

Supporting Information

Ongoing anthropogenic eutrophication of the catchment area threatens the Doñana World Heritage Site (South-west Spain)

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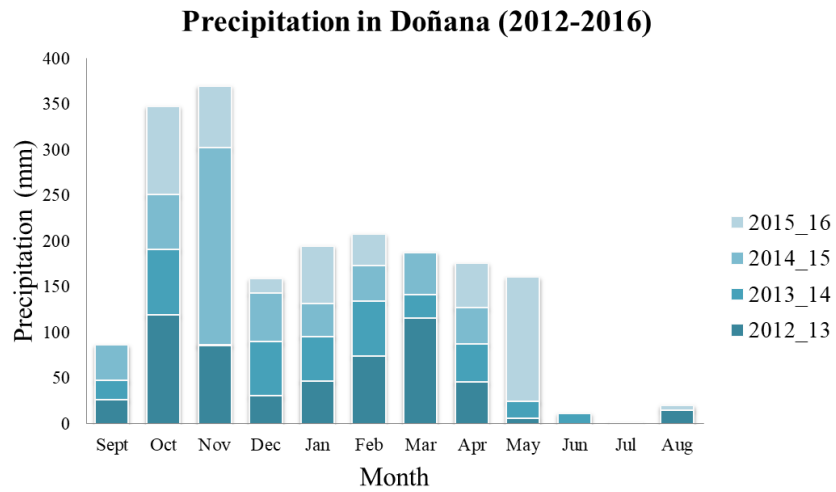


Figure S1. Monthly precipitation in Doñana National Park (DNP) over the four consecutive hydrological years included in this study. Each hydrological year extends from September until the following August. Data were collected from the Meteorological Station located at “El Palacio” within DNP.

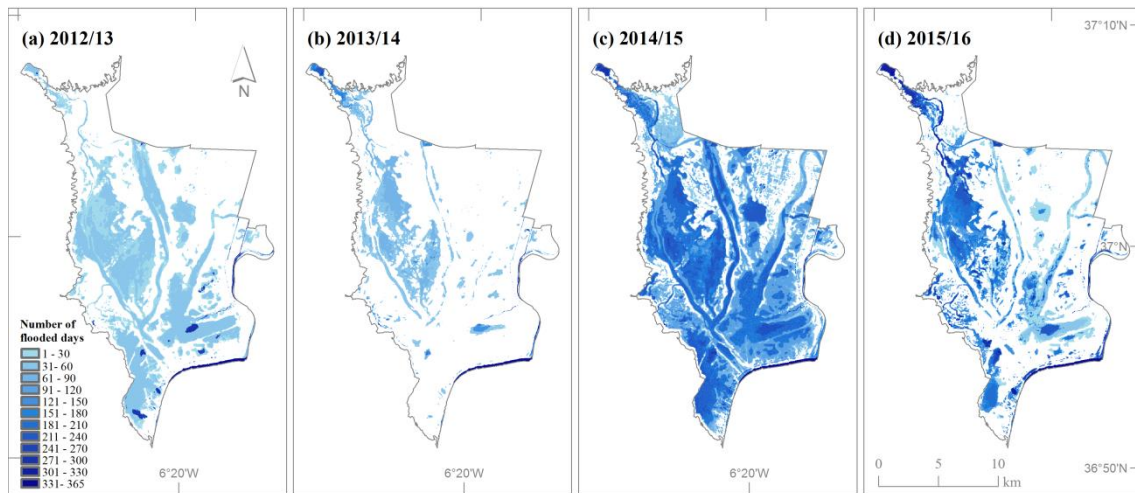


Figure S2. Estimated hydroperiod for the hydrological years 2012/13 (total precipitation=566 mm), 2013/14 (359 mm), 2014/15 (528 mm) and 2015/16 (469 mm). Colour intensity indicates the number of days each pixel was detected as flooded according to inundation masks from Landsat images (see Díaz-Delgado *et al.* 2016 for methodological details).

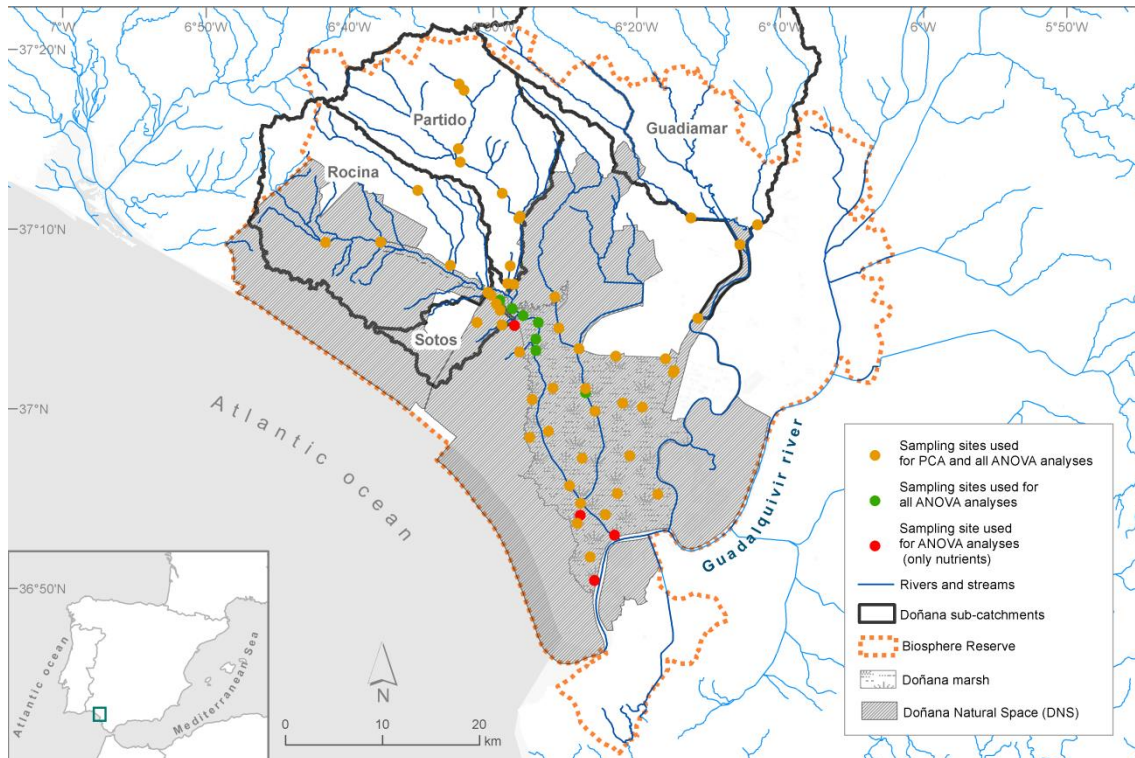


Figure S3. Location of sampling points used in the PCA analysis ($n_{\text{sites}}=49$; $n_{\text{samples}}=338$; orange dots), the two-way ANOVA analyses of nutrient concentrations ($n_{\text{sites}}=59$; $n_{\text{samples}}=434$; orange, green and red dots) and the ANOVA analyses of chl_a ($n_{\text{sites}}=55$; $n_{\text{samples}}=264$; orange and green dots). Data were collected between January 2013 and June 2016.

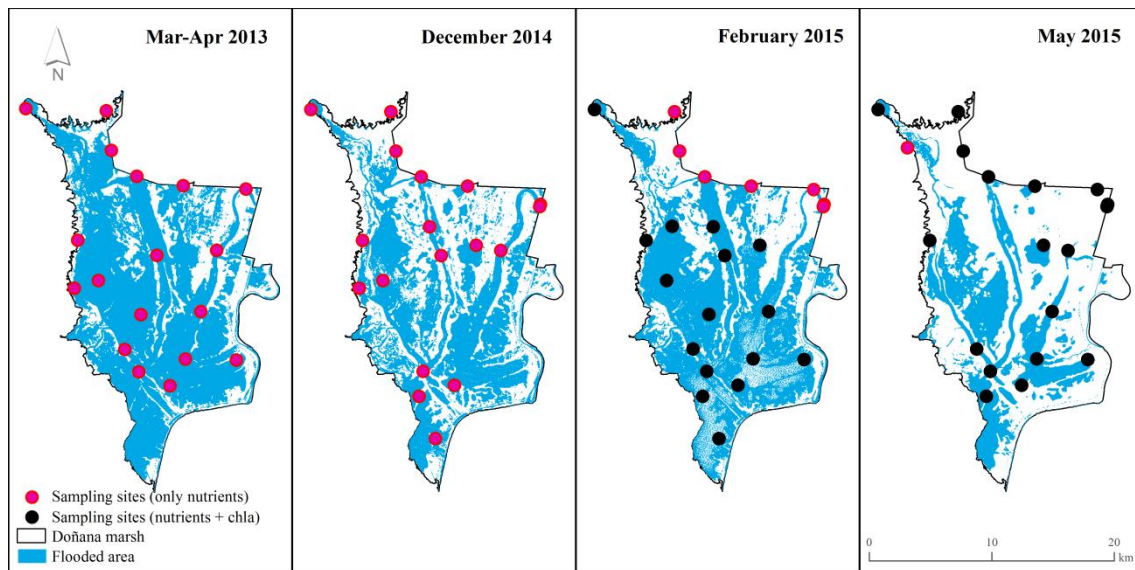


Figure S4. Location of sampling points within the Doñana marsh from which we obtained the nutrient (pink and black dots) and chl a (only black dots) data used to perform linear regression models over the four sampling periods with the highest flooding level during our entire study period. Inundation masks represent the flooded area for the following dates: 19/04/2013, 10/12/2014, 28/02/2015 and 11/05/2015.

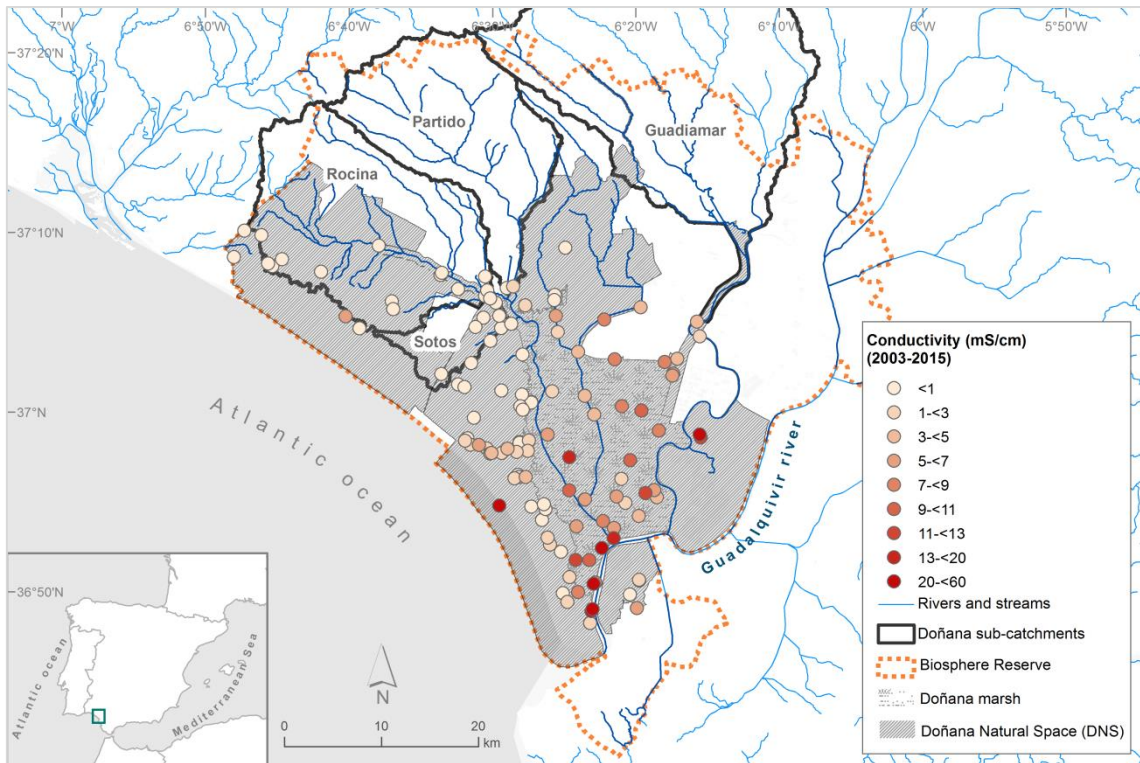


Figure S5. Conductivity (mS cm^{-1}) gradient across Doñana Natural Space (shaded area). Dots represent mean conductivity values of all samples collected at each point between 1st April and 30th June during the period 2003 to 2015. These data were collected by the Monitoring Team for Natural Resources and Processes of the Doñana Biological Station using a WTW (Weilheim, Germany) Multi-340i handheld meter for *in situ* measurements. Many of these points were not used in our own study (e.g. those in dune ponds).

Table S1. Summary of conductivity, depth and stable isotope data collected between January 2013 and June 2016 in the marsh and the three streams.

| Variables | Waterbody | | | |
|--|--------------------|-------------------------|---------------------|--------------------|
| | Partido (n= 92) | Rocina/Sotos (n= 79) | Guadamar (n= 33) | Marsh (n= 137) |
| Cond ($\mu\text{S cm}^{-1}$) | | | | |
| Min. | 267 | 222 | 325 | 156 |
| 1st Qu. | 945 | 393.5 | 719 | 1070 |
| Median | 1162 | 553 | 1439 | 2460 |
| Mean \pm s.d. | 1191 \pm 766.4 | 544.2 \pm 195.8 | 2114 \pm 1908.2 | 4330 \pm 5737.12 |
| 3rd Qu. | 1317 | 626 | 2690 | 5090 |
| Max. | 7860 | 1270 | 7820 | 32900 |
| Depth (cm) | | | | |
| Min. | 1 | 5 | 5 | 1 |
| 1st Qu. | 15 | 10 | 26 | 15 |
| Median | 23 | 20 | 60 | 28 |
| Mean \pm s.d. | 36.2 \pm 52.7 | 26.8 \pm 23.8 | 67.6 \pm 59.2 | 31 \pm 22.1 |
| 3rd Qu. | 40 | 35 | 100 | 42 |
| Max. | 480 | 130 | 300 | 150 |
| $\delta^2\text{H}$ (‰) | | | | |
| Min. | -50.1 | -55.9 | -49 | -52 |
| 1st Qu. | -28 | -29.3 | -35.6 | -21.5 |
| Median | -25.7 | -25.8 | -24.3 | -1.3 |
| Mean \pm s.d. | -26.9 \pm 5.5 | -26.5 \pm 7.3 | -23.8 \pm 13.9 | -1.5 \pm 25.9 |
| 3rd Qu. | -23.9 | -23.1 | -14.9 | 13.1 |
| Max. | -17.9 | -3.4 | 7.5 | 68.2 |

Table S2. Two-way analysis of variance (two-way ANOVA) with nutrient and chlorophyll-*a* (chl_a) concentrations as dependent variables (all log-transformed), and waterbody as categorical predictor. Coefficients for waterbody ‘Marsh’ are not shown because they are aliased, but they are effectively zero. ‘Sampling period’ refers to 16 different field campaigns carried out between January 2013 and May 2016. The degrees of freedom of the residuals were 411 for all nutrient effects and 250 for chl_a. This table corresponds to Fig. 9 in the manuscript.

| Variable | Effect | Level of effect | Estimate ± S.E. | Df | F | P | Model parameters | |
|-----------------|------------------|-----------------|-----------------|-------|---------|---------|-------------------------------|--------------------------|
| Log PO4 | <i>Intercept</i> | | -3.92 ± 0.40 | 1 | 96.51 | <0.0001 | Adj. R ² F P | 0.42 18.36 <0.0001 |
| | Waterbody | Guadamar | 0.92 ± 0.36 | 3 | 50.03 | <0.0001 | | |
| | | Rocina/Sotos | 1.82 ± 0.28 | | | | | |
| | | Partido | 3.70 ± 0.30 | | | | | |
| Sampling period | | | 15 | 12.02 | <0.0001 | | | |
| Log TP | <i>Intercept</i> | | -1.92 ± 0.15 | 1 | 155.65 | <0.0001 | Adj. R ² F P | 0.49 24.16 <0.0001 |
| | Waterbody | Guadamar | 0.38 ± 0.13 | 3 | 81.69 | <0.0001 | | |
| | | Rocina/Sotos | 1.04 ± 0.10 | | | | | |
| | | Partido | 1.80 ± 0.11 | | | | | |
| Sampling period | | | 15 | 12.65 | <0.0001 | | | |
| Log NH4 | <i>Intercept</i> | | -3.34 ± 0.57 | 1 | 34.00 | <0.0001 | Adj. R ² F P | 0.20 7.18 <0.0001 |
| | Waterbody | Guadamar | 0.79 ± 0.51 | 3 | 25.44 | <0.0001 | | |
| | | Rocina/Sotos | 0.14 ± 0.40 | | | | | |
| | | Partido | 3.31 ± 0.43 | | | | | |
| Sampling period | | | 15 | 3.53 | <0.0001 | | | |
| Log NO3 | <i>Intercept</i> | | -6.63 ± 0.73 | 1 | 82.50 | <0.0001 | Adj. R ² F P | 0.53 28.70 <0.0001 |
| | Waterbody | Guadamar | 5.31 ± 0.66 | 3 | 97.11 | <0.0001 | | |
| | | Rocina/Sotos | 6.49 ± 0.52 | | | | | |
| | | Partido | 9.08 ± 0.55 | | | | | |
| Sampling period | | | 15 | 15.02 | <0.0001 | | | |
| Log NO2 | <i>Intercept</i> | | -5.58 ± 0.31 | 1 | 319.59 | <0.0001 | Adj. R ² F P | 0.50 25.54 <0.0001 |
| | Waterbody | Guadamar | 1.4 ± 0.28 | 3 | 92.92 | <0.0001 | | |
| | | Rocina/Sotos | 1.75 ± 0.22 | | | | | |
| | | Partido | 3.95 ± 0.23 | | | | | |
| Sampling period | | | 15 | 12.06 | <0.0001 | | | |
| Log TN | <i>Intercept</i> | | 1.09 ± 0.10 | 1 | 117.31 | <0.0001 | Adj. R ² F P | 0.52 27.87 <0.0001 |
| | Waterbody | Guadamar | 0.04 ± 0.09 | 3 | 97.11 | <0.0001 | | |
| | | Rocina/Sotos | 0.49 ± 0.07 | | | | | |
| | | Partido | 1.26 ± 0.07 | | | | | |
| Sampling period | | | 15 | 14.02 | <0.0001 | | | |
| Log Chla | <i>Intercept</i> | | -5.57 ± 0.18 | 1 | 968.27 | <0.0001 | Adj. R ² F P | 0.14 4.54 <0.0001 |
| | Waterbody | Guadamar | 1.04 ± 0.26 | 3 | 8.87 | <0.0001 | | |
| | | Rocina/Sotos | 0.54 ± 0.18 | | | | | |
| | | Partido | 0.02 ± 0.19 | | | | | |
| Sampling period | | | 10 | 3.24 | <0.0001 | | | |

Table S3. Surface water flow (m^3s^{-1}) measured at different sites of the Rocina and Partido streams during February, April and May in 2016. We used a handheld Acoustic Doppler Velocimeter (Sontek FlowTracker). Final flow value is the average of several measurements along a transversal transect of the stream.

| Stream sites | Surface water flow (m^3s^{-1}) [mean velocity (m s^{-1})] | | | |
|--|--|-----------------------------|---|--|
| | 25 th Feb. 2016 | 10 th April 2016 | 20 th , *21 st April 2016 | 24 th , **25 th May 2016 |
| Rocina stream – upstream (Ortigas bridge) | 0.0079 [0.1156] | 0.0048 [0.0732] | 0.1898 [0.3594] | 0.0141 [0.1652] |
| Rocina stream - downstream (Canariega bridge) | 0.0734 [0.2860] | 0.0325 [0.2030] | 0.2020 [†] [0.5195] | 0.0564 [0.2849] |
| Partido stream – upstream (Azud de la Matanza) | 0.0956 [0.4455] | 0.0472 [0.3299] | 0.6685 [†] [0.3684] | 0.1089 ^{††} [0.4035] |
| Partido stream – downstream (Ajolí bridge) | 0.0727 [0.0971] | 0.0902 [0.0356] | 0.7859 [†] [0.2042] | 0.1770 ^{††} [0.2787] |

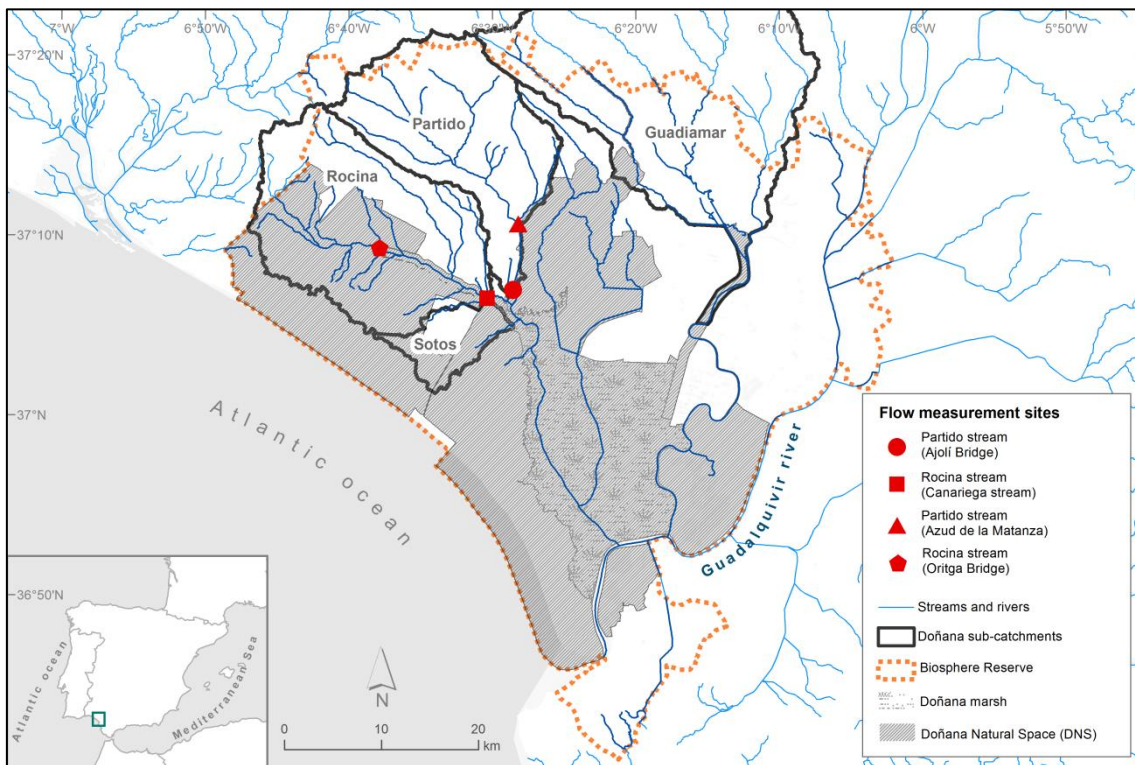


Table S4. Reference and limit values described in the Spanish Royal Decree 817/2015 for assessing the ecological status of the WFD waterbody types included within this study (R-T02, R-T18 and L-T25). Note that for type L-T25 (Marsh, Guadiamar and Soto Grande stream) there are not limit values for any nutrient species.

| Waterbody | WFD code (Spain) | Indicator | Units | Reference conditions | Limits for shifting status class | | | |
|-------------------------------------|------------------|----------------------------|-----------------------|----------------------|----------------------------------|---------------|---------------|----------|
| | | | | | Very good/Good | Good/Moderate | Moderate/Poor | Poor/Bad |
| Partido stream | R-T02 (river) | IBMWP | | 90 | 0.89 | 0.54 | 0.32 | 0.13 |
| | | IMMi-T | | 1 | 0.826 | 0.682 | 0.455 | 0.227 |
| | | IPS | | 14 | 0.94 | 0.71 | 0.47 | 0.24 |
| | | QBR | | 65 | 0.833 | | | |
| | | pH | | | 6.5-8.7 | from 6 to 9 | | |
| | | Oxygen | mg/L | | | 5 | | |
| | | % Oxygen | % | | 70-100 | 60-120 | | |
| | | Amonium | mg NH ₄ /L | | 0.3 | 1 | | |
| | | Phosphates | mg PO ₄ /L | | 0.2 | 0.4 | | |
| | | Nitrates | mg NO ₃ /L | | 20 | 25 | | |
| Rocina stream/Soto Chico stream | R-T18 (river) | IBMWP | | 78 | 0.82 | 0.5 | 0.29 | 0.13 |
| | | IMMi-T | | 1 | 0.844 | 0.696 | 0.464 | 0.232 |
| | | IPS | | 14 | 0.98 | 0.74 | 0.64 | 0.24 |
| | | QBR | | 60 | 0.833 | | | |
| | | pH | | | 6.5-8.7 | from 6 to 9 | | |
| | | Oxygen | mg/L | | | 5 | | |
| | | % Oxygen | % | | 70-100 | 60-120 | | |
| | | Amonium | mg NH ₄ /L | | 0.2 | 0.6 | | |
| | | Phosphates | mg PO ₄ /L | | 0.4 | 0.5 | | |
| | | Nitrates | mg NO ₃ /L | | 10 | 25 | | |
| Marsh/Guadiamar /Soto Grande stream | L-T25 (lake) | IBCAEL | | 6.19 | 0.78 | 0.59 | 0.39 | 0.2 |
| | | Macrophyte richness | Nº species | 23 | | 0.48 | 0.27 | 0.1 |
| | | Eutrophic macrophyte cover | % | 0 | 0.99 | 0.9 | 0.5 | 0.5 |
| | | Exotic macrophyte cover | % | 0 | 1 | 0.95 | 0.75 | 0.5 |
| | | Helophyte cover | % | 80 | 0.88 | 0.75 | 0.37 | 0.13 |
| | | Hydrophyte cover | % | 90 | 0.83 | 0.55 | 0.28 | 0.01 |
| | | pH | | | | (7.5-10) | (≤7.5-≥10) | |

Methods S1. Remote sensing of greenhouse expansion

From each hydrological year (24 in total) we selected one cloud-free Landsat image from autumn and another from spring (48 images in total). For each year, we grouped both autumn and spring images into a single 12-band image (B1: Blue, autumn; B2: Green, autumn; B3: Red, autumn; B4: NIR, autumn; B5: SWIR1, autumn; B6: SWIR2, autumn; B7: Blue, spring; B8: Green, spring; B9: Red, spring; B10: NIR, spring; B11: SWIR1, spring; B12: SWIR2, spring). For each image, we identified and classified different land covers such as greenhouses, urban areas, forested areas and other crops by simultaneously applying eight automatic target detection methods: Matched Filtering (MF) [2], Constrained Energy Minimization (CEM) [3], Adaptive Coherence Estimator (ACE) [4], Spectral Angle Mapper (SAM) [5], Orthogonal Subspace Projection (OSP) [6], Target-Constrained Interference-Minimized Filter (TCIMF) [7], Mixture Tuned Target-Constrained Interference-Minimized Filter (MTTCIMF) [8], and Mixture Tuned Matched Filtering (MTMF) [9]. Afterwards, we used two filtering options (i.e. clumping and sieving) to clean up misdetections and false positives in the category of “greenhouses”. To check the quality of performance of the classification methods, we used the image corresponding to the 2013 hydrological year. We randomly distributed 1,200 points within the area classified as “greenhouse” and used an orthophoto from April 2013 (corresponding to the peak production season for greenhouse crops) to check whether these points fell in or out the real area covered by greenhouses. This work was carried out at the Remote Sensing Lab (LAST) at the Doñana Biological Station (EBD-CSIC, Seville).

References

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