



Analyzing semiquantitative phytoplankton counts

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Abstract Semi-quantitative microscope counts of phytoplankton are often a compromise between time-consuming cell biomass analyses and no phytoplankton data. We demonstrate how semiquantitative data from a monitoring program can be used to study phytoplankton community composition, its annual cycle, and aspects of the ecosystem it inhabits. Semi-quantitative counts from Agmon Wetlands, Israel, collected monthly from 2008 to 2021, were generated by allocating a score from 1 (rare) to 6 (extremely abundant) to each taxon observed in a sample. Five samples could be analyzed at the time it takes to count one sample by the conventional Utermöhl method. Using an exponential regression equation, the scores were transformed to estimated concentrations (algal units/ml), then summed into taxonomic or other groups of species. A strong annual pattern of the sum of scores for each taxonomic group was observed. The method was useful for assessing ecosystem features based on indicator species, and for presenting the contribution of morpho-functional groups to the phytoplankton community.

If making a species list is planned, we recommend assigning scores, creating calibration curves, converting the scores to concentration estimates, and using those estimates to achieve higher resolution and better conclusions than possible with a species list alone.

Keywords Algal units · Bioindication · Functional groups · Microscope counts · Sum-of-scores

Introduction

An understanding of species composition and their relative contribution to a phytoplankton population is important for analyzing the functioning of aquatic food webs (Reynolds, 2006) as well as for water quality management (Suthers et al., 2019). The conventional methods for assessing phytoplankton species composition, abundance and biomass rely on cell counts and measurements under the inverted microscope (Utermöhl, 1958; Lund et al., 1958) followed by mathematical procedures to estimate biomass (Hillebrand et al., 1999). While these methods generate essential quantitative data for comparisons within and between systems, they are labor-intensive: it takes several hours of an expert to process a single sample. In recent years there have been growing attempts to replace the manual microscope work by automated systems, such as flow cytometer (Olson et al., 2018) and FlowCam (Poulton, 2016) that count the cells while collecting additional data on morphological

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and other parameters. But so far, in many cases these methods are not sufficiently developed to replace the human eye and have not yet replaced microscope counts in routine monitoring programs. Similarly, molecular methods such as metabarcoding, analyzing environmental DNA, do not yet replace microscope counts. The metabarcoding method is based on extraction, amplification and sequencing of a small DNA fragment found in the genomes of all organisms in the whole community. Hence species identification and abundance can be deduced from the DNA sequences obtained. However, technical biases, lack of taxa in reference libraries and of a conventional or suitable data analysis pipeline, hinder the use of such a powerful tool to replace microscopy on ongoing phytoplankton monitoring programs (Obertegger et al., 2020; Santi et al., 2021; Keck et al., 2022). Hence, microscope counts continue to be the consensus method for analyzing phytoplankton samples and is the standardized method required by the European Framework Directive (European Committee for Standardization, 2006).

In some cases, and to address limited objectives, semi-quantitative counts that are much less time consuming can be used instead of fully quantitative counts, especially if accompanied by determination of chlorophyll *a* as a proxy of algal biomass. The semi-quantitative methods usually allocate an abundance score to each species appearing in the sample, by assigning it to one of several abundance categories, e.g. from “very rare” to “extremely abundant”. Various scales of species abundance have been used for more than 100 years. Korde (1956) published a 6-score scale, ranging from 1 = rare (1–5 cells per slide/chamber) to 6 = extremely abundant (many cells in every field of view), citing earlier works (Vislouxh, 1916; Vereshagin, 1926; Knopp, 1954). While the original authors referred to cells, they actually meant ‘algal units’ (cells or colonies, as appropriate, together with any peripheral mucilage). Since then, such scores were widely used (e.g., Chandler, 1970; Whitton et al., 1991; Talling & Heaney, 2015; Barinova, 2017a). The scales applied varied in their total number of scores from 3 to 9.

To address the need to process a large number of samples from different water bodies in monitoring programs, with limited expert-time that could conduct the analyses, we had to compromise and conduct semi-quantitative phytoplankton counts for some

of the water bodies. Here, we describe our analysis method, based on the 6-score scale of Korde (1956), using data collected as part of long-term monitoring of phytoplankton in the Agmon Wetland, northern Israel. Our objectives were: (1) to describe how we collect semi-quantitative phytoplankton data by applying Korde’s scores and converting the scores to estimates of algal unit concentrations, and (2) to demonstrate how to maximize output from semiquantitative phytoplankton data collected this way.

Methods

Study site

The Agmon Wetland (also known as: Lake Agmon) is a small (area: 1.1 km²) shallow (mean depth ~0.3 m, max depth 1.5 m) man-made water body in the southern part of the Hula Valley, Israel, created in 1994 as part of a regional land rehabilitation program (Hambright & Zohary, 1998). Since its creation, Agmon has become a major stopover for migrating birds, and a major birdwatching sanctuary (Hambright & Zohary, 1999). A monitoring program was established in 1994 to follow the development of physical, chemical and biological properties of Agmon, with semi-quantitative phytoplankton scores being one of the monitored variables (Zohary et al., 1998). This study is based on the phytoplankton Korde score data collected by us during 2008–2021, as detailed below.

Sampling

Subsurface water samples (100 ml) for phytoplankton determinations were collected monthly from a dock by the Agmon outflow from 2008 till 2021, using a bucket, and preserved in Lugol’s iodine solution. A field team from MIGAL (Galilee Knowledge Center, Israel) collected the samples and brought them to the Kinneret Limnological Laboratory for phytoplankton determinations. There were several long sampling gaps (Aug 2009 to Aug 2010, Dec 2014 to Sep 2016, Apr to Dec 2020), as well as many occasions when a monthly sampling was skipped, such that out of 12 samples/year × 14 years = 168, only 111 monthly phytoplankton samples were collected. In fact, only two years (2011, 2017) had 12 monthly samples, all other

years had 11 or less. This limited the analyses that could be performed on the data.

Phytoplankton semiquantitative determinations

Ten ml of the Lugol-preserved samples were sedimented overnight in HydroBios sedimentation chambers (bottom surface area: 500 mm²), then examined under an inverted microscope (Zeiss Axiovert M135). This sedimented volume provided samples with a few to several tens of algal units per field of view at 200× magnification, which was convenient for the microscopist. The entire chamber was first scanned under low magnification (50×), to get an impression of the sample and learn which are the abundant larger species. Then, under 200× magnification, at least 10 arbitrary fields of view were examined. In each field of view, all species present were identified and recorded in a species list. Species appearing for the first time were given a score ranging from 1 to 6 that best described their frequency of appearance, according to Table 1. Additional fields of view beyond the initial 10 were scanned, until no new taxa were observed. At this point, 5 additional fields of view were examined, and if no new taxa were observed, recording stopped. If a new taxon was detected, recording continued until 5 additional consecutive fields had no new taxa. Thus, for each sample at least 15 fields of view were scanned at 200× magnification. After the last field of view, the scores allocated to each taxon were revisited and changed if the frequency didn't correspond to the previously assigned score. Algae observed were identified to genus or species level and allocated to a higher taxonomic group using the conventional taxonomic literature and the

website algaebase.com of Guiry & Guiry (2023). Scores of all species from the same higher taxonomic group (Cyanobacteria, Chlorophyta etc.) were summed, to give the 'sum-of-scores' (SoS) parameter. Species were also allocated to one of the seven morpho-functional groups described by Kruk et al. (2010a, b), and to relevant bioindicator groups (if applicable), according to Barinova et al. (2017b).

Comparison with conventional microscope counts

In order to compare the Korde scores with conventional microscope counts, we conducted both determinations on five new samples (from 2023), prepared as detailed above and counted by both methods. First, Korde scores were allocated as detailed above, then the same sample in the same sedimentation chamber was counted by the Utermöhl (1958) method as is done routinely in our lab for monitoring the phytoplankton of Lake Kinneret (Zohary, 2004), using the counting software PlanktoMetrix (Zohary et al., 2016). This enabled comparison of the time at the microscope needed for each type of determination and allowed us to compare results. Based on the agreement between the two methods and an exponential equation linking the two parameters when regressed against each other, we then transformed the Korde scores to estimates of concentrations, in algal units per ml.

Functional groups

Based on morphology, each species found in Agmon was allocated to one of the seven morpho-functional

Table 1 The 6-score frequency scale applied for phytoplankton monitoring in Agmon Wetland (modified from Korde, 1956), and the conversion factor for transforming the scores to estimated concentrations of algal units ml⁻¹ based on the expo-

ponential regression equation Y (algal units/ml) = $4.7405e^{0.8328X}$, $R^2 = 0.5218$, where X is the Korde score. One algal unit is 1 cell in single-celled species or 1 colony in colonial (filament, coenobium, colony) species

Frequency of appearance	Number of algal units observed	Korde's score (unitless)	Conversion factor to Algal units/ml
Rare	Rare, up to 3 algal units in all scanned fields of view	1	11
Occasional	Infrequent, but > 3 algal units in all scanned fields of view	2	25
Common	At least 1 algal unit in ~half of the fields of view	3	58
Frequent	One or more algal units in most fields of view	4	133
Very frequent	at least 1 algal unit in every field of view	5	305
Extremely abundant	Several to many algal units in every field of view	6	701

groups of Kruk et al. (2010a, b). The parameter used for further analyses was the sum of estimated algal unit concentrations for all taxa belonging to each functional group.

Bioindication

For assessing the aquatic environment based on the phytoplankton present in it, we applied Barinova's Combined Bioindication Method for aquatic ecosystems (Barinova, 2017b). The method is based on a species list that extracts from the published literature information on conditions under which a species is most likely to be found (termed by the author "ecological preference"). The list consists of >8000 species of algae and Cyanobacteria, each known to be associated with at least one of 10 environmental variables (Barinova and Fahima, 2017). To demonstrate the use of our semi-quantitative counts for bioindication, we applied two classification systems: one reflecting water quality based on the content of organic biodegradable matter (Rolauuffs et al., 2004), the other indicating salinity (based on Hustedt, 1957 and applied by Barinova, 2017b). For each of these systems, the number of species, sum-of-scores, and the estimated concentrations of algal units of each indicator species were summed by indicator category.

Results

Comparison of Korde scores with conventional microscope counts

The average time at the microscope for analyzing a sample by allocating Korde scores was 48 min. (range 45–50 min). The average time to count the same sample, in the same sedimentation chamber, using the conventional Utermöhl method was 221 min. (range 210–240 min.), i.e. 4.6 times longer. This excludes additional time needed for doing cell size measurements, entering raw data and computing biovolume and biomass, if biomass estimates are being made for the conventional counting method. Thus, ~5 samples can be processed by the semi-quantitative method at the same time it takes to analyze a single sample using the conventional method. Based on our previous experience, assigning a Korde score to each

species in a sample adds only 3–5 min per sample compared to constructing a species list alone.

A good agreement was found between the Korde scores allocated and the corresponding conventional counts of cells/colonies per ml (Fig. 1). An exponential regression between the two parameters, Y (algal units/ml) = $a \cdot \exp^{bx}$, where X is the Korde score, $a = 4.7405$, $b = 0.8328$, had an $R^2 = 0.5218$ (Fig. 1). ANOVA in Excel revealed that the relationship was highly significant ($P < 0.001$). Hence, by solving this regression equation, we could transform the allocated Korde scores to estimates of algal units concentrations (Table 1).

Scores 2 and 1 were allocated most frequently (83 and 53 times, respectively), whereas score 6 was least frequent (only 5 times).

Using semiquantitative counts

Community composition: Over the 14 years of the study, 211 species belonging to 8 phyla were found in the phytoplankton of Agmon (Table 2), with the number of taxa found in each sample ranging 15–35. Chlorophyta had the highest number of species (76) followed by Euglenozoa (44), Cyanobacteria (34), Bacillariophyta (27), Ochrophyta (14), Charophyta (10), Cryptista (3), and Miozoa (3). Four species were the most occurring taxa observed in Agmon samples (Table 2). The chlorophyte *Ankyra* sp. appeared in 84% of the 111 samples examined, with scores ranging from 2 to 4 most of the times. The centric diatom *Stephanocyclus meneghinianus* (Kützing) Kulikovskiy, Genkal & Kociolek (syn: *Cyclotella meneghiniana* Kützing) was present in 79% of the samples examined, with scores 3 to 4 attained most times. The cryptist *Cryptomonas ovata* Ehrenberg occurred in 78% of the samples, usually acquiring high scores, with twenty times score 6, nineteen times scores 5 and 4 each, and 21 times score 3 (Table 2). The fourth most commonly occurring taxon was the diatom *Nitzschia* sp. that appeared in 73% of the samples attaining mainly scores 3 and 2. While these four species were nearly always present in the water, other species made greater contribution to the taxonomic diversity and overall algal abundance.

Due to the many months with missing data, it was difficult to reach conclusions regarding long-term trends and patterns. Hence, to demonstrate possible

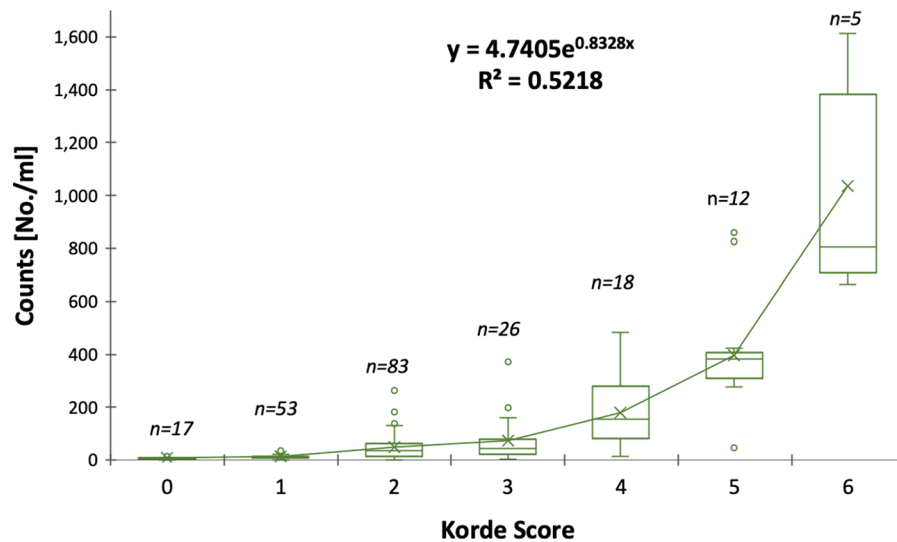


Fig. 1 Box plot of algal unit (cell or colony) counts (Y-axis) per Korde score (X-axis), based on data for 5 samples on which both parameters were determined. Korde scores were assigned as described in Methods, algal units per ml were counted using the traditional Utermöhl inverted microscope method. For each Korde score, all species counts were collected into a box, showing the median (horizontal line), mean

(X), 25th and 75th percentiles (upper and lower box borders), while the whiskers show 1.5 times of the interquartile range (lower and upper small bars). Outlier points are also shown. N is the number of incidents of taxa receiving the particular score in the five samples. The equation shown is the regression line for an exponential correlation for the same data, with Y being estimated concentrations as a function of the Korde score

use of the Korde scores we examined averaged annual cycle patterns for various parameters, based on data for all Januaries 2008–2021, all Februaries, etc.

Annual cycles

We first examine the annual cycle of three phytoplankton parameters: (1) species richness (i.e. no. of species), (2) sum-of-scores and (3) estimated algal unit concentrations computed from Korde scores (Fig. 2). A distinctive pattern of species richness is evident (Fig. 2a) with lowest values (multiannually averaged species numbers: 15–20) at the beginning and end of the year, and highest (>30) during the summer, from June to October. Chlorophyta contributed most to species richness. The number of species of Bacillariophyta (diatoms) and Cryptista (cryptophytes) remained nearly constant throughout the year. Miozoa (dinoflagellates) and Charophyta were rare, both groups were absent in December.

The seasonal pattern of species richness was replicated in the seasonal pattern of the sum-of-scores (SoS, Fig. 2b), This is expected as the no. of species

is the basic contributor to the SoS. Still, the highest SoS value occurred in October compared to the highest no. of species occurring in June (Fig. 2a, b), due to one or several species being highly abundant in October. The sum-of-scores of diatoms and cryptophytes was more or less double the no. of species in Fig. 2a, and so were the scores of Cyanobacteria and Ochrophyta in summer. The notably high SoS values of Chlorophyta at all months were mostly due to the high species richness of this group whereas high scores of Cyanobacteria in the summer months were mostly due to a few highly scoring species.

When the same semi-quantitative data were transformed to estimates of algal unit concentrations, several features of the annual cycle in the Agmon phytoplankton became much more distinct than in the SoS plot (Fig. 2b, c). These features included the numerical domination of blue-greens in August–September, with concurrent reduction of cryptophytes, chlorophytes and euglenophytes. It also became clear that cryptophytes had a significant numerical contribution from October till April, a contribution that was masked when the no. of species or SoS were presented (Fig. 2a, b, c).

Table 2 Phytoplankton taxa from Agmon Wetland, that scored 6 on the Korde scale at least twice during 2008–2021. Data given are the number of samples examined where the taxon attained the score 6, and the percentage of the times that

the species was observed (i.e. scored 1 or higher) in the 111 samples examined. Taxonomic allocation is based on Guiry & Guiry (2023)

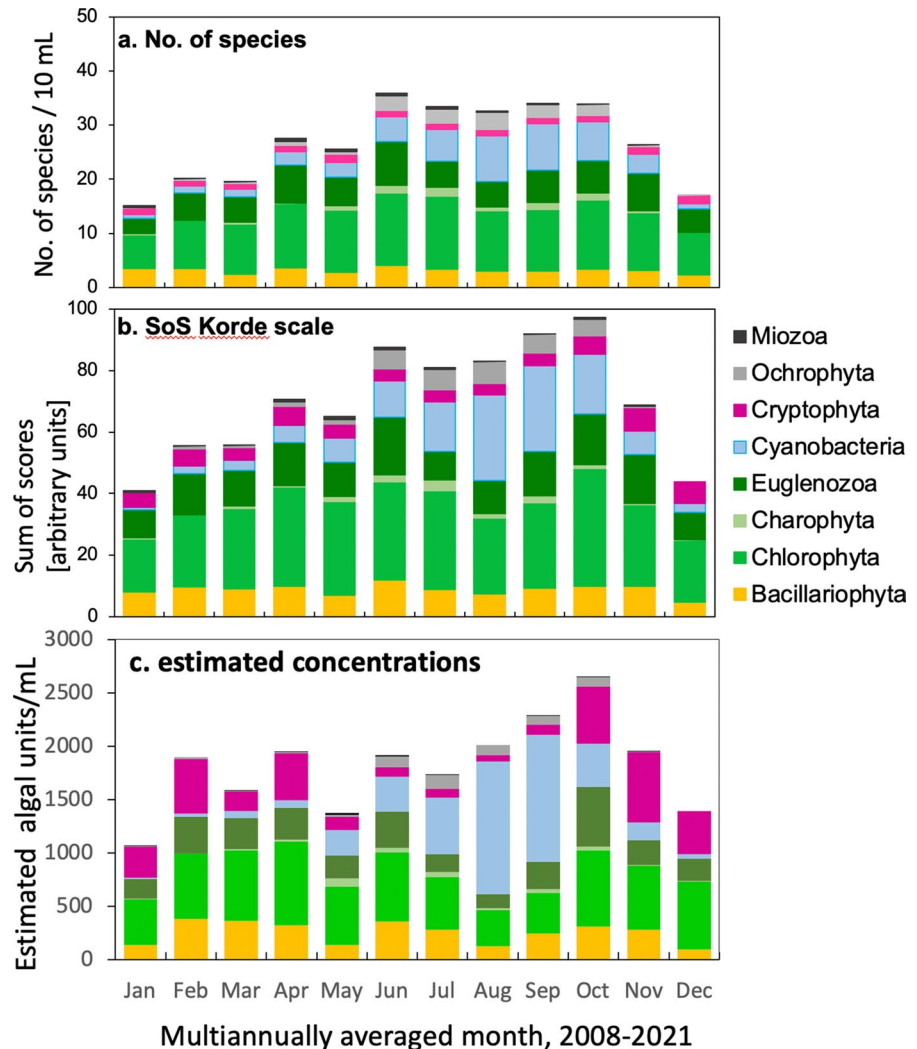
Species	# samples with score 6	Presence in samples (%)
Phylum: Cyanobacteria; Class: Cyanophyceae; Order: Chroococcales		
<i>Microcystis wesenbergii</i> (Komárek) Komárek ex Komárek 2006	2	28
Order: Nostocales		
<i>Chrysochlorum bergii</i> (Ostenfeld) E.Zapomelová, O.Skácelová, P.Pummann, R.Kopp & E.Janecek 2012	2	17
<i>Dolichospermum planctonicum</i> (Brunnthaler) Wacklin, L.Hoffmann & Komárek 2009	3	15
<i>Dolichospermum spiroides</i> (Klebahn) Wacklin, L.Hoffmann & Komárek 2009	4	22
Order: Oscillatoriales		
<i>Arthrospira platensis</i> Gomont 1892	4	15
<i>Oscillatoria</i> sp. Vaucher ex Gomont 1892	2	17
Phylum: Chlorophyta; Class: Chlorophyceae; Order: Sphaeropleales		
<i>Ankyra</i> sp. Fott 1957	5	84
Kingdom: Chromista		
Phylum: Bacillariophyta; Class: Bacillariophyceae; Order: Bacillariales		
<i>Nitzschia</i> sp. Hassal 1845	2	73
Order: Fragilariales		
<i>Fragilaria rumpens</i> (Kützing) G.W.F.Carlson 1913	2	17
Order: Licmophorales		
<i>Tabularia affinis</i> (Kützing) Snoeijis 1992	2	17
Class: Coscinodiscophyceae; Order: Aulacoseirales		
<i>Aulacoseira granulata</i> var. <i>angustissima</i> (O.Müller) Simonsen 1979	2	35
Class: Mediophyceae; Order: Thalassiosirales		
<i>Stephanocyclus meneghinianus</i> (Kützing) Kulikovskiy, Genkal & Kociolek 2022 Kützing 1844	3	79
Phylum: Cryptista; Class: Cryptophyceae; Order: Cryptomonadales		
<i>Cryptomonas curvata</i> Ehrenberg 1832	6	34
<i>Cryptomonas ovata</i> Ehrenberg 1832	20	78
Kingdom: Protozoa		
Phylum: Euglenozoa; Class: Euglenophyceae; Order: Euglenales		
<i>Lepocinclis tripteris</i> (Dujardin) B.Marin & Melkonian 2003	2	30

Morpho-functional groups

The Korde-score-based estimates of algal unit concentrations for each taxon present in the water, proved useful for following the annual pattern of morpho-functional groups in Agmon (Fig. 3). The overall highest estimated concentrations were of taxa belonging to Group IV of medium sized organisms lacking specialized traits and Group V of medium to large sized flagellated taxa. The most abundant taxa belonging to these groups were the chlorophytes

Desmodesmus maximus (West & G.S.West) Hege-
wald, *Ankyra* sp. and *Tetradasmus lagerheimii*
M.J.Wynne & Guiry (Group IV) and the cryptophytes
Cryptomonas ovata and *Rhodomonas pusilla* (Bach-
mann) Javornický (syn: *Cryptomonas curvata* Guseva)
(Group V). Group III of filaments with aerotopes
(essentially Cyanobacteria) were particularly abun-
dant in August and September, with *Phormidium*
ambiguum Gomont most abundant. Group I of small-
est size species and group VI of non-flagellated spe-
cies with siliceous exoskeletons (diatoms) were

Fig. 2 Multiannually averaged (2008–2021) annual cycle of parameters derived from semi-quantitative phytoplankton determinations for the 8 taxonomic groups (color coded) appearing in Agmon: a. no. of species per group found in 10 ml subsamples, b. sum-of-scores (SoS), c. estimated cell/colony concentrations (algal units mL^{-1})



present all year, with lower concentrations in winter, with *Stephanocyclus meneghinianus* and *Nitzschia* sp. usually most abundant. Group VII of large mucilaginous colonies were more abundant in the summer months, with *Monoraphidium contortum* (Thuret) Komárková-Legnerová and *Aphanocapsa* sp. scoring highest. Only one representative of Group II (flagellated species with siliceous structures) was ever found in the Agmon samples and only once, so this group was excluded from Fig. 3.

Water quality indication

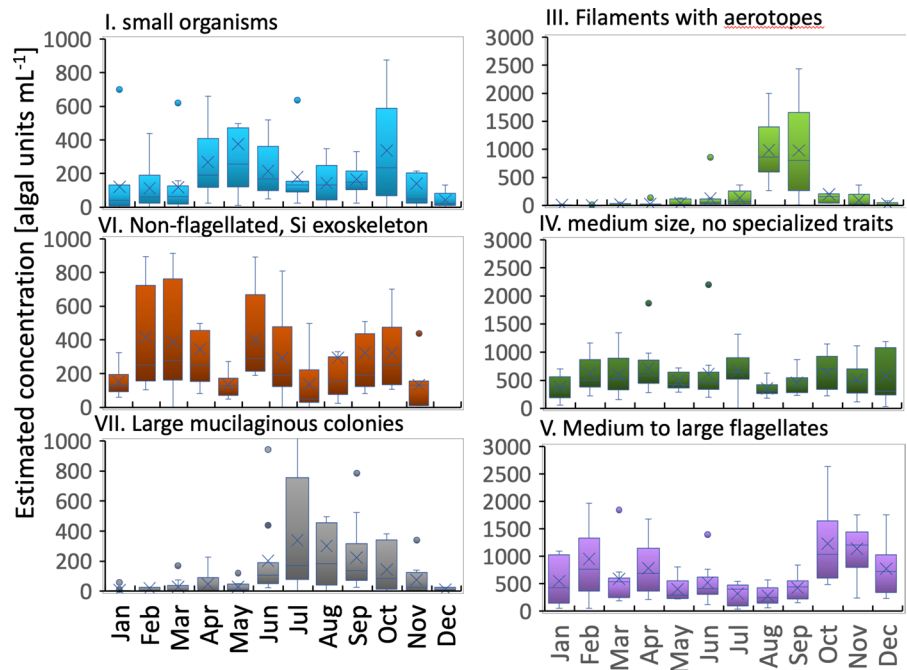
Indicator species for five saprobic classes were found in Agmon: xenosaprobity, oligosaprobity, β -mesosaprobity, α -mesosaprobity and polysaprobity.

These saprobity levels were translated into 5 pollution categories, as done by the EU Water Framework Directive (Rolauffs et al., 2004), from unpolluted (xenosaprobites) and considered high (good) water quality to highly polluted (polysaprobites) and considered bad water quality.

The annual cycle of those water quality indicators in Agmon were plotted based on the three phytoplankton parameters used previously: no. of indicator species in the sample (Figs. 4a b), sum-of-scores (Figs. 4c, d), and estimated concentrations of algal units (Figs. 4e, f). This was done to compare the usefulness of those three parameters.

At all times of year, the water quality index (Figs. 4a, c, e) was clearly dominated (usually ~70%), by species indicating moderate water quality, with a

Fig. 3 The multiannually (2008–2021) averaged annual pattern of six phytoplankton morpho-functional groups (after Kruk et al. 2010a, b) for Agmon, based on the estimated cell/colony concentrations. The box plots show the median as the horizontal line within the box, X as average, the 25th and 75th percentiles as the box boundaries, 1.5 times of the interquartile range at the end of the bars, and outliers as filled circles. Group II of Kruk et al. (2010a, b), of flagellates with siliceous exoskeletons, had no representatives in the Agmon phytoplankton and is not shown



second-largest contribution (15–30%) of taxa indicating poor water quality, and <10% contribution by taxa indicating high water quality. This was similarly evident, with only minor differences, when the parameter plotted was the no. of indicator species per category, their sum-of-scores, or the transformed estimate of algal unit concentration. The three parameters also showed highest contribution of the moderate category in July, the month when indicators of good water quality were also most abundant, and indicators of poor water quality least abundant (Fig. 4a, c, e). In winter (Dec-Feb), the relative contribution of taxa indicating poor water quality was highest.

Salinity indicators of four of the categories defined by Barinova (2017b) had representatives in the Agmon plankton: oligohalobes-halophobes (Hb, strictly freshwater species that perish with even slight increase of salinity), oligohalobe-indifferent (I, freshwater species that are indifferent to slightly elevated salt concentrations), oligo-halophile (HI, freshwater species that are stimulated by slightly elevated salinity) and meso-halobes (Mh, species of brackish water).

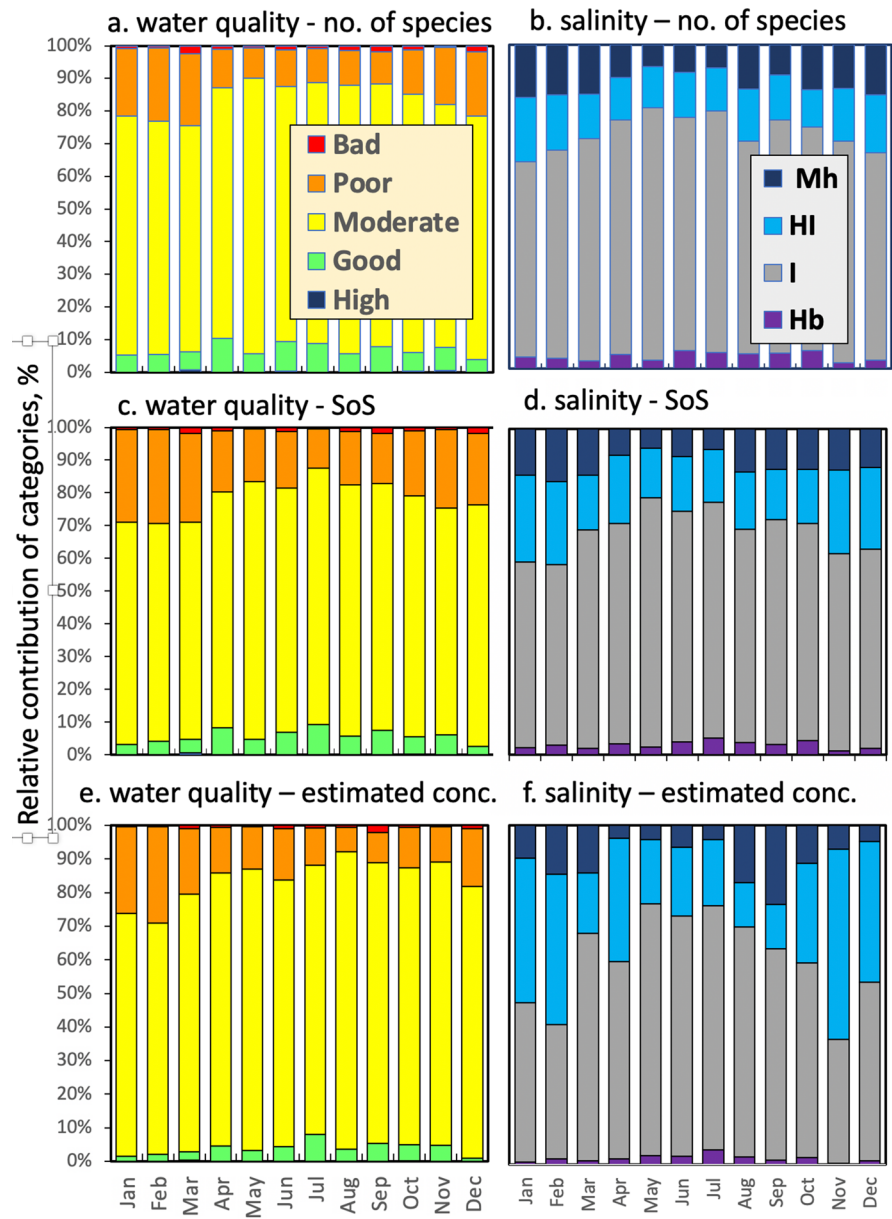
The salinity index (Fig. 4b, d, f) was dominated by species of category I indifferent to slightly elevated salt concentrations, with a second-largest

contribution of taxa of the HI and Mh groups, and a negligible contribution of HB, halophobes, the strictly freshwater species. In the case of salinity, we do find differences in the emerging seasonal pattern between the three different parameters plotted in Fig. 4: indicators of higher salinity (HI+Mh) had a much greater contribution when the estimated concentration parameter was plotted compared to their contribution when the No. of species and SoS parameters were plotted. This was mostly evident in Jan, Feb and Nov, when the contribution of the HI + Mh groups was close to or even exceeded 50% (Fig. 4f). In those months, *Cryptomonas ovata* and *Stephanocyclus meneghinianus*, (HI indicators), were particularly abundant and scored high (4–6) on the Korde scale.

Discussion

Semi-quantitative microscope counts are usually a compromise between the standard microscope cell biomass determinations and measuring only chlorophyll a, with no phytoplankton species composition data beyond, perhaps, a species list. We have shown that five samples can be processed by the Korde score system at the time it takes to count one sample by the

Fig. 4 The multiannually (2008–2021) averaged annual cycle of the relative contribution of representatives of the different bioindicator categories for organic pollution and water quality (left panels) or salinity (right panels), when percent contribution is based on the no. of indicator species in each category (a, b), the sum-of-scores (SoS) for each category (c, d) or based on estimated algal unit concentrations (e, f). Categories for water quality are according to Rolauffs et al. (2004). Categories for salinity are: Hb—freshwater halophobe (strictly freshwater species that perish with even slight increase of salinity), I—freshwater, indifferent to slightly elevated salt concentrations, HI freshwater, enhanced by slightly elevated salt concentrations, Mh -brackish, with salinity ranging 5–20‰



currently standard Utermöhl method, a substantial saving of time. Furthermore, allocating Korde scores to taxa observed under the microscope adds insignificantly to the time required to only make a species list. Yet the Korde scores, and their transformation to estimates of algal unit concentrations make a huge addition to the possible data use and analysis. In addition, if making a species list is conducted anyway, we recommend assigning Korde scores, creating calibration curves, and converting the scores to concentration estimates.

The volume of sample to be sedimented in a sedimentation chamber should be determined per body of water and kept fixed across all samples from a particular study. We found 10 ml to be a suitable volume for freshwater samples from mesotrophic to eutrophic natural waterbodies in Israel (subtropical climate), ranging in salinity from fresh to brackish. Oligotrophic or hypertrophic water bodies will likely require sedimenting larger or smaller volumes, respectively.

Korde scores provide relative estimates of the chance to meet a species in the sample, the higher the value the higher the chance. The ‘sum-of-scores’ combines species richness and relative abundance of each taxon to give a single unitless value for taxa belonging to a certain group of species, be it based on taxonomy or on other criteria. The Korde scores are not an estimate or a proxy of biomass: small and large species with the same score will have the same estimated cell concentration and the same contribution to the SoS, this limitation must be born in mind. For this reason, we argue that one shouldn’t convert the estimates of algal unit concentration to biomass, although technically it is possible to multiply abundance by a mean biovolume for each species.

Furthermore, the SoS parameter is insufficient for describing phytoplankton communities, mostly because this parameter doesn’t distinguish between diversity and abundance. For example, a SoS value of 12 can be due to two highly abundant species each scoring 6, to twelve rare species scoring 1 each, or to a wide range of intermediate situations. To overcome this problem, for each taxon we transformed the Korde score to estimated concentration of algal units, using an exponential equation derived from the calibration exercise described in Methods. Plotting the estimated concentrations increased resolution. For example, in Fig. 2c, the dominance of Cyanobacteria in August and September, and of cryptophytes from October to April is much more evident than when the no. of species or sum-of-scores are plotted (Fig. 2a, b). It also became apparent in Fig. 2c that euglenophytes and cryptophytes alternated with Cyanobacteria, they didn’t co-occur in large numbers. We recommend conducting a calibration against Utermöhl counts on at least 5 samples for each waterbody examined, and converting the Korde scores to estimates of algal unit concentrations.

The concentration estimates based on Korde-scores were also found informative for bioindication studies, especially in cases when both highly abundant and rare/infrequent indicator species are present in the samples. In such cases, assessing the waterbody based on the estimated concentrations was clearer than when the determination was based on the number of indicator species, or the sum of scores. In the case of Agmon salinity indicators (Fig. 4f), the dominance of ‘indifferent’ taxa—those

freshwater species that can tolerate low levels of added salt, alternating temporarily with dominance of salt-resistant taxa (HI + Mh categories), reflects the ambient conditions, of low background electrical conductivity ($\sim 0.5 \text{ mS cm}^{-1}$) with short peaks of elevated values ($2\text{--}3 \text{ mS cm}^{-1}$) (Barnea et al., 2020). When the plots were based on no. of indicator taxa or sum-of-scores, the contribution of salinity resistant taxa was much less evident.

The clear dominance of taxa indicating medium level water quality (Fig. 4a, c, e) defines Agmon as moderately polluted, not surprising given that the wetland is at the heart of agricultural land treated with fertilizers, and comprises a stop-over station to tens of thousands of cranes that defecate into it. The SoS parameter was widely applied by Barinova in recent bioindication studies (e.g., Barinova & Nevo, 2012; Barinova et al., 2016a, b; Khuram et al., 2017, 2019; Barinova & Mamanazarova, 2021). Studies focusing on functional groups can also be based on such semi-quantitative data. The dominance in Agmon, at all times of the year, of medium to large sized flagellates and non-flagellated species from morpho-functional groups IV and V of Kruk et al. (2010a, b), reflects its nature as a shallow water body rich in organic matter.

In conclusion, our results show that when the time and money allocated to a project are insufficient for standard biomass analyses, semi-quantitative microscope determinations can be used to achieve limited goals. Examples presented are indicating the most abundant taxa and their seasonality, showing which functional groups comprise the assemblage, and which environmental conditions are reflected by the phytoplankton association. Potential applications for the method include monitoring programs, experiments assessing impacts of various factors/additions on community composition, and other applications in which it is sufficient to find out which species increased/declined/became dominant but it is not essential to have biomass data.

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Data availability None.

Declarations

Conflict of interest The authors declare that they have no conflict of interest.

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