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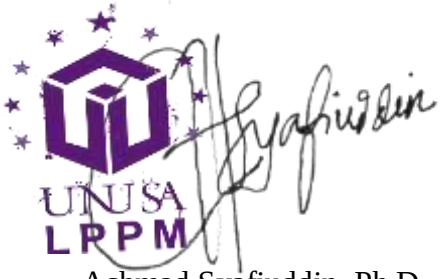

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Effect of Algal Cells on Water Pollution Control

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Abstract

Purpose of Review The use of algae for remediation of toxic pollutants seems to be promising since they also provide some advantages such as the production of valuable products and their capability to capture CO_2 during the photosynthesis, which potentially decrease greenhouse gas emission. This paper reviews the evidence for highlighting the effectiveness of the use of living or non-living algal cells for treating polluted waters.

Recent Findings Removal efficiency and sorption capacity of algal non-living cells are higher than in living cells because of cell membrane disruption (leading to enhancement of intracellular pollutants binding) and the improvement of specific surface area. For the kinetic and isotherm modeling, there is no single powerful model for a wide range of pollutants and type of algae, indicating that the mechanism is quite specific depending on the type of algae, type of pollutants, and environmental conditions. The removal mechanism of pollutants by living and non-living algae can be considered as an exothermic reaction and physical sorption from many published reports.

Summary The use of non-living cells was more effective compared to living cells for a wide range of pollutants since the non-living cells performed better removal efficiency and sorption capacity as well as easy to handle. This review is useful to pave a good strategy for designing a greener technology for future environmental pollutants remediation particularly within the domain of algal-based technology.

Keywords Algae · Living and non-living cells · Bioaccumulation · Biotransformation

Introduction

Recently, remediation of polluted water using biosorbents have emerged because they are considered eco-friendly and effective as well as have the potential as low-cost material option because they are freely available in environment [1, 2]. These biosorbents include fungi, algae, and bacteria [3–13]. Among these, algae seem to be a promising organism for treating polluted waters [14–18]. This is partly due to high efficiency, surface area, and binding affinity as well as free from nutrient requirement [1, 19, 20]. In addition, the use of algae has several advantages such as the production of valuable products such as biofuel, proteins, carbohydrates, pigments, and vitamins during the treatment of polluted water [21, 22]. Moreover, algae has also the capability to capture carbon dioxide (CO_2) during photosynthesis, leading to the decrease in greenhouse gas emission [23, 24].

The pollutant uptake by algae is commonly associated with two stages, which are biosorption and absorption [25]. Biosorption is commonly characterized by the rapid adsorption behaviors and ion exchange processes as well as commonly occurs at surface of cells. Hence, this mechanism can occur in both living and non-living cells, but in some cases, non-living cells are more effective to adsorb toxic pollutants. When the uptake is controlled by absorption mechanism, the pollutant removal is slower due to the metabolism dependent activity in living cells. Therefore, it has been widely reported that after biosorption and absorption, toxic pollutants can also be degraded to the less toxic chemicals facilitated by enzymes [26–32]. For this process, the capability of algae to adsorb, absorb, and degrade toxic pollutants depends highly on algal species, composition of cell wall, as well as enzyme systems as illustrated in Fig. 1.

The confirmation of pollutants degradation by using algae has been reported in literature, and their metabolic pathways

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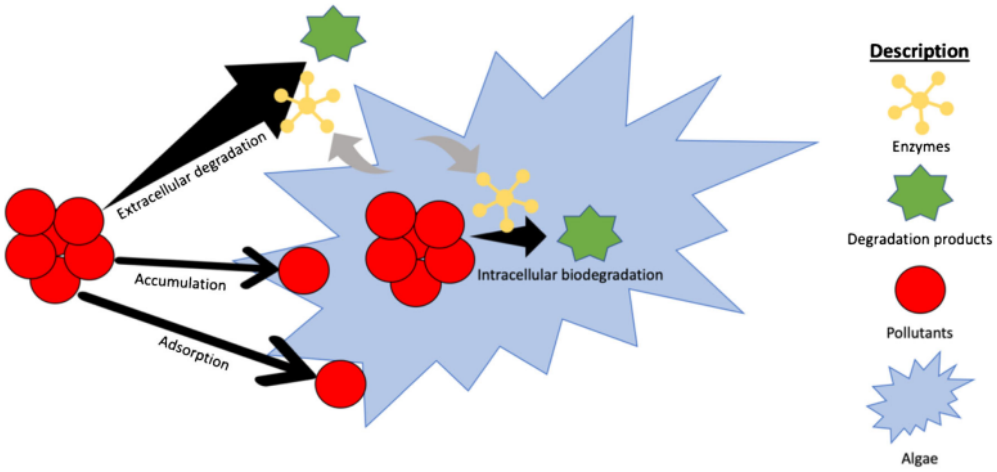

Fig. 1 Mechanism of pollutants removal by algae

have also been proposed as shown in Fig. 2 [33–35]. For instance, *Selenastrum capricornutum* can transform ethinylestradiol (EE) to three products (see Fig. 2) [33]. Alternatively, EE can also be transformed to 17α-ethinyl-1,4-estradien-10,17β-diol-3-one by using *Scenedesmus quadricauda*. It is also possible to transform EE to 3-β-d-glucopyranosyl-6β-hydroxyethinyl estradiol by employing *Ankistrodesmus braunii*. In general, the transformation process can be controlled by single or combination of a series of mechanisms. For example, the hydrogenation is the main mechanism to transform climbazole to climbazole-alcohol (CBZ-OH) by *Scenedesmus obliquus* [36]. Alternatively, a series of mechanisms was responsible for various pollutants degradation such as progesterone and norgestrel by *Scenedesmus obliquus* and *Chlorella pyrenoidosa* [37], naproxen by *Cymbella* sp. and *Scenedesmus quadricauda* [38], and galaxolide (HHCB) by *Navicula* sp. and *Scenedesmus quadricauda* as summarized in Table 1 [34, 36–43]. For a comprehensive overview, biotransformation mechanism of various pollutants by different algal species is listed in Table 1.

Considering the significance of this topic particularly for pollutants remediation using algal-based technology, this paper reviews the evidence for highlighting the effectiveness of the use of living or non-living cells for treating polluted waters. Although numerous studies have been conducted for the evaluation of non-living or living algal cells for treating polluted waters, this paper focuses only on studies that conducted the direct comparison between living and non-living cells. This is carried out in order to reduce uncertainties in the literatures on the use and performance of similar algal species and pollutants reporting contrary results because algal performance also depends on the environmental conditions and experimental procedures.

Removal Performance

It is useful to start the discussion by presenting the removal performance of living and non-living algae to remove pollutants since this indicator is very crucial for designing bioremediation technology. Direct comparison of living and non-living algal cells for the remediation of toxic pollutants was initiated about 4 decades ago by Sakaguchi et al. [44] who evaluated the bioaccumulation of cadmium in *Chlorella regularis*. Eleven years later after the investigation by Sakaguchi et al. [44], Maeda et al. [45] also reported that *Chlorella vulgaris* in the living and non-living cells had the capability to remove zinc and cadmium. In general, their study proved that the pollutant uptakes can be enhanced when the non-living cells were used.

Twelve years later, several algal species were investigated by Tam et al. [46] for the removal of tributyltin (TBT) as listed in Table 2. By using the living cells, the removals of TBT can be achieved up to 46, 42, 72, and 92% after 3 days for *Chlorella miniata*, *Chlorella sorokiniana*, *Scenedesmus dimorphus*, and *Scenedesmus platydiscus*, respectively. When the non-living cells were used, the corresponding removal percentages can be enhanced to 77, 90, 92, and 99%. Moreover, the most significant in terms of percentage removal can be achieved using *Scenedesmus platydiscus*. Another study investigated the capability of living and non-living *Chlorella vulgaris* for the removal of Cu(II) from water environment [55]. By using the distilled water, the removal of Cu(II) by 96.8 and 95.2% can be achieved using the non-living and living cells, respectively. However, when using the natural waters collected from various sources, a lower removal efficiency was found by 61.57 and 85.13 % for the living and non-living cells.

The removal of Cr (VI) using *Phaeodactylum tricornutum* and *Navicula pelliculosa* was examined [57]. The study

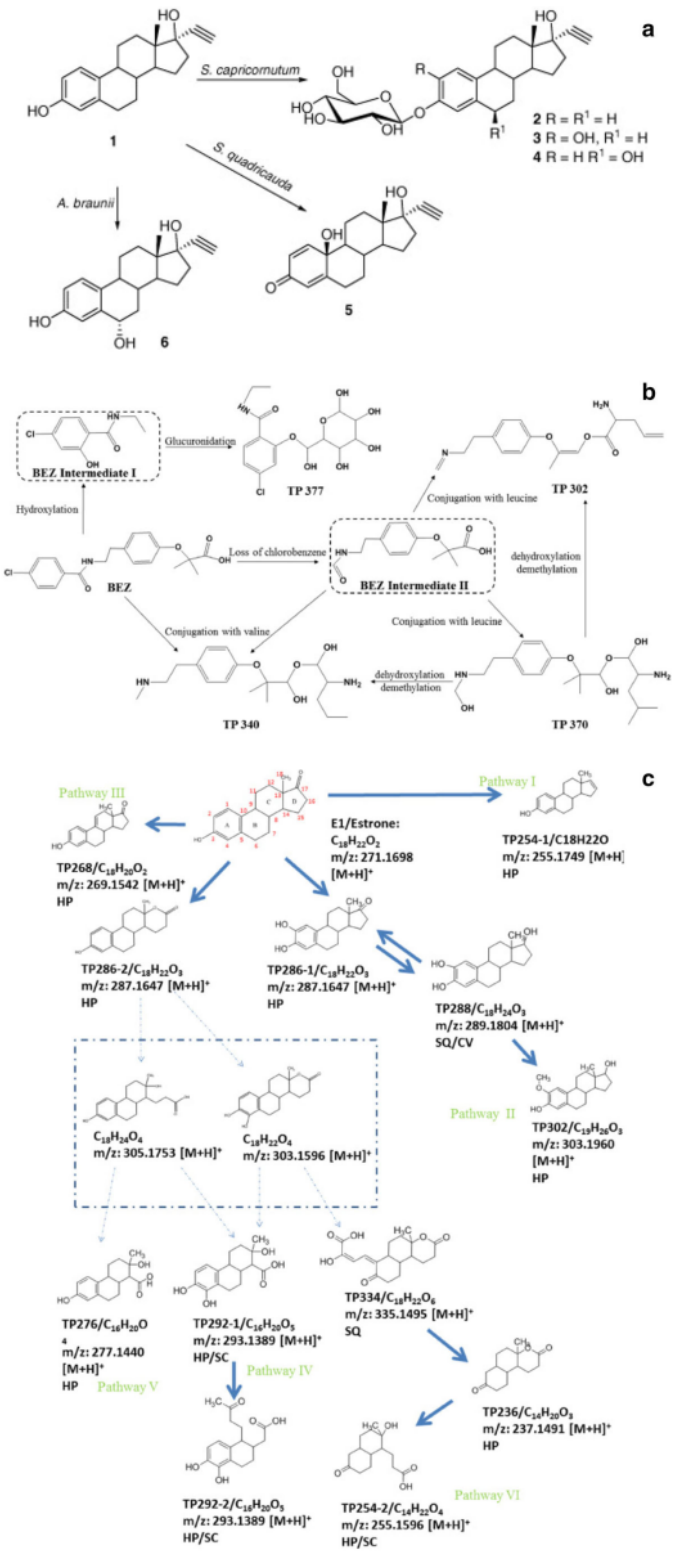

Fig. 2 Proposed biodegradation pathways of **a** ethinylestradiol by different microalgae [33], **b** bezafibrate by *Navicula* sp. [34], and **c** estrone by different microalgae [35]. All figures are adapted with permission

Table 1 Biotransformation mechanism of various pollutants by different algal species

Organism	Type of pollutant	Transformation mechanism	Reference
Scenedesmus obliquus	Climbazole	Hydrogenation	Pan et al. [36]
Scenedesmus obliquus and *Chlorella pyrenoidosa*	Progesterone and norgestrel	Hydroxylation, reduction, oxidation, and side chain breakdown	Peng et al. [37]
Cymbella sp. and *Scenedesmus quadricauda*	Naproxen	Hydroxylation, decarboxylation, demethylation, tyrosine conjunction, and glucuronidation	Ding et al. [38]
Navicula sp. and *Scenedesmus quadricauda*	Galaxolide (HHCB)	Hydroxylation, methoxylation, methylation, ketonization, demethylation, and oxaloacetate conjunction	Ding et al. [39]
Tetraselmis suecica	Benzalkonium chloride	Hydroxylation and dehydration	Jaén-Gil et al. [40]
Chlorella pyrenoidosa	Sulfamethoxazole (SMX)	Oxidation, hydroxylation, formylation, side chain breakdown, and pterin-related conjugation	Xiong et al. [41]
Picocystis sp. and *Graesiella* sp.	Diclofenac	Dehydration and dechlorination	Ouada et al. [42]
Sargassum horneri and *Pyropia yezoensis*	Arsenic	Reduction and methylation	Mamun et al. [43]
Navicula sp.	Bezafibrate	Hydroxylation, demethylation, and glucuronidation	Ding et al. [34]

observed that the non-living *Phaeodactylum tricornutum* can remove Cr (VI) by 32% but only achieve 24% when using the living cells. For *Navicula pelliculosa*, the removals of Cr (VI) using the non-living and living were 37 and 27%, respectively, indicating an improvement in the removal capability when the non-living cells were used. *Chlorella pyrenoidosa* was evaluated to remove methylene blue (MB) from the textile wastewater [54]. Maximum dye removals by

Table 2 The use of living and non-living algae for removal of various pollutants

Algae	Type of pollutant	Contents	Reference
Chlorella regularis	Cd	Cadmium uptake investigation	Sakaguchi et al. [44]
Chlorella vulgaris	Zn and Cd	Zinc and cadmium uptake investigation	Maeda et al. [45]
Chlorella vulgaris	Ni and Cu	Kinetic, isotherm, and pretreatment studies	Mehta and Gaur [47]
Chlorella miniata, *Chlorella sorokiniana*, *Scenedesmus dimorphus*, and *Scenedesmus platydiscus*	TBT	Removal and degradation studies	Tam et al. [46]
Chlorella vulgaris	BPA	Removal and parametric effects studies	Peng et al. [48]
Spirulina sp.	Cr^{3+}, Ni^{2+}, Cu^{2+}, and Cr^{6+}	Kinetic, isotherm, FTIR, fluorescence microscopy, and SEM studies	Doshi et al. [49]
Spirulina sp.	Cd^{2+}	Kinetic, isotherm, FTIR, and SEM studies	Doshi et al. [50]
Nitzschia hantzschiana, *Chlorella vulgaris*, *Chlamydomonas sajao*, and *Anabaena cylindrica*	Aniline	Removal and parametric effects studies	Wang et al. [51]
Chlamydomonas reinhardtii	Cu(II) and Pb(II)	Biosorption and bioaccumulation studies	Flouty and Estephane [52]
Selenastrum capricornutum	PAHs (BaA, BbF, BkF, BaP, DA, BghiP, and IP)	Removal and metabolites studies	Luo et al. [53]
Chlorella pyrenoidosa	MB	Kinetic, isotherm, removal studies	Pathak et al. [54]
Chlorella vulgaris	Cu(II)	Kinetic, isotherm, removal studies	Cheng et al. [55]
Chlorella vulgaris	Flutamide	Kinetic, isotherm, and optimization studies	Habibzadeh et al. [56]
Phaeodactylum tricornutum and *Navicula pelliculosa*	Cr (VI)	Parametric effects studies	Hedayatkhah et al. [57]
Parachlorella kessleri	Silver	Removal studies	Sedláková-Kaduková and Pristaš [58]

the non-living cells for the initial dye concentrations of 10, 20, 40, and 60 mg/L were 95.92, 90.46, 76.83, and 56.64%, respectively. By using the living cells, the maximum dye removals for the initial dye concentrations of 10, 20, 40, and 60 mg/L were lower to be 83.17, 77.25, 62.65, and 41.72%, respectively. For another dye removal, the photodegradation of bisphenol A (BPA) in simulated lake water using *Chlorella vulgaris* was carried out using living and non-living cells [48]. The removal of BPA using the living cells was only 37% in 4 days. However, an improvement was achieved by using the non-living cells by achieving the removal up to 49% in 4 days.

The degradation of aniline was investigated using four algal species under metal halide light as tabulated in Table 2 [51]. The study found that the removals of aniline were 87% (non-living) and 69% (living), 88% (non-living) and 68% (living), 89% (non-living) and 82% (living), and 84% (non-living) and 68% (living) by *Nitzschia hantzschiana*, *Chlorella vulgaris*, *Chlamydomonas sajao*, and *Anabaena cylindrica*, respectively. Removal of Cu(II) and Pb(II) by *Chlamydomonas reinhardtii* was conducted by Flouty and Estephane [52]. For Cu(II), the removal efficiency of the non-living cells can be improved about 2 times than the living cells (55 and 28%). As a comparison, the removal efficiencies of Pb(II) using the non-living cells and living cells were 40 and 8%, respectively. It has been proposed that amino and hydroxyl groups are responsible for lead removal since they have capability to combine intensively with lead, but the amino groups are responsible for the removal of copper [59]. Removal of silver by freshwater green alga *Parachlorella kessleri* was investigated [58]. When the non-living cells were used for silver removal, up to 75% can be removed from the solution. However, when the living cells were employed, the removal was decreased to 68%.

Inconsistency was observed when living and non-living *Selenastrum capricornutum* for the removal of some PAHs such as benz[a]anthracene, benzo[b]fluoranthene, benzo[k]fluoranthene, benzo[a]pyrene, dibenzo[a, h]anthracene, benzo[g, h,i]perylene, and indeno[1,2,3-c, d]pyrene abbreviated as BaA, BbF, BkF, BaP, DA, BghiP, and IP under light irradiation [53]. Under white light irradiation, the removal of DA, BghiP, BaA, and BaP using the living cells were 13.3, 0.8, 77.7, and 59.5% after 4 days, which are lower than those using the non-living cells by 25.4, 31.9, 90.9, and 90.2%, respectively. Similar observation was also reported by Habibzadeh et al. [56] when living and non-living *Chlorella vulgaris* were used to remove flutamide (FLU) from wastewater. Using the non-living alga, the FT can be removed up to 97.9%. In addition, up to 98.5% of the flutamide can be removed when the living algal cells were employed. Table 3 summarizes a comprehensive overview and percentage removal comparison between non-living and living algae for different pollutants. The above discussion on the basis of the removal performance has clarified that the capability of algae to remove pollutants are quite specific based on the type of algae, type of pollutants, environmental chemistry, and experimental procedure. In general, the use of non-living cells has been more attractive in terms of the removal performance.

Biotransformation of pollutants using algae has also been investigated. The degradation of TBT to di- and monobutyltin (DBT and MBT) was observed due to a stepwise enzymatic debutylation mechanism as reported by Tam et al. [46]. The study confirmed that the algae can transform TBT to DBT and MBT within 14 days. Specifically, *Chlorella miniata*, *Chlorella sorokiniana*, *Scenedesmus dimorphus*, and *Scenedesmus platydiscus* produced 11.6% DBT and 38.6 % MBT, 7.3% DBT and 41.3% MBT, 10.2% DBT and 11.1% MBT, and 0.4% DBT and 3.0% MBT, respectively. It is interesting to note that the investigated algal cells can degrade TBT depending on the capability of the TBT debutylating enzymes. Moreover, transformation from BaA to BaA-7,12-dione and from BaP to BaP-1,6-dione and BaP-6,12-dione can be facilitated by *Selenastrum capricornutum* [53].

Biosorption Capacity

The accumulation of cadmium in *Chlorella regularis* was investigated at different pH (3–7) and temperatures (0–30 °C) [44]. Increase in pH can improve the uptake of cadmium by both living and non-living cells. The uptake of the cadmium at the highest evaluated pH (7) by the living and non-living cells were 4700 and 5766 μg/g, respectively. At the maximum temperature (30 °C), the uptakes of cadmium by the living and non-living cells were 1989 and 4856 μg/g, respectively. Another investigation by using *Chlorella vulgaris* for the uptake of zinc and cadmium in *Chlorella vulgaris* was carried out [45]. The uptake of cadmium in living and non-living cells was 11.7 and 16.1 × 10^3 mg/kg after 18 days, respectively. The finding exhibited that the use of the non-living cells can enhance (1.4 times) the uptake of cadmium. However, a contrary finding was observed when the living and non-living cells were tested for the removal of zinc. The study observed that the uptakes of zinc in living and non-living cells were 6.6 and 4.7 × 10^3 mg/kg, respectively, after 11 days [45].

Study on the removal of Ni and copper Cu using living and non-living *Chlorella vulgaris* was investigated by Mehta and Gaur [47]. Biosorption capacity at equilibrium (q_e) of the non-living cells for the removal of Ni (10 mg/L pollutant concentration) was 9.00 mg/g but for the living cells was only 7.14 mg/g. For the Cu (10 mg/L pollutant concentration) removal, the corresponding biosorption capacities were 14.45 and 8.61 mg/g, respectively. In addition, the maximum biosorption capacities (q_{max}) of the non-living and living cells for Ni removal were 20.23 ± 1.03 and 14.10 ± 0.58 mg/g, respectively, and for Cu removal were 14.48 ± 0.70 and 10.12 ± 0.41 mg/g, respectively.

Table 3 Percentage removal and biosorption capacity comparison between non-living and living algae for different pollutants

Type of algae	Experimental procedures	Pollutant	Percentage removal (%)		q_{max} (mg/g)		Reference
			Living	Non-living	Living	Non-living	
Chlorella miniata	TE = Batch study ED = up to 14 days	TBT	46	77	-	-	Tam et al. [46]
Chlorella sorokiniana	TE = Batch study ED = up to 14 days	TBT	42	90	-	-	Tam et al. [46]
Scenedesmus dimorphus	TE = Batch study ED = up to 14 days	TBT	72	92	-	-	Tam et al. [46]
Scenedesmus platydiscus	TE = Batch study ED = up to 14 days	TBT	92	99	-	-	Tam et al. [46]
Chlorella vulgaris	TE = Batch study ED = up to 150 mins	Cu(II)	95.2	96.8	16.34	16.65	Cheng et al. [55]
Phaeodactylum tricornutum	TE = Batch study ED = up to 3 days	Cr (VI)	24	32	-	-	Hedayatkhah et al. [57]
Navicula pelliculosa	TE = Batch study ED = up to 3 days	Cr (VI)	27	37	-	-	Hedayatkhah et al. [57]
Chlorella pyrenoidosa	TE = Batch study ED = up to 60 mins	MB	83.17	95.92	20.8	21.3	Pathak et al. [54]
Chlorella vulgaris	TE = Photochemical reactor ED = up to 240 mins	BPA	37	49	-	-	Peng et al. [48]
Nitzschia hantzschiana	TE = Photochemical reactor ED = up to 300 mins	Aniline	69	87	-	-	Wang et al. [51]
Chlorella vulgaris	TE = Photochemical reactor ED = up to 300 mins	Aniline	68	88	-	-	Wang et al. [51]
Chlamydomonas sajao	TE = Photochemical reactor ED = up to 300 mins	Aniline	82	89	-	-	Wang et al. [51]
Anabaena cylindrica	TE = Photochemical reactor ED = up to 300 mins	Aniline	68	84	-	-	Wang et al. [51]
Chlamydomonas reinhardtii	TE = Batch study ED = up to 370 mins	Cu(II)	28	55	-	-	Flouty and Estephane [52]
Chlamydomonas reinhardtii	TE = Batch study ED = up to 370 mins	Pb(II)	8	40	-	-	Flouty and Estephane [52]
Parachlorella kessleri	TE = Batch study ED = up to 4 days	Ag	68	75	-	-	Sedláková-Kaduková and Pristaš [58]
Chlorella vulgaris	TE = Batch study ED = up to 120 min	FLU	98.5	97.9	12.5	28.8	Habibzadeh et al. [56]

It is noted that TE and ED refer to the type of experiments and experimental duration, respectively

A study on the MB removal from simulated textile wastewater by *Chlorella pyrenoidosa* was evaluated using the living and non-living cells and exhibited different biosorption capacities [54]. The q_e (at 60 mg/L dye concentration) of the non-living cells was 31.1 mg/g but 16.6 mg/g for the living cells. The value of q_{max} of the non-living cells was 21.3 mg/g and 20.8 mg/g for the living cells. The values of q_{max} for the removal of Cu(II) using living and non-living *Chlorella vulgaris* were 16.34 and 16.65 mg/g [55].

Inconsistency can be observed when *Spirulina* sp. was used for the removal of Cr^{3+}, Ni^{2+}, Cu^{2+}, and Cr^{6+} proposed by Doshi et al. [49]. The non-living cells performed q_{max} by 167, 515, 100, and 143 mg/g for the removal of Cr^{3+}, Ni^{2+}, Cu^{2+}, and Cr^{6+}, respectively. The corresponding values for the living cells were 304, 1378, 389, and 333 mg/g. In a different study, Doshi et al. [50] reported a consistent finding when Cd^{2+} was used as a pollutant model. The study found that the q_e of the *Spirulina* sp. for the removal of Cd^{2+} were 601 and 313 mg/g for the living and non-living cells, respectively, and the q_{max} values were 625 and 355 mg/g for the living and non-living cells, respectively. Alternatively, similar observation on the biosorption capacity of living and non-living *Chlorella vulgaris* to remove FLU from wastewater was reported by Habibzadeh et al. [56]. It was observed that the q_e (at the drug concentration of 100 μM) of the non-living cells was 2.87 mg/g, but an improvement can be obtained when living cells were used (3.07 mg/g). In addition, the study also found that the living alga had q_{max} of 28.8 and 12.5 mg/g for the non-living cells.

In general, the above discussion presented on the performance of some algae for the removal of various organic and metal pollutants. This paper has shown that the properties of q_e and q_{max} are quite specific depending highly on the type of algae and pollutants. In the case of organic pollutants, study

by Pathak et al. [54] reported better performance of the non-living compared to living cells but the study by Habibzadeh et al. [56] found their contradiction. In the case of metal pollutants, several studies agreed that the non-living outperformed compared to living cells [44, 45, 47, 55], but other studies reported the opposite findings [49, 50].

Increase in adsorption capacity for the non-living algal cells compared to living cells to remove some pollutants as reported by Mehta and Gaur [47] indicated that the non-living alga can be used more efficiently than the living algal cells. In addition, the value of *n* was also found to be enhanced for the non-living algal cells than the living cells. The non-living cells, for instance, by oven drying can cause cell membrane disruption, leading to the enhancement of binding between sorbate and sorbent via intracellular binding [47]. In addition, the non-living cells are settled relatively faster than living cells because they can form aggregates, which can make easier for the separation of the cells from solution. The improving sorption capacity for the non-living cells can also be suggested by the slight improvement of specific surface area as estimated by Pathak et al. [54] by using *Chlorella pyrenoidosa* as the algal model. The study found that the non-living and living cells had the specific surface areas of 67.6 and 66.0 m^2/g, respectively. Although the specific surface area was not statistically significant, it can possibly contribute for sorption efficiency improvement as reported by several studies [54, 60].

In the cases of higher sorption capacity by living cells, the possible explanation is as follows. Increase in the uptakes of metals (Cr^{3+}, Ni^{2+}, Cu^{2+}, and Cr^{6+}) by living *Spirulina* sp. were because of their metal transport via the cell, which can be facilitated by enzymes present in the cells surface. For this mechanism, functional groups of intercellular are responsible for the adsorption [49]. For the non-living cells, the study exhibited that there were no enzymatic activities observed like in the cells. It was also hypothesized that the binding of Cd^{2+} with the carboxylic and phosphate group and to lesser extent the amino group present in *Spirulina* sp. is responsible for the Cd^{2+} uptake in living and non-living *Spirulina* sp. as observed via infrared (IR) spectroscopy [50]. The uptake of the metal ions was then observed via surface morphology analysis using the SEM that confirmed the increase in the width of the filament of living and non-living cells to be in the range of 5 to 25 μm (before adsorption = 2 to 5 μm) and 3 to 10 μm (before adsorption = 1.4 to 3.7 μm), respectively. A greater width of the filament of living after adsorption of Cd^{2+} indicated the improvement of adsorption capability.

Kinetic Behavior

Kinetic behaviors of removal of Ni and Cu using living and non-living *Chlorella vulgaris* was investigated by Mehta and Gaur [47] and modeled using the first- and second-order models. The kinetic studies were conducted using batch studies with 100-ml cell suspension having 10 mg dry weight and initial pollutant concentration of 10 mg/L at 30 rpm and 25 °C. The study found that all kinetic data can be best described by the second-order than the first-order model. By evaluating two models, the kinetic adsorption data of non-living *Spirulina* sp. for the removal of Cr^{3+}, Ni^{2+}, and Cu^{2+} fitted well using the pseudo first-order (PFO) model but followed the pseudo second-order (PSO) model for Cr^{6+} removal [49]. The experiment was conducted via batch studies with the use of dry biomass of 0.3 g and initial pollutant concentration ranging from 0.05 to 0.5 g/25 mL. In the living form, the kinetic uptake of all pollutants by living *Spirulina* sp. agreed well with the PSO model. In another study by using the same two models, the kinetic sorption of Cd^{2+} by using the *Spirulina* sp. fitted well with the PSO than the PFO model [50]. The study conducted batch studies for evaluating the kinetic behaviors and tested at initial pollutant concentration ranging from 0.05 to 0.5 g/25 mL.

Removal of MB in simulated textile wastewater by *Chlorella pyrenoidosa* using the living and non-living cells followed the PSO model than the PFO model [54]. The kinetic mechanism of living and non-living *Chlorella vulgaris* for the removal of Cu(II) was modeled using three models, which are the PFO, PSO, and Elovich models [55]. The removal using the living cells can be well described using the PFO, but for the non-living, the kinetic behaviors fitted well with the PSO, as judged by low mean squared error (MSE) and R^2. By employing six kinetic models, which are the PFO, PSO, intraparticle diffusion (IPD), modified Freundlich, Sigmoidal Chapman (SC), and Elovich, the kinetic behaviors of living and non-living *Chlorella vulgaris* to remove FLU from wastewater followed the modified Freundlich model [56]. This suggested that the removal of FLU by living and non-living cells can be characterized as the heterogeneous adsorption [56]. It has been proposed that functional groups present on the cell surface are responsible for the biosorption mechanism [61]. It is noted that the Elovich equation is the common model used for the description of pollutants adsorption via chemisorption mechanism. The study exhibited that Elovich kinetic model did not perform well the data characteristics, suggesting that the mechanism is not a chemisorption.

Kinetic behaviors of oxytetracycline (OTC) removal by living and non-living *Phaeodactylum tricornutum* showed different patterns judged by nine statistical error analyses [62]. Batch studies were conducted with the biomass concentration used in the study was 0.4 g/L of dry biomass and tested at the initial concentration of 2.5, 5, 7.5, 10, 12.5, or15 mg/L of OTC. The study investigated the kinetic behaviors using four models, which are the PFO, PSO, IPD, and SC models. It was found that the experimental kinetic data followed the sigmoidal kinetic model compared to other evaluated models. Interestingly, the removal of OTC using the non-living cells followed the PFO model.

It is interesting to note in this review that the common powerful kinetic model such as the PSO did not show the best accuracy on the basis of statistical error analyses. This is crucial since common studies in the kinetic investigation that only used R^2 as the indicator to evaluate the performance of a model has limitation. Basically, this indicator (R^2) partially measures goodness of fit how the data points are close with the predicted model [63]. Therefore, additional statistical indicators are needed to judge the model performance as proposed by Santaeufemia et al. [62] by using nine statistical error analyses. In the future, the selection of the best kinetic model for the removal of pollutants is highly recommended not only based on R^2 but also the combination with other error analyses as proposed by Santaeufemia et al. [62] and proposed by other studies [64–66].

Isotherm Behavior

It is useful to evaluate isotherm behaviors of pollutant adsorption using some mathematical models. Several existing isotherm models can also be used for describing the possible mechanism. The usefulness of existing models can be presented as follows. The use of the Langmuir isotherm model can be very useful since it has a constant that describe the dimensionless separation factor (RL) [67–70]. The RL describes the type of isotherm such as unfavorable, favorable, linear, and irreversible if RL > 1, 0 < RL < 1, RL = 1, and RL = 0, respectively. Alternatively, the Freundlich isotherm model is also useful for explaining the mechanism [62, 71, 72]. The value of 1/n in the model shows heterogeneity factor, which suggests that a smaller value (n = 1 to 10) exhibits more favorable by sorption process. In addition, this also shows the strong interaction between sorbate and sorbent and indicates more heterogeneous material surface [54]. Temkin isotherm model also provides a useful constant, b_T, which describes heat of sorption in the unit of J/mol. The mechanism is defined as exothermic and endothermic reaction if the value of $b_T > 1$ and $b_T < 1$, respectively [73–75]. Moreover, the nature of adsorption can also be predicted using the Dubinin-Radushkevich isotherm model by estimating the E_D (apparent energy of sorption) value. The E_D between 8 and 16 kJ/mol and below 8 kJ/mol indicates the chemisorption and physical sorption processes, respectively.

Isotherm behaviors of Ni removal using living and non-living *Chlorella vulgaris* fitted well with the Freundlich model compared to the Langmuir model, judged by R^2 ranging from 0.87 to 0.88 for the Langmuir model and 0.98 to 0.99 for the Freundlich model [47]. Similar finding was also observed for the removal of Cu using both living and non-living algae [47]. By providing R^2 ranging from 0.72 to 0.78 and 0.98 to 0.99 for the Langmuir and Freundlich models, respectively, the isotherm data can be well predicted using the Freundlich model. Moreover, the values of 1/n for the Ni removal using the non-living and living cells were 0.39 and 0.45, respectively. For the Cu removal, the corresponding values were 0.19 and 0.27, indicating the lower value for non-living than living cells. The removal of MB dye in simulated textile wastewater by *Chlorella pyrenoidosa* exhibited to follow the Langmuir isotherm model [54]. Moreover, the shape of isotherm was analyzed by estimating the RL. The study found that the values of RL were in the range of 0.02 to 0.1 and 0.08 to 0.3 for the non-living and living cells, respectively. In addition, the value of 1/n for the non-living cells was 0.32 and for living cells was 0.38. The lower 1/n value of the non-living cells showed more favorable adsorption than the living cells and indicated strong bond between the sorbate and sorbent.

The antibiotic, oxytetracycline (OTC) removal by living and non-living cells of *Phaeodactylum tricornutum* were investigated using four isotherm models [62]. The study found that the isotherm data for the living and non-living cells can be best described in the following order: Langmuir > Temkin > Freundlich > Dubinin-Radushkevich. In addition, the RL values found in the study ranged from 0 to 0.5. Specifically, RL decreased from 0.130 to 0.024 in the case of living cells at the initial pollutant concentration from 2.5 to 15 mg/L. For the non-living cells, the RL values varied from 0.482 to 0.134 for the corresponding initial pollutant concentration. This indicates that the adsorption is more favorable at higher initial pollutant concentration. It is also interesting to note that the RL values for living cells were closer to 0, which suggested the possible irreversible mechanism. The values of 1/n (non-living and living: 0.35 and 0.39) obtained in this study indicated favorable sorption. The b_T values were positive for both living (391.52 J/mol) and non-living cells (2329.81 J/mol), which indicated that the reaction was exothermic. Increase in the b_T value for the non-living cells exhibited the more exothermic process. This can be explained because the adsorption of OTC in living cells consumes energy. The values of E_D for the non-living and living cells were 0.95 and 3.26 kJ/mol, indicating the physical process controlling the mechanism.

Alternatively, isotherm data for the removal of Cu(II) using living and non-living *Chlorella vulgaris* for the removal of Cu(II) was modeled using four models, which are the Langmuir, Freundlich, Sips, and Khan models [55]. By evaluating the MSE and R^2, the isotherm data followed the Sips model. In addition, the values of 1/n for the removal of Cu(II) using living and non-living cells were 0.71 and 0.76, which is not significantly different. The isotherm behaviors of living and non-living *Chlorella vulgaris* to remove FLU followed the Freundlich model and then followed by the Langmuir, Temkin, and Dubinin-Radushkevich [56]. The 1/n values of living and non-living cells were in the range of 0 to 1, which indicated that the adsorption is favorable. The RL values for living and non-living cells were 0.054 and 0.064. The b_T values were also positive for both cells (non-living and living:

1383.54 and 382.59 J/mol) and exhibited more exothermic for the non-living cells by providing a greater b_T value. Moreover, the values of E_D for the non-living (0.48 kJ/mol) and living cells (0.66 kJ/mol) were below 8 kJ/mol, suggesting that the biosorption is a physical process.

For the evaluation of the isotherm characteristics, the use of *Spirulina* sp. for the removal of Cr^{3+}, Ni^{2+}, Cu^{2+}, and Cr^{6+} was carried out by Doshi et al. [49]. It was found that the adsorption using the living and non-living cells followed the Freundlich model. A consistent finding was also achieved when the algae was tested for another pollutant removal in another investigation [50]. By using two isotherm models, the isotherm sorption of Cd^{2+} by using the *Spirulina* sp. in the form of living and non-living cells can be described well using the Freundlich model compared to the Langmuir model. The study also found that the value of 1/n for the living cells was 0.4 and near 1 for the non-living cells.

Based on the aforementioned knowledge, it is interesting to discuss all findings in the basis of RL, 1/n, b_T, and E_D. In the case of *Chlorella pyrenoidosa* to remove MB, the values of RL ranged from 0 to 1, suggesting the favorable adsorption with the lower values were found for the non-living cells [54]. At the highest tested initial FLU concentration (60 mg/L), the values of RL were closer to zero (0.02 and 0.08: non-living and living), indicating the irreversible adsorption, which was also similarly observed by Santaeufemia et al. [62] when living and non-living cells of *Phaeodactylum tricornutum* were used for OTC removal. In addition, the removal of FLU by *Chlorella vulgaris* was also considered as irreversible adsorption because the RL values were closer to zero [56]. Based on the value of 1/n, all abovementioned studies indicated the beneficial adsorption and strong bonding between sorbate and sorbent.

Moreover, only two studies comprehensively analyzed for the values of b_T and E_D *as reported by* Habibzadeh et al. [56] and Santaeufemia et al. [62]. For the study by Santaeufemia et al. [62], the b_T values were positive for both living and non-living cells, which indicated exothermic reaction with a superior b_T value for the non-living cells. The values of E_D for the non-living and living cells were below 8 kJ/mol, indicating the physical process. For the study by Habibzadeh et al. [56], the b_T values were also positive for both cells and exhibited more exothermic for the non-living cells by providing a higher b_T value. Moreover, the values of E_D for the non-living were also below 8 kJ/mol, suggesting that the biosorption is a physical process. In general, the aforementioned studies exhibited that the removal of pollutant by using living and non-living algae was exothermic reaction and controlled by physical process.

General Perspectives

Several studies have agreed that the use of non-living cells is more preferable because of the following reasons. Non-living cells are commonly not affected by the toxic pollutant, and they can be handled easily [76]. In addition, the use of non-living cells can eliminate the need for nutrients and can be used for many cycles [53]. It is also possible to store non-living cells for extended periods at room temperature without the need for purification [47]. However, it is also potential to explore the living cells particularly when the toxicity of the pollutant is relatively low or is in a pollutant concentration which does not cause the total inhibition. For this particular case, the use of living cells seems to be more promising since they can retain their activity for more storing a pollutant or for transforming a pollutant to less toxic chemicals. This probably can improve the removal efficiency since the removal mechanism is not only controlled by adsorption and absorption but also by biotransformation [77–82].

It is therefore important to select between living and non-living algae for the cleanup of polluted water. This selection should be based on the basis of percentage removal and sorption capacity of pollutants. The above discussion has highlighted that non-living algae are more effective on the basis of percentage removal as reported by Tam et al. [46] for the removal of TBT, Cheng et al. [55] for the removal of Cu(II), Hedayatkhah et al. [57] for the removal of Cr (VI), Pathak et al. [54] for the removal of MB dye, Peng et al. [48] for the removal of BPA, Wang et al. [51] for the removal of aniline, Flouty and Estephane [52] for the removal of Cu(II) and Pb(II), Sedláková-Kaštánková and Pristaš [58] for the removal of silver, and Luo et al. [53] in the case of removal of DA, BghiP, BaA, and BaP. In contrast, only studies reported by Habibzadeh et al. [56] for the removal of FLU from wastewater and by Luo et al. [53] in the case of removal of BbF, BkF, and IP exhibited the best removal efficiency can be achieved when living cells were used. Specifically, although lower removal efficiency reported by Habibzadeh et al. [56], the difference was not significant as only by 97.9 and 98.5% for the non-living and living cells, respectively, which is very small (0.8%). In the case of removal of BbF, BkF, and IP reported by Luo et al. [53], the removals of BbF, BkF, and IP using living *Selenastrum capricornutum* were up to 3.0, 1.8, and 1.2 times higher than the non-living cells. Considering the aforementioned basis (percentage of removal), it is suggested that the use of non-living cells was more efficient for a wide range of pollutants.

Secondly, the selection is based on the sorption capacity. Increase in the adsorption capacity has been shown for non-living cells as reported by Sakaguchi et al. [44] for the removal of cadmium, Maeda et al. [45] for the removal of zinc and cadmium, Mehta and Gaur [47] for the removal of Ni and Cu, Pathak et al. [54] for the removal of MB, and Cheng et al. [55] for the removal of Cu(II). A contrary finding was observed only by Doshi et al. [49] for the removal of Cr^{3+}, Ni^{2+}, Cu^{2+}, and Cr^{6+}, by Doshi et al. [50] for the removal of Cd^{2+}, and by Habibzadeh et al. [56] for the removal of FLU from

wastewater. It is noted that Doshi et al. [49] employed the same algae, *Spirulina* sp., and Habibzadeh et al. [56] used *Chlorella vulgaris* as algae model. Considering the sorption capacity, the use of non-living cells seems to be more efficient for a wide range of pollutants.

In the future, studies on the investigation of living and non-living algae are recommended to focus on the improvement of removal efficiency. For instance, the use of living and non-living *Phaeodactylum tricornutum* and *Navicula pelliculosa* for the removal of Cr (VI) can only achieve 24–32% and 27–37% [57]. Several strategies can be proposed for the improvement such as pretreatment methods or adjustment of physio-chemical parameters as carried out by several studies [51, 56, 57]. In addition, evaluation of capability of algae for the removal of pollutions in real water samples such as river water, wastewater, and lake water is crucial to be carried out to improve their practicality and applicability in real application in the field. This is because study conducted by Cheng et al. [55] found that the removal efficiency of *Chlorella vulgaris* for the removal of Cu(II) were much lower when the natural water was used compared to the distilled water.

This paper has shown that all lab-scale studies for the implementation of algal-based technology for the removal of environmental pollutions have been significant although their implementation in pilot scale or full-scale operations is still very limited. The Algal Turf Scrubber® (ATS) technology is an established algal-based technology designed and produced by HydroMentia, which is a water pollution control company, for treating wastewater [83]. A pilot scale implementation of ATS was tested using effluent water from a constructed wetland in the Everglades agricultural area and can remove the total phosphorus by 23% [84]. Moreover, a full-scale implementation of this technology with the treatment capacity of 15 million gallons per day (MGD) has been applied at Taylor Creek, Florida, which discharges to Lake Okeechobee, Florida, USA [84]. The technology can remove 92 g/m^2-yr of total phosphorus and 727 g/m^2-yr of total nitrogen in stormwater and can reduce the concentration of phosphorus by 900 g/m^2-yr of total phosphorus in wastewater. Moreover, several algal-based technologies such as the AlgaeWheel® system by OneWater and the revolving algal biofilm (RAB) system by Gross-Wen Technologies have been put into practice for wastewater remediation [85].

Moreover, harvesting microalgae from growth medium after removing pollutants is a necessary process and still challenging [86–90]. This is partly due to their small sizes and colloidal stability in growth medium. In general, methods for microalgae harvesting can be categorized into four, which are chemical, physical, biological, and magnetic with their specific advantages and disadvantages, which depend on the culture methods and product types of microalgae [32, 91]. The selection of harvesting method is not only by the harvesting efficiency but also by cost effectiveness, which are crucial particularly for large-scale application. Currently, magnetic nanoparticles (MNPs) were proposed for the harvesting of algae [92]. The study observed that the harvesting efficiency achieved 95% within 5 min and can be improved for the longer time. The estimated cost for the harvesting of microalga was USD 2.4 to 3.2 per one cubic meter of the algal medium. Interestingly, the costs can be reduced up to USD 0.45/m^3 by recycling the proposed MNPs for 5 times. Although the harvesting cost by MNPs seems to be competitive, alternative current study found that flocculation seems to be more promising in terms of cost reduction as reported by Labeeuw et al. [93]. The study evaluated the synthetic (polyacrylamides) and natural (chitosan) flocculants. It was estimated that the harvesting costs using polyacrylamides and chitosan were $0.04/m^3 and $0.80/m^3, respectively, suggesting that the cheapest price was provided when polyacrylamides were used compared to chitosan. Moreover, it is also interesting to note that the capability of the flocculation method was tested in a pilot scale (350 L photobioreactor) and exhibited a good performance by providing above 80% harvesting efficiency after 10 min, which has the potential for further large-scale application. After harvesting, microalgal biomass can be utilized for bioenergy production [94, 95] or it can also be explored for the production of health supplements, bioactive compounds, food additives, and biotechnology applications [96, 97].

It has been well known that most of microalgae species have the potential for biodiesel production because of high lipids contents ranging from 50 to 70% and can reach 80% in case of the *Botryococcus braunii* [98, 99]. Microalgae have capability to produce algal oil by 58700 L/hac, which can produce biodiesels by 121104 L/hac [100]. However, their productivity depends highly on environmental conditions such as the presence of pollutants. Fatty acid profiles are commonly used as potential indicators for production of biodiesel. The effects of pollutants on the lipid or fatty acid profiles of algae have been reported in the literatures. For instance, the effects of nonylphenol and bisphenol on the fatty acid of *Nannochloropsis salina* for biodiesel production were evaluated [101]. In the presence of nonylphenol and bisphenol, the percentages of saturated fatty acids were 41.06 and 38.73%, which were higher compared to the control (35.38%). The study also found that the presence of nonylphenol and bisphenol can increase the lipid contents by 135 and 139 mg/g cell dry weight compared to the control (96.4 mg/g cell dry weight). It is noted that biodiesel is composed mainly from palmitic acid (C16:0), stearic acid (C18:0), oleic acid (C18:1), and linolenic acid (C18:3) and increase in palmitic acid can be used as an indicator for perfect quality biodiesel. Study by Mohy El-Din [101] also ratified that the presence of nonylphenol and bisphenol can increase the palmitic acid of *Nannochloropsis salina*. It is noted that some algae can adapt at certain environmental pollutants concentration. In the future, more studies need to be conducted at higher pollutant

concentration and complex pollutant mixture since such condition can be commonly observed particularly in wastewater.

Conclusion

This paper has highlighted the use of living and non-living algae for the cleanup of polluted water. In general, the use of non-living algae was more effective as indicated by higher removal efficiency and higher adsorption capacity as well as easy to handle compared to living cells. This review also showed there is no single powerful model for the modeling kinetic and isotherm data for a wide range of pollutants and type of algae, indicating that the mechanism is quite specific depending on the type of algae, type of pollutants, environmental conditions, and experimental procedures. The removal of pollutants by living and non-living algae can be considered as an exothermic reaction and physical sorption. Further studies are needed to use living and non-living algae to remove pollutants from real water samples to confirm their practicality and applicability in field application.

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Declarations

Human and Animal Rights and Informed Consent This article does not contain any studies with human or animal subjects performed by any of the authors.

Conflict of Interest The authors declare no competing interests.

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