

Supplements

All data and R scripts used for this article are open access and are available at Zenodo (<https://doi.org/10.5281/zenodo.5517381>), the digital slide scans and annotations can be viewed online in BIIGLE (direct link: <https://biigle.de/images/1933537/annotations?scaleLine=1&zoomLevel=1&labelTooltip=1>, login: "digimic@example.com", password: "microscopy"). A video summary of the digital workflow can also be found here (<https://uni-duisburg-essen.sciebo.de/s/rT4AtzAYXAxwsEq>).

Figure S1: Complete hierarchical clustering with triplicate annotation and complete list of diatoms using OMNIDIA 4-letter code.; Figure S2: Pairwise comparison of diversity and biomonitoring indices of all samples depending on analysis method. Diversity and species values: a) species richness, b) Shannon diversity entropy, c) Simpson diversity entropy. Trophic biomonitoring indices: d) Indice de polluosensitivité (IPS), e) Indice biologique des diatomées (IBD), f) Trophic diatom index (TDI), g) indice des diatomées générique (IDG), h) Rott Trophic index (Rott TI). Saprobic biomonitoring indices: i) Rott saprobic index (Rott SI), j) Sladeczek saprobic index (Sladeczek). Methods summarized as trad = "traditional" light microscopy and digi = digital microscopy. Video S1: Virtual diatom slide preparation process. Table S1: Species distribution along the sites. Percentage distribution on the triplicates. Methods: mic = traditional light microscopy, scan = digital microscopy. Percentages: +++ >50%, 50%>++>25%. 25%>+>10%, 10%> r (rare)>5%, 5%> vr (very rare)>0

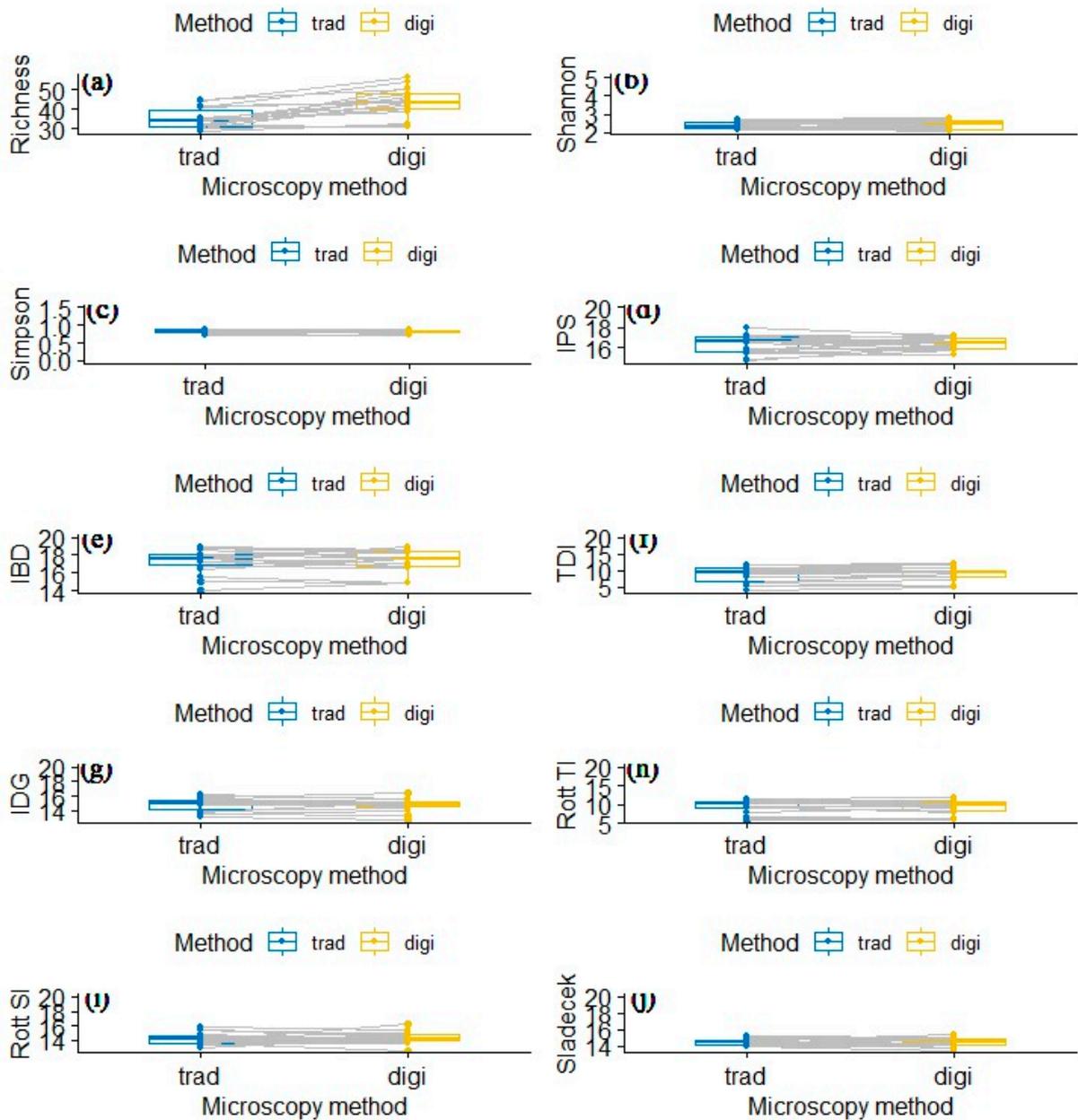


Figure S2. Pairwise comparison of diversity and biomonitoring indices of all samples depending on analysis method. Diversity and species values: a) species richness, b) Shannon diversity entropy, c) Simpson diversity entropy. Trophic biomonitoring indices: d) Indice de polluosensitivité (IPS), e) Indice biologique des diatomées (IBD), f) Trophic diatom index (TDI), g) indice des diatomées générique (IDG), h) Rott Trophic index (Rott TI). Saprobic biomonitoring indices: i) Rott saprobic index (Rott SI), j) Sladecek saprobic index (Sladecek). Methods summarized as trad = “traditional” light microscopy and digi = digital microscopy.

Table S1. Species distribution along the sites. Percentage distribution on the triplicates. Methods: mic = traditional light microscopy, scan = digital microscopy. Percentages: +++ >50%, 50%>++>25%. 25%>+>10%, 10%>r (rare)>5%, 5%>vr (very rare)>0.

Taxon Name	Sites →	M1		M2		M3		M4		M5		M6	
	Code	mic	scan										
<i>Acbnanthidium</i>	ACHD	0	vr	0	0	0	0	0	vr	0	vr	0	vr
<i>Acbnanthidium affine</i>	ACAF	0	0	0	vr	0	vr	0	0	0	vr	0	vr
<i>Acbnanthidium atomoides</i>	ADAM	vr	vr	vr	vr	vr	vr	vr	vr	vr	0	vr	vr
<i>Acbnanthidium delmontii</i>	ADMO	0	vr	0	vr	vr	0	vr	0	0	0	vr	0
<i>Acbnanthidium druartii</i>	ADRU	vr	0	vr	0	vr	0	0	0	vr	0	vr	vr
<i>Acbnanthidium eutrophilum</i>	ADEU	0	0	0	vr								
<i>Acbnanthidium exile</i>	ADX1	0	0	0	0	0	0	0	0	0	vr	0	0
<i>Acbnanthidium gracillimum</i>	ADGL	0	0	vr	0	0	0	0	0	0	0	0	0
<i>Acbnanthidium jackii</i>	ADJK	0	0	vr	vr	0	vr	0	0	vr	0	0	0
<i>Acbnanthidium kranzii</i>	ADKR	vr	0	0	vr	0	vr	0	vr	0	0	0	vr
<i>Acbnanthidium lineare</i>	ACLI	0	0	0	0	0	vr	0	0	0	0	0	0
<i>Acbnanthidium minutissimum</i>	ADMI	++	++	++	++	++	++	++	+++	+	+	++	++
<i>Acbnanthidium pyrenaicum</i>	ADPY	0	vr	vr	0	vr	vr	vr	vr	0	0	vr	vr
<i>Acbnanthidium rivulare</i>	ADRI	r	r	vr	vr	vr	vr	vr	vr	vr	0	vr	vr
<i>Acbnanthidium saprophilum</i>	ADSA	0	0	0	vr	0	vr	0	0	0	vr	0	0
<i>Acbnanthidium straubianum</i>	ADSB	0	vr	0	0	0	0	0	0	0	0	0	vr
<i>Acbnanthidium subatomus</i>	ADSU	0	vr	0	0	0	0	0	0	0	0	0	vr
<i>Adlafia minuscula</i> var. <i>Minuscula</i>	ADMS	0	0	vr	vr	vr	vr	vr	0	0	0	vr	vr
<i>Adlafia muralis</i>	ADMU	vr	0	vr	0	vr	vr	vr	0	0	0	vr	0
<i>Amphora copulata</i>	ACOP	0	vr	0	vr	0	0	0	0	0	0	0	vr
<i>Amphora excimia</i>	AEXM	0	0	0	0	0	0	0	vr	0	0	0	vr
<i>Amphora inariensis</i>	AINA	vr	vr										
<i>Amphora indistincta</i>	AMID	0	vr	0	vr	0	vr	0	vr	0	0	0	vr
<i>Amphora pediculus</i>	APED	+	+	r	r	r	r	vr	vr	vr	vr	+	+
<i>Ampora minutissima</i>	AMNU	vr	vr	0	0	vr	0	0	0	vr	0	vr	vr

<i>Caloneis lancettula</i>	CLCT	vr	vr	vr	vr	vr	vr	0	0	0	0	vr	vr
<i>Cocconeis euglypta</i>	CEUG	vr	vr	vr	0	vr	vr	0	0	0	vr	vr	0
<i>Cocconeis lineata</i>	CLNT	0	vr	0	0	0	0	0	0	0	0	0	0
<i>Cocconeis pediculus</i>	CPED	0	vr	0	0	vr	vr	0	vr	0	0	0	0
<i>Cocconeis placentula</i> var. <i>Euglyptoidea</i>	CEUO	vr	vr	0	0	0	0	0	0	0	0	0	0
<i>Cocconeis placentula</i> var. <i>Placentula</i>	CPLA	0	vr	0	0	0	0	0	0	0	0	0	0
<i>Cocconeis pseudolineata</i>	COPL	0	vr	0	0	0	vr	0	0	0	0	0	vr
<i>Craticula molestiformis</i>	CMLF	0	vr	0	vr	0	vr	0	0	0	0	0	0
<i>Craticula subminuscula</i>	CSNU	vr	vr	0	vr	0	0	0	0	0	0	vr	vr
<i>Cymatopleura solea</i>	CSOL	0	0	vr	vr	0	vr	0	vr	0	0	0	0
<i>Delicata delicatula</i>	DDEL	0	0	vr	0	0	0	0	0	0	0	0	0
<i>Denticula tenuis</i>	DTEN	vr	0	vr	r	r							
<i>Diademsis confervacea</i>	DCOF	0	vr	0	0	0	0	0	0	0	0	0	0
<i>Diploneis oculata</i>	DOCU	0	0	0	0	vr	0	0	0	0	0	0	0
<i>Discostella</i>	DISC	0	vr	0	0	0	0	0	vr	0	0	0	0
<i>Encyonema</i>	ENCY	0	0	0	0	0	vr	0	vr	0	0	0	0
<i>Encyonema caespitosum</i> var. <i>Caespitosum</i>	ECAE	0	0	0	0	0	0	0	0	0	0	vr	0
<i>Encyonema lange-bertalot</i>	ENLB	vr	0	0	0	0	0	0	0	0	0	0	0
<i>Encyonema minutum</i>	ENMI	0	vr	vr	vr	r	r	r	r	0	vr	r	r
<i>Encyonema neogracile</i>	ENNG	0	0	0	0	0	0	0	vr	0	0	0	0
<i>Encyonema reichardtii</i>	ENRE	0	0	0	0	vr	vr	vr	vr	0	0	vr	vr
<i>Encyonema silesiacum</i>	ESLE	0	0	0	0	vr	vr	vr	vr	0	vr	vr	vr
<i>Encyonema ventricosum</i>	ENVE	vr											
<i>Encyonopsis krammeri</i>	ECKR	0	0	0	0	0	0	0	0	0	vr	0	0
<i>Epithemia frickei</i>	EFRI	0	0	0	0	0	0	0	0	vr	0	0	0
<i>Fistulifera pelliculosa</i>	FPEL	0	vr	0	vr	0	vr	0	vr	0	0	0	vr
<i>Fistulifera saprophila</i>	FSAP	0	vr	0	vr								
<i>Fragilaria famelica</i>	FFAM	vr	0	0	0	0	0	0	0	0	0	0	0
<i>Fragilaria perminuta</i>	FPEM	0	0	0	0	0	0	0	0	0	vr	0	0

<i>Navicula notha</i>	NNOT	0	0	0	0	0	0	0	vr	0	0	0	vr
<i>Navicula pelliculosa</i>	NPEL	0	0	0	0	0	0	vr	0	0	0	0	0
<i>Navicula phyllepta</i>	NPHY	0	0	0	0	vr	0	0	0	0	0	0	0
<i>Navicula recens</i>	NRCS	0	vr										
<i>Navicula rhyncocephala</i>	NRHY	0	0	0	0	0	0	vr	0	0	0	0	0
<i>Navicula salinarum</i>	NSAL	vr	0	0	0	0	0	0	0	0	0	0	0
<i>Navicula tenelloides</i>	NTEN	0	0	0	0	0	0	0	vr	0	0	0	vr
<i>Navicula tripunctata</i>	NTPT	vr											
<i>Navicula vandamii</i>	NVDA	0	0	0	0	0	0	0	0	0	0	0	vr
<i>Navicula veneta</i>	NVEN	vr	vr	0	vr	vr	vr	vr	vr	vr	0	vr	vr
<i>Nitzschia</i>	NITZ	0	vr	0	0	0	0	0	vr	0	0	0	vr
<i>Nitzschia amphibia</i>	NAMP	vr	0	0	0	0	0	0	0	0	0	0	0
<i>Nitzschia archibaldii</i>	NIAR	0	vr										
<i>Nitzschia aurariae</i>	NAUR	0	vr	vr	vr	0	0	0	0	vr	vr	0	vr
<i>Nitzschia clausii</i>	NCLA	0	0	0	0	0	0	0	0	0	0	vr	0
<i>Nitzschia communis</i>	NCOM	0	vr	0	0	0	0	0	0	0	0	0	0
<i>Nitzschia dealpina</i>	NDLP	0	0	0	0	0	0	vr	vr	0	vr	0	0
<i>Nitzschia dissipata</i>	NDIS	vr	vr	vr	vr	r	r	vr	vr	vr	vr	vr	vr
<i>Nitzschia dissipata var. Media</i>	NDME	0	vr	0	0	0	vr	0	0	0	0	0	0
<i>Nitzschia dubia</i>	NDUB	0	vr	0	vr	0	0	0	0	vr	vr	0	0
<i>Nitzschia fibulafissa</i>	NFIB	0	vr	0	0	0	0	0	0	0	0	0	0
<i>Nitzschia frustulum</i>	NIFR	0	0	0	vr	0	vr	vr	vr	0	vr	0	0
<i>Nitzschia intermedia</i>	NINT	0	0	vr	0	vr	0	0	0	0	0	vr	0
<i>Nitzschia liebetruthii</i>	NLBT	0	0	0	0	0	0	0	0	vr	0	0	0
<i>Nitzschia linearis</i>	NLIN	vr	vr	vr	vr	vr	0	vr	vr	0	vr	0	0
<i>Nitzschia palea</i>	NPAL	vr	0	vr	vr	vr	0	vr	vr	0	vr	0	0
<i>Nitzschia palea var. Debilis</i>	NPAD	vr	0	0	0	vr	0	vr	vr	0	0	0	0
<i>Nitzschia paleacea</i>	NPAE	vr	vr	r	vr	vr	vr	vr	vr	vr	0	vr	vr
<i>Nitzschia perminuta</i>	NIPM	0	vr	0	0	0	0	vr	vr	0	0	0	0
<i>Nitzschia pura</i>	NIPR	0	0	0	vr	0	0	0	0	0	0	0	0

<i>Nitzschia pusilla</i>	NIPU	0	0	vr	vr	0	vr	0	vr	0	0	0	0
<i>Nitzschia recta</i>	NIRE	vr	vr	vr	vr	0	0	0	0	vr	vr	0	0
<i>Nitzschia sigmoidea</i>	NSIO	0	0	vr	0	0	0	0	0	0	0	0	0
<i>Nitzschia sociabilis</i>	NSOC	0	vr	0	vr	0	0	0	0	0	vr	0	0
<i>Nitzschia solita</i>	NISO	0	vr	0	0	0	0	0	0	0	0	0	0
<i>Nitzschia soratensis</i>	NSTS	0	0	0	0	0	0	0	0	vr	0	vr	0
<i>Nitzschia subacicularis</i>	NSUA	0	0	0	0	0	0	0	vr	0	0	0	0
<i>Nitzschia sublinearis</i>	NSBL	0	0	0	0	0	vr	0	0	0	0	0	0
<i>Nitzschia supralitorea</i>	NZSU	0	0	0	0	0	0	0	vr	0	vr	0	0
<i>Nitzschia tenuis</i>	NITE	vr	0	0	0	0	0	vr	0	vr	0	0	0
<i>Nitzschia umbonata</i>	NUMB	vr	0	vr	vr	0	0	0	0	vr	0	0	0
<i>Nitzschia wuellerstorfi</i>	NWUE	vr	0	vr	0	0	0	0	0	0	0	0	0
<i>Placoneis elginensis</i>	PELG	vr	0	0	0	0	0	0	0	0	0	0	0
<i>Planorbidium</i>	PLTD	0	vr	0	0	0	0	0	0	0	0	0	0
<i>Planorbidium capitatum</i>	PCAP	0	0	0	0	0	0	0	0	vr	0	vr	0
<i>Planorbidium caputium</i>	PCPU	vr	vr	vr	0	0	0	vr	vr	vr	vr	0	0
<i>Planorbidium delicatulum</i>	PTDE	vr	0	0	0	0	0	vr	0	vr	0	0	0
<i>Planorbidium frequentissimum</i>	PLFR	r	r	vr	vr	vr	vr	vr	vr	+	+	vr	vr
<i>Planorbidium hauckianum</i>	PTHA	0	0	0	0	0	0	0	0	vr	0	0	0
<i>Planorbidium incuriatum</i>	PICU	vr	vr	vr	0	0	0	vr	vr	0	0	0	0
<i>Planorbidium joursacense</i>	PJOU	0	vr	0	0	0	0	0	0	0	0	0	0
<i>Planorbidium lanceolatum</i>	PTLA	r	r	vr	vr	vr	vr	vr	vr	++	++	vr	vr
<i>Planorbidium rostratobolarticum</i>	PROH	0	vr	0	0	0	0	0	0	0	0	0	0
<i>Platessa conspicua</i>	PTCO	r	0	vr	0	0	0	vr	0	0	0	0	0
<i>Platessa holsatica</i>	PLHO	0	vr	0	0	0	0	0	0	0	0	0	0
<i>Psammothidium</i>	PSMT	0	vr	0	0	0	vr	0	0	0	0	0	0
<i>Psammothidium acidoclinatum</i>	PACD	0	vr	0	0	0	0	0	0	0	0	0	0
<i>Psammothidium bioretii</i>	PBIO	vr	0	vr	0	0	0	0	0	0	0	0	0
<i>Psammothidium daonense</i>	PDAO	0	0	vr	0	0	0	0	0	0	0	0	0
<i>Psammothidium grischunum</i>	PGRI	vr	vr	0	0								

<i>Psammothidium lauenburgianum</i>	PLAU	0	0	0	vr	0	vr	0	0	0	0	0	0
<i>Psammothidium marginulatum</i>	PMRG	0	0	0	vr	0	0	0	0	0	0	0	0
<i>Psammothidium microscopicum</i>	PMCR	vr	0	0	0	0	0	0	0	0	0	0	0
<i>Psammothidium subatomoides</i>	PSAT	0	r	0	vr	0	0	0	0	0	0	0	vr
<i>Pseudostaurosira brevistriata</i>	PSBR	0	0	0	0	0	0	0	0	vr	0	0	0
<i>Reimeria</i>	REIM	0	vr	0	0	0	0	0	0	0	0	0	0
<i>Reimeria sinuata</i>	RSIN	vr	vr	vr	vr	vr	0	0	vr	0	0	0	0
<i>Reimeria uniseriata</i>	RUNI	0	vr	vr	vr	0	vr	0	0	0	0	0	0
<i>Rhoicosphenia abbreviata</i>	RABB	vr	vr	vr	vr	vr	vr	0	vr	0	0	vr	vr
<i>Rhoicosphenia abbreviata</i>	RHOI	0	vr	0	0	0	0	0	0	0	0	0	0
<i>Rossithidium petersenii</i>	RPET	vr	0	vr	0	vr	0	vr	vr	vr	vr	vr	vr
<i>Sellaphora</i>	SELL	0	0	0	0	vr	0	0	vr	0	vr	0	vr
<i>Sellaphora crassulaeexigua</i>	SCRA	0	0	0	0	0	0	0	vr	0	0	0	0
<i>Sellaphora nigri</i>	SNIG	vr											
<i>Sellaphora saengerresii</i>	SSGE	0	vr										
<i>Sellaphora seminulum</i>	SSEM	vr	vr	0	vr								
<i>Sellaphora verecundiae</i>	SVER	0	vr	0	0	0	0	0	0	0	0	0	0
<i>Stauroforma exiguiformis</i>	SEXG	0	0	0	0	0	0	0	0	0	vr	0	0
<i>Staurosira venter</i>	SSVE	0	vr	0	0	0	0	0	vr	0	0	0	0
<i>Stephanodiscus hantzschii</i>	SHAN	vr	0	vr	0	0	0	0	0	0	0	0	0
<i>Surirella</i>	SURI	0	vr	0	0	0	vr	0	0	0	0	0	0
<i>Surirella angusta</i>	SANG	0	0	vr	0	0	0	0	0	vr	0	0	0
<i>Surirella angustata</i>	SUAA	0	vr	0	vr	0	0	0	vr	0	vr	0	0
<i>Surirella brebissonii</i>	SBRE	0	vr	vr	0	0	vr						
<i>Surirella brebissonii</i> var. <i>Kuetzingi</i>	SBKU	0	vr	vr	vr	vr	vr	vr	0	vr	vr	0	0
<i>Surirella crumena</i>	SCRU	vr	0	vr	0	0	0	vr	vr	0	vr	0	0
<i>Surirella minuta</i>	SUMI	vr	vr	0	vr	0	vr	vr	0	vr	vr	0	vr
<i>Surirella ovalis</i>	SOVI	0	0	vr	vr	0	vr	0	0	0	vr	0	0
<i>Surirella terricola</i>	STER	vr	vr	0	vr	vr	0	0	0	vr	vr	vr	0
<i>Ulnaria</i>	ULNA	0	0	0	0	0	0	0	0	0	vr	0	0

<i>Ulnaria acus</i>	UACU	0	0	vr	0	0	0	0	0	0	0	0	0
<i>Ulnaria danica</i>	UDAN	0	0	0	0	0	0	0	0	0	vr	0	0
<i>Ulnaria ulna</i>	UULN	0	0	0	0	0	0	0	0	vr	0	0	0