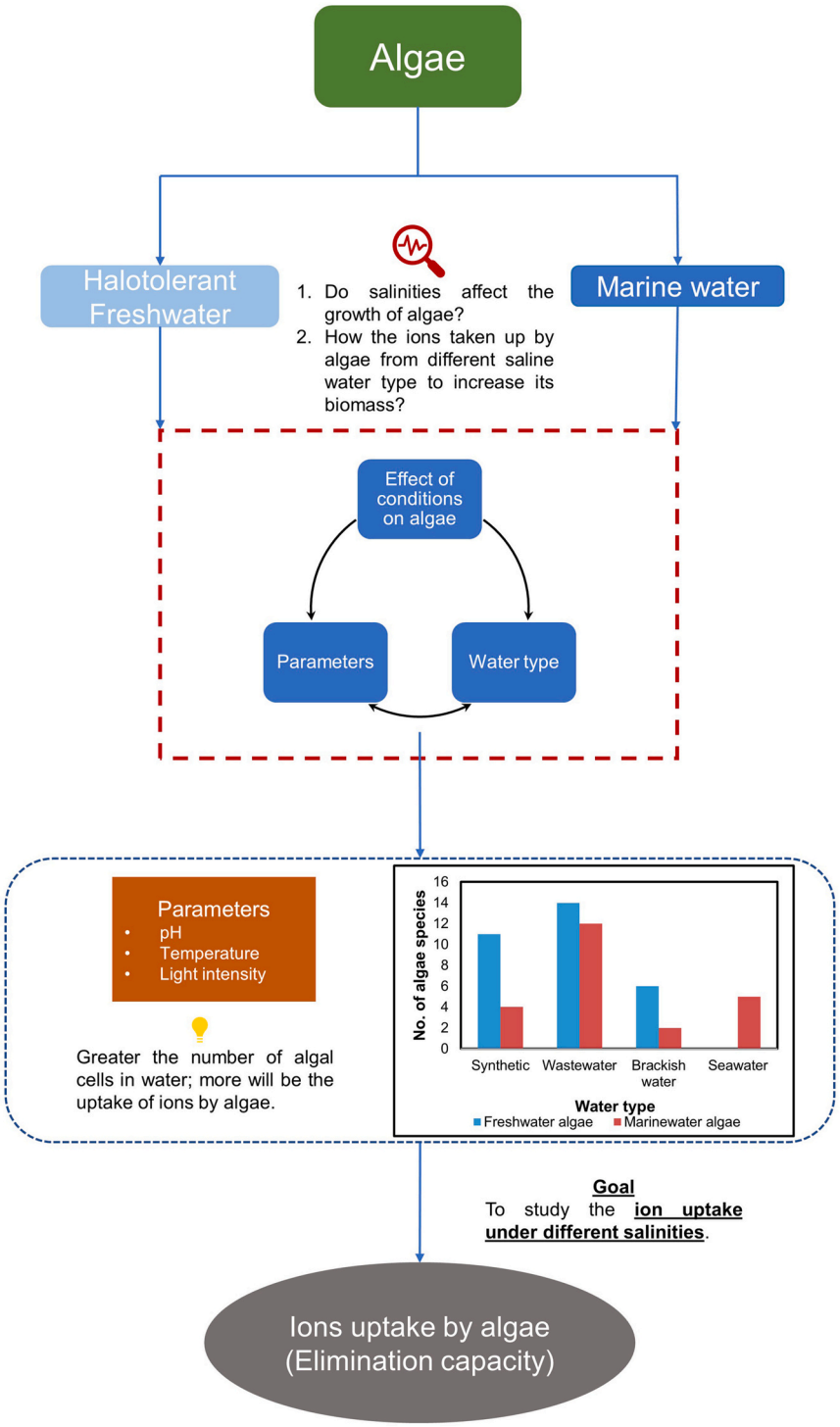

Fig. 2. A schematic layout of the scope of this review, indicating open research questions regarding the effect of salinity on algal growth and biomass; the figure describes the focus of this study, which is ion uptake by algae.

water, seawater, and wastewater with high salinity. The total 23 marine water species include *Nannochloropsis salina*, *Chattonella marina*, *Enteromorpha* sp. in different water types. Similarly, 21 freshwater algae species, including *Scenedesmus obliquus*, *Chlorella vulgaris*, *Desmodesmus armatus*, *Chlorella sorkiniana*, were studied for ion uptake. As the collected salinity data were irregular, we re-scaled the salinities at intervals of 5 ppt. All the collected ion data were averaged, and the standard deviation is presented in all figures from Section 4.

The theme and scope of this review study are shown in Fig. 2. The algae discussed in this review are halotolerant freshwater algae and marine algae. The following open research questions are addressed: 1) Does salinity affect algal growth? and 2) How does ion uptake by algae and increased biomass levels differ among different saline water types? The effects of environmental parameters such as pH, light intensity, and temperature were ignored in preparing the dataset; however, the type of water was used to classify the data in Microsoft Excel, as shown in Fig. 2. However, nutritional ions were examined under different salinities for this review. The focus of this review is based on the finding that the greater the number of algal cells produced in water, the greater is the uptake of ions/nutrients by algae. Thus, this review aimed to clarify the dynamics of ion/nutrients uptake by algae under different salinities and process conditions.

This review considers important environmental parameters such as pH, temperature, and light intensity and focuses on identifying the maximum growth in algae with respect to growth rate and biomass production. However, these parameters, except water type, were not considered in the analysis. However, the effect of pH, light intensity, and temperature on algal growth was discussed in this review briefly.

2.1. Environmental parameters that affect algal growth and ion uptake by algae

2.1.1. Effect of pH

pH plays a vital role in algal cell growth as it directly affects lipid accumulation by algae [32]. Algal lipid content remained stable at a pH range of 7.5–10 and was reduced by 9% with an increase in pH to 11 [33]. Higher alkalinity reduces the concentration of algal cells because, in alkaline conditions, algal cells have a greater demand for adenosine triphosphate (ATP) to transport bicarbonate and sodium ions through the cell membrane [34]. pH levels below 11 are suitable for algal cell growth. However, if CO_2 is injected into the reactors, the pH may decrease to below 6.5. As reported by Guckert and Cooksey [35], high alkaline stress reduces lipid accumulation by algae. In general, higher pH does not affect either cell density or cell growth rate [36,37]. At a pH of >9.5, the rate of photosynthesis in marine algae inhibit the rate of oxygen evolution. The level of bicarbonate ions increases at high pH. High pH levels restrict the penetration of ions into algal cells and reduce the rate of photosynthesis [26].

Injection of CO_2 into the solution made the pH more acidic, and therefore, at low pH, there was insufficient algal growth and cell density. Sahle-Demessie et al. [9] observed that CO_2 consumption increased with an increase in biomass, but a higher rate of CO_2 injection reduced the pH to the acidic scale because of carbonic acid (H_2CO_3) formation. In water treatment by microalgae, precipitation at a pH of 8–10 can remove ions such as phosphate, ammonium, and magnesium in the aqueous phase [38]. As reported by Larsdotter et al. [39], photosynthesizing algae increased the pH, primarily because of the formation of chemical precipitates of ammonium, phosphate, and carbonate ions.

2.1.2. Effect of temperature

Light, temperature, growth media, and salinity are essential parameters for the growth of marine algae [4]. Temperature is an important parameter for cultivating and achieving maximum output from algae under saline conditions. Algae are found in cold aquatic zones close to the North and South Poles and can survive at temperatures above 51 °C and in tropical regions close to the equator [40]. *Chaetoceros* sp. are marine algae that can even survive at −5 °C [4]. Barati et al. [20] evaluated the adverse effect of temperature on *Chlorella* species. They found that Antarctic and temperate algal growth was inhibited at temperatures above 38 °C–40 °C. Maximum cell growth or increase in cell density is essential to study ion removal by algae under different temperatures. Maximum algal growth occurred between 20 °C–33 °C [10,41]. Similarly, *C. vulgaris* and *S.* sp. could be cultivated at 25 °C–30 °C and 10 °C–40 °C, respectively. Hemens and Mason [42] reported that the removal rate of ions such as phosphate and nitrogen increased by 90% in warmer seasons compared with that in the winter season. The low rate of phosphate and nitrogen removal during winter was due to decreased pH and temperature [42]. This suggests that temperature affects algal growth and ion/nutrients uptake.

2.1.3. Effect of light intensity

The growth of the unicellular green algae *Chlorella* depends on the photoperiod, irradiance, and temperature [41]. *Chlorella* sp. contains the green pigments chlorophyll *a* and chlorophyll *b*, which promote photosynthesis. Light intensity is an important parameter for the growth of green microalgae. Li et al. [43] studied the growth of two algal species, *C. kessleri* and *C. protothecoide*, cultivated under different light intensities. They determined that the optimum light intensity for these species was 120 and 30 μmol of photon m^{-2} s^{-1}, respectively, and observed that both species demonstrated sufficient ion removal under all lighting conditions. Yeesang and Cheirslip [44] studied four green microalgae from lakes and ponds in southern Thailand. An improvement in lipid accumulation of up to 35% efficiency was observed with a combination of nitrogen deficiency and light intensity of 82 μmol of photon m^{-2} s^{-1}. Ho et al. [45] studied the effect of light intensity on the growth of *S. obliquus* and found that the algae showed maximum biomass productivity of 840.56 $mg.L^{-1}.d^{-1}$ at 420 μmol of photon m^{-2} s^{-1}. They also performed experiments using 540 μmol of photon m^{-2} s^{-1}, but the biomass growth significantly reduced [45].

3. Pathways of ion uptake by microalgae for their growth

The increase in the levels of a pollutant in the biomass of a living organism is known as bioaccumulation [46,47]. Ions that are taken up by algae accumulate in the cell structure, thereby increasing their concentration [48–50]. Algae are introduced in environments where large amounts of ions are present, which is favorable for biomass growth and the breakdown of cellular compounds to increase algal metabolism. Ion uptake by algae is defined by direct and indirect methods involving phosphate precipitation and nitrogen volatilization. The direct method involves ion uptake in which the biochemical pathways are interrelated to convert target ions into biomass for storage [51,52]. As shown in Fig. 3, nitrogen uptake is processed mainly by the thylakoid membrane and the Calvin cycle. The thylakoid membrane is present within cell chloroplasts. In the thylakoid, a light-dependent reaction breaks down H_2O to form O_2 and generates ATP and nicotinamide-adenine dinucleotide (NAD). The Calvin cycle occurs in the cell stroma. This cycle is the light-independent part of the reaction wherein CO_2 is converted to glucose using the ATP, and nicotinamide adenine dinucleotide phosphate ($NADP^+$) generated from the light-dependent reaction. Second, ions are assimilated into nucleic acids and proteins for biomass growth [53]. Assimilation occurs in all eukaryotic organisms, such as algae, and also contain nitrogen in the form of nitrate (NO_3^-), nitrite (NO_2^-), and ammonium (NH_4^+) ions and is translocated through the cell membrane in the order of $NH_4^+ > NO_3^- > organic\text{–}N$. The oxidized nitrogen species are converted into NH_4^+ ions and are assimilated into amino acids to form proteins [53,54]. The conversion of amino acids through NH_4^+ ion uptake occurs because of the lower energy requirement for the reduction and assimilation of these ions. Therefore, microalgae can be employed for total nitrogen removal (nitrification and denitrification), leading to the formation of NO_3^- after the uptake of NH_4^+ from the water source [55].

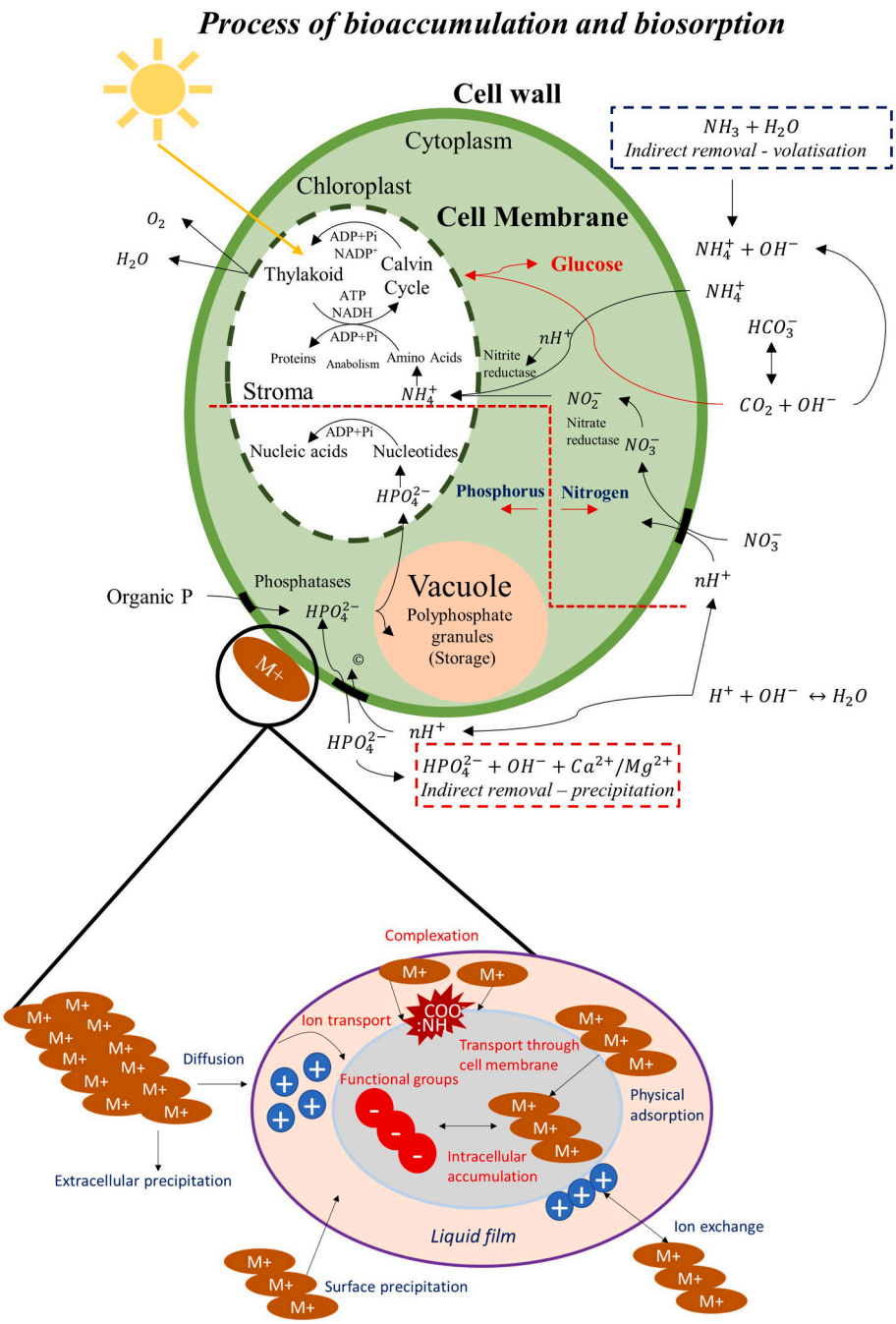

Fig. 3. Schematic mechanism of ion uptake by microalgae indicating 1) direct uptake through biochemical pathways resulting from deposition into storage (vacuoles) in cells and 2) indirect removal of nitrogen (volatilization) and phosphorus (precipitation). Encircled is a schematic illustration of the generalized biosorption mechanism of heavy metals/cations in algal cells.

Phosphates are present in the form of $H_2PO_4^-$ and HPO_4^{2-} transported through energized transport panels in the cell membrane [53,56]. These phosphates are then assimilated into nucleotides converting them into phosphorylation for storage in the cell vacuole. Production of ribosomal RNA nucleotides converts the phosphates into nucleic acids [57]. Therefore, a nitrogen source is essential to initiate protein synthesis to enable phosphorus assimilation, limiting either an ion in low cell protein content and reduced biomass growth [57]. In excess phosphate concentration, algae can utilize phosphates through a unique luxury uptake pathway [51,54,58], as shown in Fig. 3, as a storage channel for insoluble polyphosphate granules for future use in the cell vacuole. These stored granules can be utilized during phosphate insufficiency. Owing to higher nitrogen biomass content, nitrogen removal is always greater than phosphorus removal [59]. Nurdogan and Oswald [59] also determined experimentally that only 1–3 mg.L^{-1} of every 10 mg.L^{-1} of phosphorus was consumed by algae. For the cultivation of *C. vulgaris* and *S. sp.*, the external N:P ratio should be maintained at 8.5–32 and 4.1–32, respectively [57,60].

The byproduct of direct ion uptake and growth is the alkalization of the local environment, which leads to the indirect method of defining algal ion uptake. This alkalization is based on 1) hydroxyl ion (OH^-) generation during consumption of bicarbonate in photosynthesis [39,59,61] and 2) uptake of hydronium ions (H^+) from the dissipation of H_2O for the co-transportation of NO_3^- and PO_4^- through the algal cell membrane, as shown in Fig. 3 [39,62]. The physicochemical environment is controlled through pH, facilitating indirect volatilization and

precipitation [59]. When the pH increases to <10.5, phosphate precipitation decrease because of the shift toward calcium carbonate formation [63].

Biosorption is a physicochemical process that occurs in biomass (algae) that binds to a substrate [64–67]. Biosorption has extensive applications, such as in the heavy metal removal, production of micronutrient fertilizers, and removal of organic pollutants. Depending on biomass activity, the mechanism of biosorption is either metabolic-dependent or metabolic-independent [67]. The metabolic-dependent and metabolic-independent mechanisms can be further classified [68]. The metabolic-dependent mechanism comprises 1) transport through the cell membrane and 2) precipitation. The metabolic-independent mechanism comprises 1) physical adsorption, 2) ion exchange, 3) complexation, and 4) extracellular precipitation (Fig. 3) [69]. The mechanisms of biosorption are mostly interrelated and can occur in both metabolic and non-metabolic activity depending on algal classification. Metabolic-independent activity is usually faster and involves reversible physicochemical interactions between metals and functional groups on the algal cell surface [70]. On the other hand, intercellular accumulation (bioaccumulation [Fig. 3]) is slower and is a metabolic-dependent activity [71]. Metal precipitation is primarily affected by metabolic activity either when compounds interact with microorganisms or when a chemical action occurs between metals and cell surfaces [72,73]. The mode and characteristics of metal binding to biomass depends upon the heavy metal concentration and the conditions surrounding the biomass [74]. Therefore, the efficiency of metals' sorption to algae is affected by whether the biomass is living or dead. Hence, the growth rate of biomass is not a significant parameter to assess metal uptake, but active cells assimilate metals from water to be treated.

Similarly, extracellular polymeric substances (EPS) are essential parameters that increase biosorption yields, particularly when cell colonies form biofilms [67]. EPS are biopolymers produced by active cell secretion, shredding of cell surfaces, and cell lysis materials. EPS can be determined from the adsorption of organic compounds [75,76]. EPS are composed of organic substances with a higher amount of carbohydrates and proteins and lower fractions of uronic, humic, and nucleic acids. EPS have large, negatively charged functional groups, which renders them suitable for metal sorption. These discussions aim to provide background knowledge on the biosorption mechanism of algal cells. Processes such as bioaccumulation and biosorption promote ion uptake in water to enhance algal growth. Acclimation of algae may involve Na^+ and K^+ ions using a redox-driven Na^+ pump [77,78]. If Na^+ and Cl^- ions are translocated to the cell vacuole of microalgae, K^+ ions and organic chemicals such as proline and glycine are deposited in the cytoplasm. The algal cytoplasm then balances the osmotic pressure [79]. This mechanism was confirmed by [80], who showed that salt accumulates within *C. autotrophica* to regulate osmotic pressure within the cell.

4. Algal growth and biomass production in saline conditions

Changes in salinity affect algae in three possible ways: 1) osmotic stress with an impact on the cellular water potential; 2) ionic stress caused by the uptake or loss of ions—this loss of ions then becomes a part of acclimation; and 3) exchange of ions through the cell membrane due to the change in cellular ionic ratios [4]. Different marine species in different parts of the world are exposed to different salinities. For example, salinity varies from 37 $g.L^{-1}$ in tropical seas to 33 $g.L^{-1}$ in polar seas [4]. Halotolerant algae exhibit more resilience in conditions of extreme salt stress than in the environment in which they usually live [81]. Another halophyte species, *Pheridia tenuis*, cultivated in seawater, showed the ability to deposit chloride ions into cell vacuoles [82]. Therefore, as the water salinity increases, the capacity of halotolerant algae to remove NaCl increases with the uptake of other ions [9]. *S.* sp. and *C. vulgaris* have shown the greatest chloride ion RE and have produced higher amounts of lipids under conditions of high salinity [11]. Reed et al. [83] monitored algal growth by studying three cyanobacterial strains under the influence of salinity. They revealed the accumulation of organic solutes depending upon the temperature, salinity, and time duration.

von Alvensleben et al. [84] studied four species of freshwater algae—*Dunaliella armatus*, *S. quadricauda*, *Tetraedron* sp., and *Mesotaenium* sp.—in brackish water with salinities of 2, 8, 11, and 18 ppt. There was no significant effect on the growth rate of *D. armatus*, *S. quadricauda*, and *Tetraedron* sp. at salinities of 2, 8, and 11 ppt. At 18 ppt, the growth of all three species was significantly inhibited. *Mesotaenium* sp. showed maximum growth at 2 and 8 ppt, whereas the growth decreased significantly at 11 and 18 ppt. *D. armatus* exhibited maximum growth at 18 ppt and lowest growth at 2 ppt. These species are halotolerant freshwater species. They consumed sufficient amounts of nitrite from the growth medium to survive at higher salinities [85]. Gan et al. [5] observed that lipid production decreased with an increase in NaCl concentration in brackish water used to cultivate *S. obliquus*. They observed a reduction in the specific growth rate (red line) due to high salt stress, which ultimately inhibited the growth of *S. obliquus* cells and increased the inhibition rate. The marine algae *Dunaliella* sp. and *Nannochloropsis* sp. were cultivated in salinities >37 ppt [14,15]. This indicates the scale of the difference in salinity between halotolerant freshwater algae and marine algae.

At higher salinities, water contains ions rich in Na^+ and Cl^-. The growth of halotolerant freshwater algae was inhibited at higher salinities due to 1) overproduction of reactive oxygen species, which is not the reason for oxidative stress; 2) enzyme deactivation due to high salt stress; 3) decreased rates [86]; and 4) cellular ionic disproportion and major water loss [87]. The inhibition of algal growth arises from a limitation in the metabolic machinery/activity rather than a lack of organic carbon and nitrogen sources in the medium [87]. Acclimation to higher salinities involves 1) restoration and maintenance of cell turgor and volume, 2) changes in cell membrane porosity and adjusted uptake (K^+) and exclusion (Na^+) of ions, and 3) accumulation of osmoprotectant, compatible solutes, and stress proteins [88,89]. Therefore, high salinity conditions deactivate enzymes, reduce the rate of photosynthesis, and lead to loss of water from algal cells, thereby reducing cell density. The reduction in the number of algal cells leads to low ion uptake.

4.1. Effect of salinity on biomass production

A higher algal biomass production rate increases ion uptake [8,84]. In these studies, the nutrients required for algal growth were supplied mainly in growth media such as BG-11, Bold basal medium, and Bristol medium. At higher salinities, algal biomass production varied depending on the algal species and growth media provided. However, it is necessary to ascertain which algae species can take up ions under which conditions. For instance, if marine algae are cultivated in a freshwater medium, biomass growth will be negligible or undetectable. Thus, it is essential to determine which conditions should be provided for specific algae to maximize biomass yield.

The light intensity must be determined before cultivating algae under specific conditions. Von Alvensleben [84] observed lower biomass concentrations of *D. armatus*, *Mesotaenium sp.*, *Tetraedron* sp., and *S. quadricauda* by limiting the light provided to them to 40 μmol photons m^{-2} s^{-1}. In *S. quadricauda*, higher biomass was observed at light intensities of 100–200 μmol photons m^{-2} s^{-1} [85,90–92]. The studies reviewed for halotolerant freshwater algae in synthetic water were conducted mainly at irradiances of 40 and 105 μmol photons m^{-2} s^{-1} [8,12,93]. Tests on halotolerant wastewater algae were conducted at light intensities of 40, 50, 63, 85, 96, and 150 μmol photons m^{-2} s^{-1}, and the results were used to estimate biomass production [6,7,66,94–96]. Similarly, marine algae were cultured in wastewater and brackish water at irradiances of 40, 70, 85, and 96 μmol photons m^{-2} s^{-1} for biomass production [12,14,94,96]. The variation in the datasets at different salinities was taken into account (indicated by error bars, e.g., Fig. 4 a, b, c,

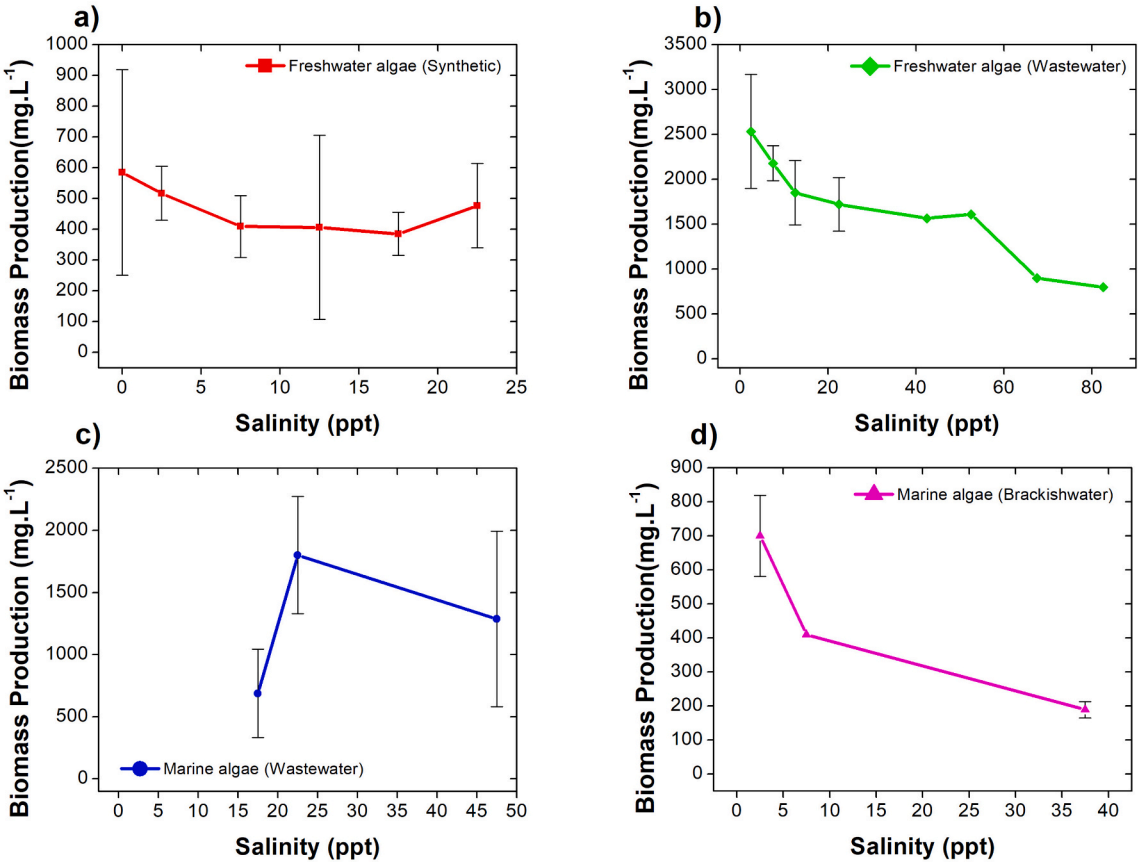

Fig. 4. Biomass production by a) halotolerant freshwater algae in synthetic water (red) [8,12,93], b) halotolerant freshwater algae in wastewater (green) [6,7,66,94–99], c) marine algae in wastewater (blue) [94,96], and d) marine algae in brackish water (magenta) [12,14]. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

and d).

As shown in Fig. 4, biomass production for both halotolerant freshwater and marine algae decreased with increasing salinity. For halotolerant freshwater algae cultured in the laboratory, the tilted effect was observed (Fig. 4a). Biomass production started at ~600 mg.L^{-1} and decreased to ~500 mg.L^{-1} at salinities of 0–22.5 ppt. One reason for this effect could be the different parameters used in the laboratory for different algal species. The effect of higher salt concentrations on biomass production was not investigated in many halotolerant freshwater algae.

Similarly, for halotolerant freshwater algae cultivated in wastewater, increased salinity was accompanied by reduced biomass production (Fig. 4b). An interesting trend was observed for marine algae. Biomass production by marine algae in wastewater increased from ~750 mg.L^{-1} to ~1800 mg.L^{-1} in salinities from 17.5 to 22.5 ppt, respectively. With a further increase in salinity, i.e., 47.5 ppt, biomass production decreased to ~1250 mg.L^{-1}. For marine algae cultivated in brackish water, biomass production showed a decreasing trend with increasing salinity. A biomass production of ~700 mg.L^{-1} was observed at a salinity of 2.5 ppt, which decreased to ~200 mg/L at a salinity of 37.5 ppt (Fig. 4d).

Figler et al. [8] found that *S.* sp. showed maximum chloride ion uptake at a salinity of 5 ppt, with a biomass production of 410.7 mg.L^{-1}. They also found that the maximum biomass production for *S. obliquus*, i.e., 508.7 mg.L^{-1}, occurred at 0 ppt and that for *Monoraphidium komarkove*, i.e., 374 mg.L^{-1}, occurred at 0 ppt; this production decreased gradually to 206 mg.L^{-1} at 12.5 ppt and was the lowest among nine species studied. *C. sorokiniana* showed the highest resilience to 22.5 ppt salinity and exhibited a biomass production of 503 mg.L^{-1}. The growth output of *S.* sp. was also significant but occurred at lower salinities, i.e., 4–8.8 ppt [5,8,9]. *C. vulgaris* was cultivated by Shen et al. [66] at salinities of 0–50 ppt, and it showed a maximum biomass production of 1610 mg.L^{-1} at 50 ppt. The answer to one of the open questions is that salinity does affect biomass production, with algal biomass decreasing with increasing salinity. Another cause of reduced biomass production by algae could be excessive salt stress on algal cells, which slows down enzymatic processes [86], reduces the rate of photosynthesis, and causes excessive water loss [87].

4.2. *Effect of salinity on algal growth rate*

The growth rate is an important parameter in estimating algal growth. Two parameters —growth rate and specific growth rate—have been used to estimate the algal size. The specific growth rate has been used to compare algal strains [100]. However, the specific growth rate changes under different conditions such as light intensity, pH, temperature, rate of CO_2 supply, and composition of growth media [100–103]. With respect to the measurement of the growth rate, there are few phases of algal growth. First, the exponential phase of microalgae cultivation ends with the transition of the cell culture to the stationary phase, which initially takes 7 days [100]. Usually, there are no significant changes in the growth rate during the stationary phase, and stable conditions persist [104]. Measuring the growth rate sheds light on algal behavior under certain conditions.

Lutzu and Dunford [94] reported a higher growth rate of 0.48 d^{-1} for *Pseudoanabaena* sp. in wastewater from shale oil and gas exploration sites. This wastewater is highly toxic and very saline (~80 ppt). Jiang et al. [14] cultivated *Nannochloropsis* sp. in F/2 medium at 37 ppt salinity and observed a growth rate of 0.57 d^{-1}. According to Gan et al. [5], the cell densities of *S.* sp. in water decreased from 0.46 d^{-1} to 0.35 d^{-1} as the salinity increased from 1.2 to 8.8 ppt, respectively. A similar

trend of reduced growth rate for *S.* sp. with increased salinity was observed in other studies [105–107]. Data for halotolerant freshwater algae were collected from various research articles [5,9,10,66,95–97,99,103,108,109]. For marine algae, data for growth rate estimation were collected from several articles [14,15,24,96,108–112]. The relationship between salinity range and growth rate has been estimated statistically for all the collected data. This conversion of data helped in the comparison of halotolerant freshwater and marine algae (Fig. 5). The halotolerant freshwater algae demonstrated the highest growth rate: ~ 0.8 d^{-1} at 2.5 ppt. However, they showed very high variation at 2.5 ppt, possibly due to the different algae types tested under different conditions. As the salinity increased, the growth rate of halotolerant freshwater algae decreased. Similarly, the marine algae exhibited a bell curve-like pattern. Compared with halotolerant freshwater algae, marine algae showed a shift of scale of 5 ppt from the first data point. Marine algae required an initial salinity of 7.5 ppt and exhibited a growth rate of ~0.2 d^{-1}.

The growth rate trend of marine algae increased to the maximum value of ~1.5d^{-1} at 27.5 ppt, followed by an increase in salinity. A sudden decrease in the growth rate was observed. Thus, it is evident that marine algae grew at higher salinities than freshwater algae and exhibited higher growth rates. Even for algae cultivated in a marine environment, a gradual decrease in growth rate was observed after increasing the salinity. Fig. 5 supports our hypothesis that algal growth decreases with an increase in salinity.

4.3. Cultivation of algae in seawater

Algae require specific nutrient media containing particular ions to grow. However, there are some studies performed on the cultivation of algae directly into the seawater. Ahamefule et al. [113] cultivated three freshwater strains, *Desmodesmus subspicatus*, *Desmodesmus armatus,* and *Dictyosphaerium spp.,* in the seawater having 37.5 g.L^{-1} of salinity. They observed the change in salinity for *Desmodesmus subspicatus* (30%), *Desmodesmus armatus* (27.5%), and *Dictyosphaerium spp.* (20%) on the fourth, eighth, and tenth week of the experiment, respectively. During the cultivation process, there was an increase in the pH due to the photosynthesis process. This increase could be due to converting the carbonate and bicarbonate for CO_2 consumption, and therefore the pH scale of seawater increased toward alkaline scale [99,113].

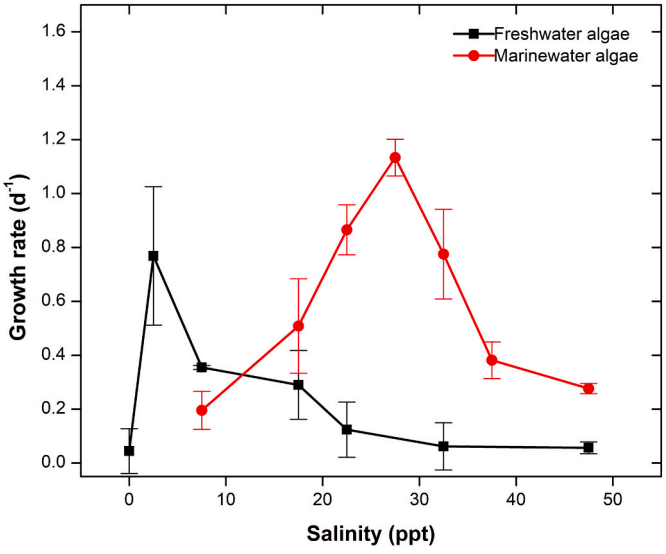

Fig. 5. Graph of halotolerant freshwater and marine algae under the influence of salinity, as analyzed from different studies. The solid black line indicates the growth rate (d^{-1}) of halotolerant freshwater algae in relation to salinity, and the red line depicts the growth rate of marine algae. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

Ummalayma et al. [114] isolated *Chlorococcum* sp. in the seawater samples where the strain could grow naturally. The cultivation of algae in seawater is more beneficial as it already contains nutrients essential for algal growth, and the seawater cannot be contaminated with fungi and bacterial growth. Hence, providing sterile conditions for algae to grow and flourish in seawater. Therefore, freshwater algae can be cultivated in seawater. It has been observed that by cultivating freshwater algae in high salinities (or seawater), more lipid extraction can be yielded to produce more biofuels [114]. There is another observation that algae can adapt to extreme acidic seawater, where the algae were introduced to water with pH 2.0 [115]. It is proposed that under acidophilic conditions, there are fewer chances of contamination to the algae. However, the acidophilic condition could be a positive strategy to cultivate algae in natural open ponds where chances of contamination would be minimal.

5. Ion and nutrients uptake in halotolerant freshwater and marine algae at different salinities

Several plants can take up Na^+ ions by decreasing their absorption of K+ ions' to maintain the K^+/Na^+ ratio [117]. As algae behave like plants; therefore, Cl^-, the selective pump was discovered in *Scenedesmus species* [120]. However, the chemical energy carrier ATP powers Na^+ export from cells or by Na^+, pumping ATPases or H^+–ATPases [78,121]. If the cell encounters high salinity during the growth phase, the growth phase stops, Na^+ ions are exported, and the Cl^- ions absorb light through halorhodopsin [9]. The Cl^- in the cell drives Na^+ through the protein synthesis channel by penetration through the cell membrane [121]. Before the Na^+ requirement, numerous cell morphology mechanisms occur, including ion uptake, pH homeostasis in an alkaline environment, photosynthesis, and nitrate assimilation [77,122,123]. In this section, different ion analysis has been discussed.

5.1. Uptake of chloride ions under salinity

The literature reviewed covers various relevant ions, including chloride, nitrate, phosphate, calcium, magnesium, and potassium. Ion uptake analyses for chloride ions have been reviewed [8,103]. Figler et al. [8] analyzed chloride ion removal in nine algal species. They found that *M. komarkove* takes up 44.59% of chloride ions at 0 ppt salinity. Similarly, with *S. obliquus*, they found that the maximum removal of chloride ions occurred at 5 ppt. Sahle-Demessie et al. [9], in their study of *S.* sp. and *C. vulgaris*, found promising results in the uptake of chloride ions by 30% after separating algae from a suspension. Lutzu and Dunford [94] experimented on freshwater and marine algae and found that *Phormidium keutzingianum* showed maximum chloride and nitrate ions uptake. Wei et al. [118] experimented on fresh and dry *Scenedesmus obliquus* in the salinity range of 2.8 to 8.8 g/L. The findings concluded that fresh algae removed NaCl approx. 20% and 15% by dry algae in 8.8 g/L of NaCl in the solution. Thus, the studies involved various experimental setups and presented clear evidence of the existence of ion uptake by algae. Chloride ion analysis is shown in Fig. 6, respectively, which shows RE in % and elimination capacity in g.m^{-3}.h^{-1}.

The box-whisker plot better illustrates the prediction of a trend of algal RE. The first box represents RE at 0 ppt salinity, which is less than ~10% at the mean value. Other boxes show salinities of 2.5, 7.5, 12.5, 17.5, 22.5, and 27.5 ppt. The midpoint of boxes illustrates the median point. The maximum RE of ~28% for chloride ions occurs at 22.5 ppt. The box at 27.5 ppt shows only a single point due to a lack of data in that salinity range. Similarly, for elimination capacity, the line plot increases gradually from 0 ppt. At 22.5 ppt, elimination capacity reaches its maximum uptake capacity of ~7 g.m^{-3}.h^{-1}. However, as salinity increases to 27.5 ppt, elimination capacity decreases to ~0.8 g.m^{-3}.h^{-1}. Ultimately, chloride ion uptake by algae shows significant elimination

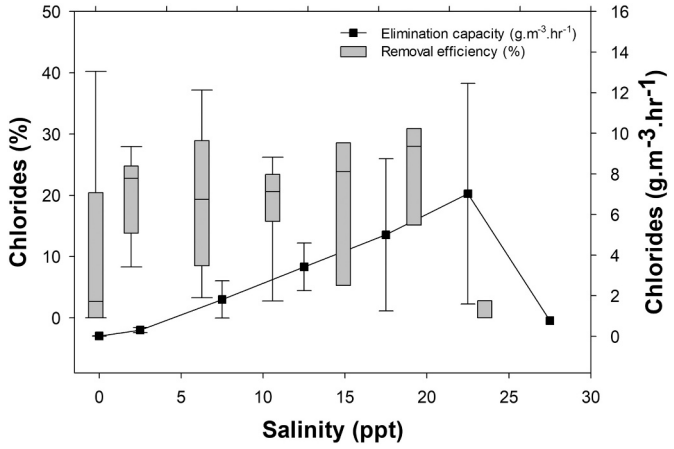

Fig. 6. Chloride removal efficiency (%) in the box-whisker plot—the line with error bars illustrates algae's elimination capacity (g.m − 3.h − 1) of chloride ions.

capacity under the influence of salinity. However, more thorough experiments are needed to reach better conclusions to estimate the effect of chloride ions on the algae uptake phenomenon.

5.2. Uptake of nitrate and phosphate ions

Nitrate and phosphate are essential for algal growth. Gordillo et al. [124] studied three brown algae species in seawater and 50% diluted seawater and found that these species can store inorganic nitrogen through intracellular pathways. Faster uptake of nitrate and phosphate due to the accumulation of cell vacuoles was observed. When the cell vacuoles reached maximum saturation, the rate of uptake was reduced [124]. However, the faster rate of uptake of phosphate and nitrate ions by algae could be attributed to either greater biomass production or a capacity to store these ions [8,84]. The more algal cells in water, the more nitrate and phosphate ions can be stored in the cell vacuoles.

Furthermore, if the intracellular nitrate ion concentration is low, algae will take up nitrate ions from the external source [124]. The data on nitrates and phosphates were collected from reputed articles [5,8,94,97,99,103,116]. In this review, the results of nitrate and phosphate ion RE and elimination capacity are summarized in relation to salinity (Fig. 7).

Fig. 7a depicts the nitrate ion RE in a box-whisker plot. The box plot represents the data points for salinities of 0, 2.5, 7.5, 12.5, 17.5, and 22.5 ppt. These points contain the box plot data representing the range of data collected from different literature sources. The maximum RE occurs at 0 and 22.5 ppt, i.e., ~ 90% and ~ 88%, respectively. However, on comparing the median value of the box plot, there is a slight increase in RE from 7.5 ppt, with an increase in salinity for nitrate ions. This could be due to the different algal species included in this review. An inline plot for nitrate ion elimination capacity is shown in Fig. 7a. Elimination capacity remains constant throughout all the salinity levels, i.e., ~0.45 $g.m^{-3}.h^{-1}$. The variation in nitrate ion elimination capacity is not significant because most algal species have a unique capacity for storage of nitrate ions in the cell vacuoles [124]. Another reason for the low degree of variation could be that algal cells require nitrate ions to increase their growth and biomass. Therefore, there is no large effect of salinity on nitrate ion uptake. There is a considerable variation in the nitrate ion dataset, which may be due to different experimental setups and algal species.

There is a large degree of variation in the dataset for uptake of phosphate ions (Fig. 7b). The median value of the box plot remains <10% for all salinities. These large variations are attributed to the diverse experimental setups with different parameters. In addition, the large variation in the data may be due to the use of different algal

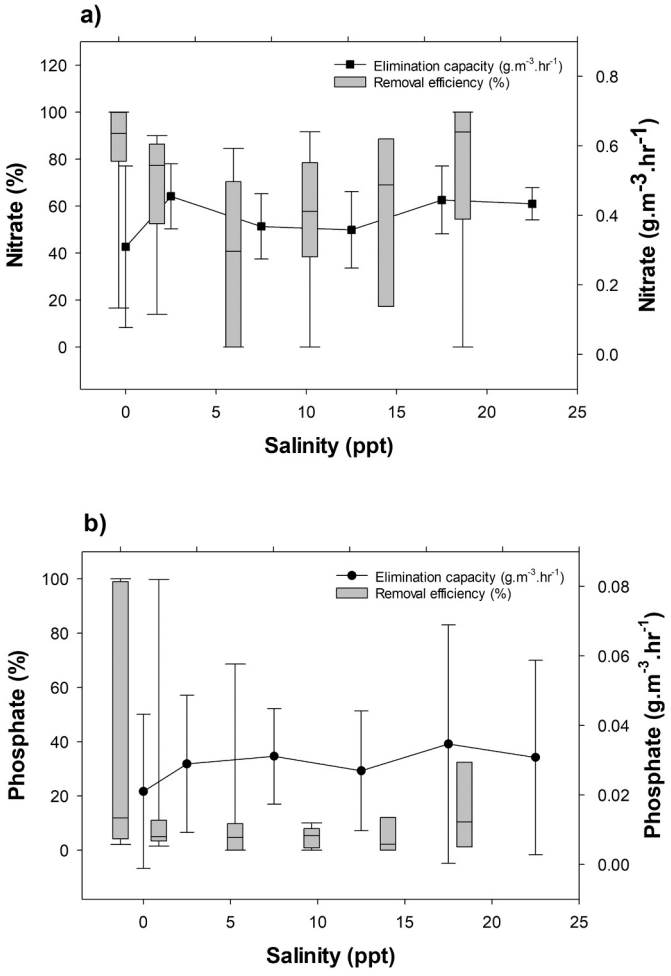

Fig. 7. a) Nitrate removal efficiency (%) in the box-whisker plot—the line with error bars indicates the elimination capacity ($g.m^{-3}.h^{-1}$) of nitrate ions by algae. 6b) Phosphate removal efficiency (%) in the box-whisker plot—the line with error bars illustrates the elimination capacity ($g.m^{-3}.h^{-1}$) of phosphate ions by algae.

species. However, if we compare the elimination capacity using box plots, the elimination capacity values remain constant at ~0.03 $g.m^{-3}.h^{-1}$. The main activation energy channel for the algae, nitrates, and phosphates is taken up by algal cells through nitrogenases and phosphatases [125]. Nitrates enter the cell through the cell membrane and are converted into nitrite by nitrate reductase [39,62]. This nitrite is further broken down into ammonium ions [54]. These are then consumed by the cell stroma to generate amino acids, which further synthesize proteins within the cell (Fig. 3). Similarly, phosphate is an important constituent of algal cells, but the requirement for phosphate is less than that for nitrogen. Usually, algae consume 3 $mg.L^{-1}$ of phosphate ions for every 10 $mg.L^{-1}$ of nitrogen ions [57,60].

5.3. Uptake of sodium, potassium, magnesium, and calcium ions by algae

Positive ions can adhere to algal cell walls because they contain a negative charge, thereby removing cations from water [126]. Algae take up cations through physical adsorption, biosorption, surface deposition, active transport, and passive diffusion [67,126]. However, there is still considerable debate about the role of algae in consuming these cations under saline conditions. Fig. 8a shows the elimination capacity for calcium, potassium, sodium, and magnesium.

RE data have been extracted from various experimental investigations on algae [15,94,117,119]. Elimination capacity has been

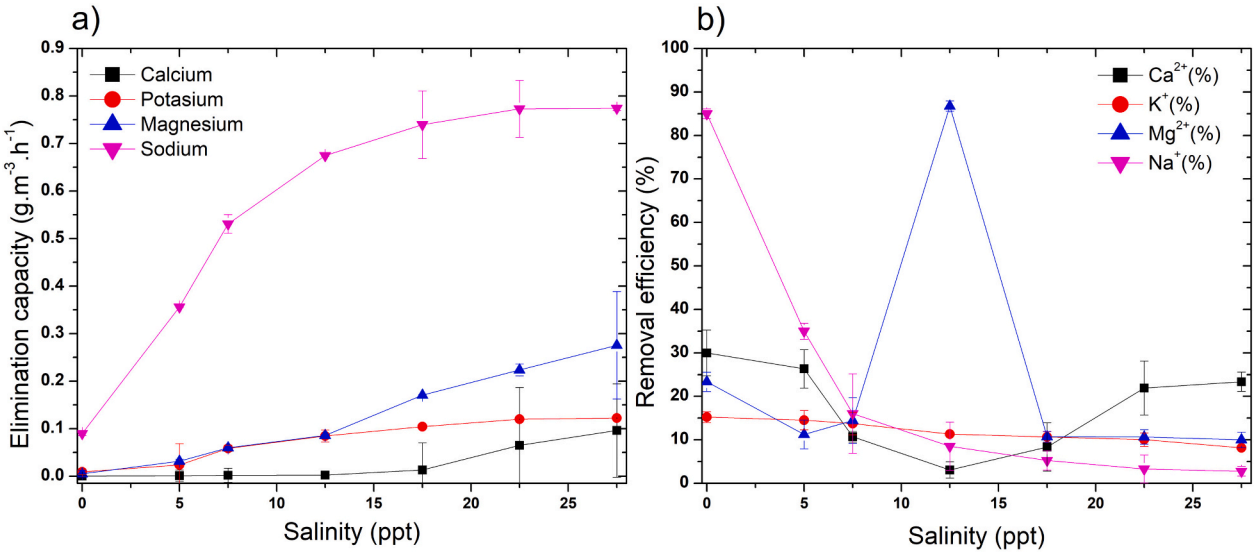

Fig. 8. a) Elimination capacity of cations i.e., sodium (Na^{+}), potassium (K^{+}), calcium (Ca^{+2}), and magnesium (Mg^{+2}). b) Removal efficiency of cations displayed in the form of a bar chart, where each bar represents sodium (Na^{+}), calcium (Ca^{+2}), magnesium (Mg^{+2}), and potassium (K^{+}) on the scale of salinity in ppt.

calculated for sodium, potassium, magnesium, and calcium ions using the data from the same studies. The salinity has been converted to a unified scale using the same normalizing technique as those used for all previous results. Fig. 8a illustrates the elimination capacity of cations, with Na^{+} ions showing elimination capacity values of ~0.55 $g.m^{-3}.h^{-1}$ at 7.5 ppt salinity. However, at 22.5 ppt salinity, the Na^{+} ions' curve stabilizes at an elimination capacity of ~0.75 $g.m^{-3}.h^{-1}$. The results for Ca^{+2} and K^{+} ions show no significant increases or decreases. The elimination capacity values for Ca^{+2} ions reach 0.09 g. $m^{-3}.h^{-1}$ at 27.5 ppt salinity. For K^{+} ions, there is a stable trend from 7.5 ppt to 27.5 ppt at an elimination capacity value of ~0.1 $g.m^{-3}.h^{-1}$. However, the uptake of Mg^{+2} ions increases from 12.5 ppt for elimination capacity and reaches a maximum value of ~0.3 $g.m^{-3}.h^{-1}$at 27.5 ppt.

Fig. 8b depicts the RE of Na^{+}, K^{+}, Ca^{+2}, and Mg^{+2}. The data for RE are extracted from the same source as those for elimination capacity. The bar chart indicates the RE of cations at salinities of 7.5, 12.5, 17.5, 22.5, and 27.5 ppt. Na^{+} is represented first, and its RE decreases from ~85% to ~3% at 0 to 27.5 ppt salinities, respectively. Ca^{+2} ions show a decreasing trend at 12.5 ppt and increase after 17.5 ppt to an RE of ~23% at 27.5 ppt. Similarly, Mg^{+2} ions show a maximum RE of ~87% at 12.5 ppt, which is reduced to ~10% at 27.5 ppt. RE of K^{+} ions declines with an increase in salinity. The RE decreases from ~14% to ~8% as the salinity increases from 7.5 to 27.5 ppt. It is evident that RE in algae is inhibited as salinity increases. These ions should be investigated further using other algal species cultivated under various salinities. A complete removal efficiency analysis of different species has been shown in Table 1.

5.4. Uptake of chemical oxygen demand (COD), total nitrogen, total phosphorus, and ammonium ions by algae

Algae also utilize COD in wastewater treatment. Only studies of high salinity, TDS, and chloride values have been used for analysis in this review. Lutzu and Dunford [94] cultivated algae in produced water. Produced water tends to be more saline, with a TDS of approximately 463,000 ppm. For this review, data for COD, total phosphorus, total nitrogen, and ammonium ions were collected from studies by [6,94,98]. The response of algae is measured in elimination capacity, i.e., elimination capacity. Similarly, for salinity, data are normalized to the scale 0, 2.5, 7.5, 12.5, 17.5, 22.5, 27.5, and 32.5 ppt.

Fig. 9 displays the results of ion uptake by algae in wastewater. Fig. 9a depicts COD uptake by algae. COD is an indication of the oxygen available in the water to oxidize organic matter. The trend of COD at 2.5 ppt of salinity was ~18 $g.m^{-3}.h^{-1}$; however, an increasing trend of elimination capacity was observed up to ~36 $g.m^{-3}.h^{-1}$at 12.5 ppt. At 22.5 ppt, the elimination capacity of COD uptake was reduced to ~1 g. $m^{-3}.h^{-1}$. This indicates a decrease in algal COD uptake with an increase in salinity; this decline can be attributed due to different reasons.

Similarly, wastewater characteristics, algal growth, and algal type depend on algal behavior under different conditions. This analysis, shown in Fig. 9, is based on salinity and COD, irrespective of algal species and growth conditions. Nevertheless, Fig. 9a shows a general decreasing trend for COD after an increase in salinity. As nitrogen is essential for the Calvin cycle and thylakoid membrane in algal growth, it is important to consider total nitrogen. Fig. 9b displays the elimination capacity of total nitrogen uptake by algae. Total nitrogen uptake increases gradually as the salinity increases. At 27.5 ppt salinity, the elimination capacity of total nitrogen uptake by algae is ~0.9 $g.m^{-3}.h^{-1}$. This suggests that nitrogen uptake by algae will be high irrespective of the water type. Thus, there will be less algal growth if there is insufficient nitrogen in the water. Algae can consume high amounts of nitrogen, even with increased salinity (Fig. 9b).

Phosphorus is also an important parameter. Fig. 9c depicts the elimination capacity of total phosphorus uptake by algae. From 0 ppt salinity, there is no significant uptake of total phosphorus ions, but the uptake increases to ~0.25 $g.m^{-3}.h^{-1}$at 2.5 ppt. The highest peak in the elimination capacity of total phosphorus uptake of ~0.26 $g.m^{-3}.h^{-1}$ occurs at 7.5 ppt salinity. Thereafter, the total phosphorus uptake decreases steadily to an elimination capacity of ~0.08 $g.m^{-3}.h^{-1}$at 27.5 ppt salinity. Thus, it is evident that there is a considerable reduction in total phosphorus with an increase in salinity. Further, the elimination capacity of total phosphorus is lower than that of total nitrogen.

Similarly, as reported in the reviewed experimental studies, ammonium ions are essential. Fig. 3 shows that ammonium ions are converted in the cell stroma to produce amino acids. These amino acids then further synthesize proteins in algae. Thus, we evaluated the capacity of algae to take up ammonium ions under the influence of salinity. Fig. 9d illustrates the elimination capacity of ammonium ion uptake by algae against salinity. At 0 ppt salinity, algae consume ~0.6 $g.m^{-3}.h^{-1}$ of ammonium ions. The elimination capacity is 0.6 $g.m^{-3}.h^{-1}$ up to 12.5 ppt salinity, but after a further increase in salinity, there is a sudden reduction in ammonium ion uptake elimination capacity. At 47.5 ppt salinity, the elimination capacity of ammonium ion uptake by algae decreased to ~0.3 $g.m^{-3}.h^{-1}$. We conclude that ammonium ion uptake

Table 1
Data for different algae species cultivated in other conditions of temperature, irradiance, type of water, and reactors used. Salinity range (ppt) has been shown in minimum to the maximum range with corresponding values of growth rate (d^{-1}), biomass production (mg. L^{-1}), biomass productivity (mg. L^{-1}. d^{-1}), and removal efficiencies of specified targeted ions measured in experiments.

Sr. No.	Algae specie	Water type	Temp (°C)	Irradiance (μ.mol m^{-2} s^{-1})	Type of reactor	Salinity (ppt) Min - Max	Nutrient media	Growth rate (d^{-1})	Biomass production (mg. L^{-1})	Biomass productivity (mg. $L^{-1}d^{-1}$)	Ions measured	Removal efficiency (%)	Ref.
1	*Chlorella sorkiniana* *Chlorella vulgaris* *Chlorococcum sp.* *Desmodesmus communis* *Desmodesmus spinosus* *Scenedesmus obliquus* *Scendesmus obtusus* *Monoraphidium komarkove* *Monoraphidium pusillum*	Synthetic	24	40	Batch Reactor	0–20 0–20 0–10 0–10 0–10 0–5 0–5 0–5 0.5–5	Bold Basal Medium	–	380–503 457.7–329 625.3–578 354.7–507.3 584–406 508.7–410.7 534–488 374–225 490–414	–	Cl^{-}	12.06–28.21 14.69–26.99 39.11–18.75 23.53–15.75 20.59–17.10 2.65–40.87 16.61–31.63 44.59–19.33 26.65–10.08	[8]
2	*Chlorella vulgaris* *Scenedesmus obliquus*	Brackish	25	40	Continuous PBR	2–4	Bristol Medium	0.21 1.82	– –	– –	Cl^{-} Cl^{-}	25 30	[9]
3	*Phormidium kuetzingianum*	Wastewater	25	85	Batch Reactor	24	BG-11 + 1% NaCl	0.25	1350	51	Cl^{-}	64	[94]
4	*Chlorella vulgaris*	Wastewater	25	40	Batch Reactor	0–50	Artificial Seawater Medium	1.51	1460–1610	–	–	–	[66]
5	*Desmodesmus armatus* *Mesotaenium sp.* *Scenedesmus quadricauda* *Tetraedron sp.*	Freshwater	24	42	Batch Reactor	2–18	Modified L1 medium	–	–	41–17 54–9 37.9–0 37.8–10.4	–	–	[84]
6	*Scenedesmus obliquus*	Brackish	25	50	Batch Reactor	1.2–8.8	BG-11	0.46–0.35	–	–	Cl^{-}	8.33–28.99	[5]
7	*Chlorella vulgaris*	Synthetic	26	150	Batch Reactor	0	Bristol Medium	–	–	–	$NO3^{-}$ $PO4^{-}$	1.72–100 10–100	[116]
8	*Chlorella vulgaris*	Synthetic	–	105	Batch Reactor	0–30	–	–	–	1500–0	–	–	[93]
9	*Scenedesmus dimorphus*	Wastewater	25	100	Bubble column tube reactor	0	BG-11	–	–	–	$NO3^{-}$ $PO4^{-}$ COD	99.99 99.66 59	[97]
10	*Scenedesmus sp.*	Wastewater	25	63	Batch Reactor	0	BG-11	0.0685	875.60	32.43	COD	38.88	[95]
11	*Chlorella vulgaris*	Wastewater	25	50	Batch Reactor	4.2–9	TAP media	–	1800–2400	–	Cl^{-} $PO4^{-}$ $NH4^{+}$ COD TN	8.8–2.6 80.4–38.4 99.18–98.246 42.2–34.1 71.4–51.3	[6]
12	*Chlorella sorkiniana*	Brackish	28	150	Batch Reactor	0–11.36	BG-11	–	2600–1700	213.6–157.7	–	–	[7]
13	*Nanochloropsis sp.*	Brackish	26	70	Photobioreactor	37	f/2 medium +50% MW	0.54	212	–	–	–	[14]
14	*Nannochloropsis salina* *Dunnaliella tertiolecta*	Wastewater	22	29–55	Batch Reactor	45.5 45.65	f/2 medium +2% AD f/2 medium +4% AD	0.276 0.316	–	173 239	–	–	[15]
15	*Dunaliella sp.* *Pseudanabaena sp. (SP 46)*	Wastewater	23	96	Batch Reactor	15	f/2 medium A+	0.76 0.61	620 1320	51.8 55.9	–	–	[96]
16	*Enteromorpha sp*	Seawater	20	90	Batch Reactor	35	SM medium	0.387	–	–	–	–	[110]
17	*Chattonella ovata (CO8)*	Seawater	30	15–45	Batch Reactor	30	Modified SW3-medium	1.47	–	–	–	–	[24]
18	*Chattonella marina*	Seawater	25	400	Batch Reactor	28	Seawater	1.08	–	–	–	–	[111]
19	*Nitzschia thermalis*	Synthetic	22	150	Batch Reactor	34	f/2 medium	0.35	–	–	–	–	[109]
20	*Phaeocystis globosa*	Synthetic	18	150	Batch Reactor	30	Seawater	1.17	–	–	–	–	[112]

(*continued on next page*)

Table 1 (*continued*)

Sr. No.	Algae specie	Water type	Temp (°C)	Irradiance (μ.mol m^{-2} s^{-1})	Type of reactor	Salinity (ppt) Min - Max	Nutrient media	Growth rate (d^{-1})	Biomass production (mg. L^{-1})	Biomass productivity (mg. $L^{-1}d^{-1}$)	Ions measured	Removal efficiency (%)	Ref.
21	*Scenedesmus sp.* *Chlamydomonas mexicana*	Synthetic	27	40	Batch Reactor	1.9–6.2	Bold Basal Medium	–	670–440 820–410	–	–	–	[12]
22	*Scenedesmus obliquus*	Synthetic	25	70	Batch Reactor	10–25	Bristol Medium	–	–	–	Ca^{+2} Mg^{+2} K^{+}	3.09–23.37 86.75–9.99 13.73–8.17	[117]
23	*Scenedesmus sp.*	Synthetic	25	25	Batch Reactor	5–25 0	BG-11	0.82	–	–	NO_3^{-} PO_4^{-} NH_4^{+}	82.5 100 25	[103]
24	*Scenedesmus obliquus (Fresh)* *Scenedesmus obliquus (Dry)*	Synthetic	25	50	Batch Reactor	2.8–8.8	BG-11	–	–	–	TDS	5–20 6.3–15	[118]
25	*Chlorella vulgaris*	Synthetic	25	81	Batch Reactor	0–4	Modified BG-11	–	–	–	Na^{+}	85–35	[119]
26	*Botryococcus sp.*	Synthetic	18–38	2.7-324	Batch Reactor	– –	Bold Basal Medium	0.572–0.804 0.412–1.160 0.959–0.554	– – –	– – –	– – –	– – –	[10]

by algae reduces as salinity increases.

6. Concluding remarks and recommendations

Algal-based water treatment receives maximum attention from the scientific community as an alternative to desalination technologies due to reasonable chloride and sodium ion removal. Other potential pollutants in water, such as nitrate and phosphate ions, can be fundamental nutritional sources for algae. Besides these advancements, algae in biofuels, lipids, fatty acids, and protein extractions have been highly investigated in the literature. This current review has presented that ion uptake by algae decreases under conditions of high salinity. However, in the ranking, marine water > freshwater in terms of growth rate from the analysis because marine water species are adaptive to more saline environments than freshwater species. Moreover, halotolerant freshwater microalgae were tolerant up to brackish salinities; e.g., *C. vulgaris* and *S.* sp. showed growth in brackish water consuming unwanted ions [8,9]. A constant increase in the salinity for halotolerant freshwater and marine algae led to a decrease in the growth rate, as shown in Fig. 5. Another reason for the slow growth at higher salinities could be that fatty acids in the cell membrane are destabilized and stop cell growth through photosynthesis [127]. This reason could explain why growth was inhibited at higher salinities in most studies included in this review.

Another hypothesis has been evaluated; i.e., higher salinity affects biomass production. The literature considered in Fig. 4 shows that higher salinities affect biomass production by both halotolerant freshwater and marine algae. Various studies have reported on algae behavior at higher salinities. Greater biomass production by algae eventually increases the ion uptake capacity of algae [8,84]. However, biomass production decreases with an increase in salinity. Therefore, at higher salinities, reduced biomass production could also explain the reduced ion uptake capacity of algae.

Further research is still needed to enhance our understanding of the ion uptake capacity of algae. However, it is recommended to explore the ion uptake mechanism by algae under different growth conditions. There are no fixed parameters, such as growth media, light intensity, temperature, and pH, for algal growth. Algal growth depends upon the source of origin, such as freshwater and marine water. Therefore, as evidenced by these studies, it is challenging to develop a uniform pattern for ion uptake by algae due to the versatile nature and fragility of the surrounding environment. Hence, a comprehensive study on the effect of parameters with regard to salinity may improve our understanding of the behavior of halotolerant freshwater and marine algae. It is also expected that the ion uptake capacities of various other algal species that are not listed in this review will be explored. We hope that additional novel algal species will be explored in the forthcoming years.

7. Limitations and future prospects

The scope of algae in water treatment is limited because algae-based water treatment performs adequately when provided in suspension, and water appears in a greenish colour. In this method, water is directly exposed to unicellular algal organisms for the exchange of ions. The water could be treated efficiently, but aesthetically, water remains unfeasible for direct use. Further, the small size, cell density, opposing charge cell surface, reduced cell dry weight cause challenges to harvest biomass. Treating water through suspended algae requires algal harvesting techniques, which include sedimentation [128], bio-flocculation [129], centrifugation [130], pressure filtration [129], and dissolved air flotation (DAF) [131–134]. Sedimentation is the most economical treatment of these technologies, while centrifugation, pressure filtration, and DAF are the expensive algae harvesting techniques because more energy is required to operate these technologies. The cost-efficient natural treatment using algae parallels the cost of treatment by conventional RO plants using these technologies other than sedimentation. Therefore, the scientific community is spending time and effort to

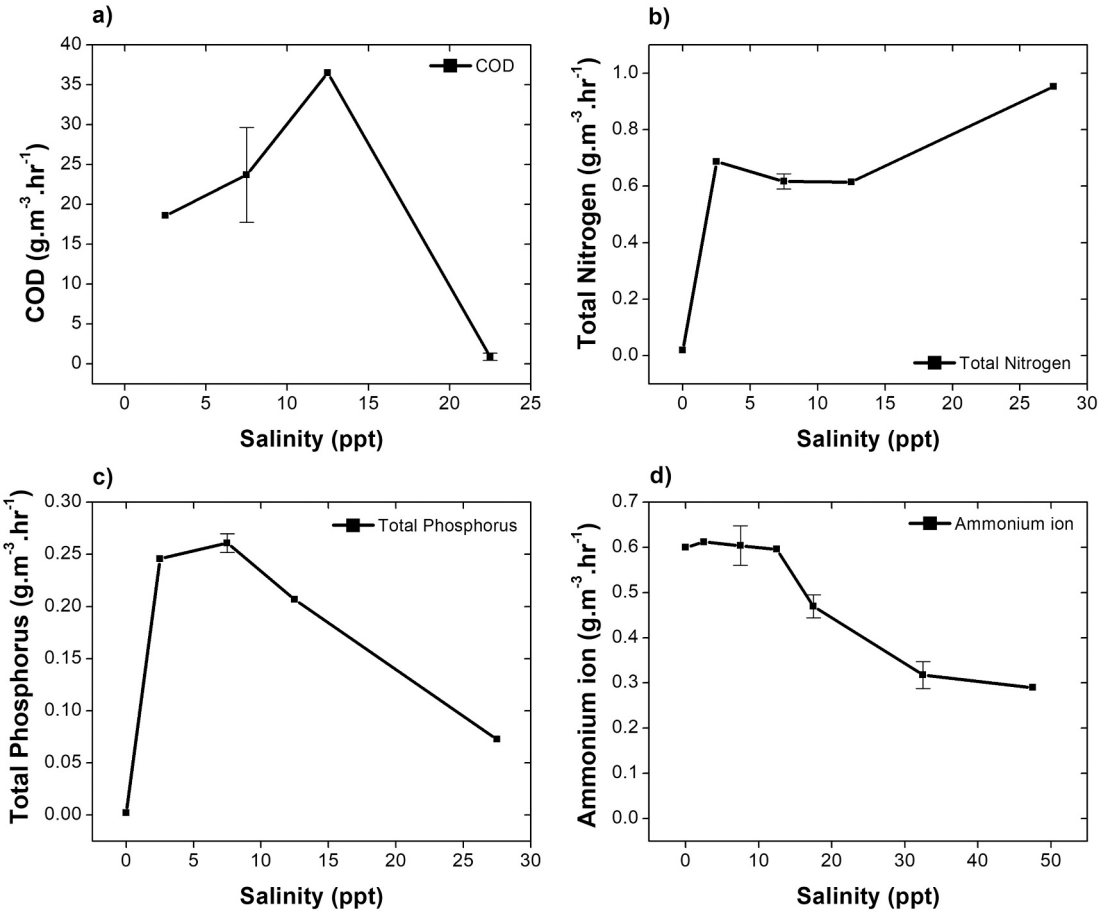

Fig. 9. Analysis of elimination capacities of algae in wastewater. The subparts illustrate a) COD reduced by algae in salinity, b) Total nitrogen uptake rate, c) Total phosphorus uptake rate, and d) ammonium ion uptake rate.

develop the best possible solutions to harvest algae.

Further, the slower rate of ion recovery is another major constraint of the algal-based treatment methods. The treatment time of saline water through reverse osmosis is faster and produces freshwater at the rate of approx. 10–20 L.min^{-1}. However, there are possibilities to design an engineered system to recover the saline water through biological species at faster rates. Many research groups are targeting to remove chloride and sodium ions from water using microalgae, this technique is known as biodesalination. These research areas will explore new methodologies in biological water treatment techniques to be discovered in upcoming years.

Some improvements are required in biodesalination, such as biomass separation in suspension for studies as mentioned above. One solution could be using an immobilized form of algae, where algal beads are formed. The algal growth occurs inside the bead, and water can be treated effectively and efficiently, and the algal biomass can be harvested easily in the immobilized form of algae. In immobilizing algae, water could be contaminated from the growth of fungi and bacteria. For that reason, as mentioned earlier in Section 4.3, algae could be grown in acidophilic conditions to avoid any contamination.

Furthermore, this study could help the designers of water and wastewater treatment plants using algae as a primary treatment unit to consume unwanted ions from the water. The pre-treated water with lower pollutant loading rates will reduce the energy consumption in water and wastewater treatment plants. The review also targets saline water treatment where performance has been evaluated based on salinity to uptake the ions from desalination water plants or petrochemical industrial waste. Nevertheless, the countries consuming seawater can also use halotolerant algae to uptake some of the ions such as sodium and chloride, reducing the loading potential on desalination treatment plants. Hence, algae can be utilized in many fields. The strain selection of algae is a critical step to achieve the attributed goal. For the biodesalination process, halotolerant species should be used to achieve maximum salt removal. There are a variety of species that are not well documented. There are more areas to be explored by the newly isolated species for several new processes, and biodesalination is among them, which could be researched more in the upcoming years.

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CRediT author statement

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Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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