

*Supporting Information for*

## **Dielectrophoretic Characterization and Selection of Non-spherical Flagellate Algae in the Parallel Channels with Right-angle Bipolar Electrodes**

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## Section 1: Calculation of capture rate and escape rate

The calculation formula of flagellate microalgae<sup>1, 2</sup>:

$$\text{Capture Rate} = \frac{N_{i-\text{capture}}}{N_{i-\text{capture}} + N_{i-\text{flowout}}} \quad (\text{S1})$$

where  $N_{i-\text{capture}}$  represents the capture number of cells  $i$  to the electrodes,  $N_{i-\text{flowout}}$  represents flow-out number of cells  $i$  in channels.

The calculation formula of flagellate microalgae<sup>1</sup>:

$$\text{Escape Rate} = \frac{N_{i-\text{escape}}}{N_{i-\text{captured}}} \quad (\text{S2})$$

where  $N_{i-\text{escape}}$  represents the escape number of cells  $i$  from electrodes,  $N_{i-\text{captured}}$  represents the captured number of cells  $i$  at electrodes.

## Section 2: Escape behaviors of flagellate algae

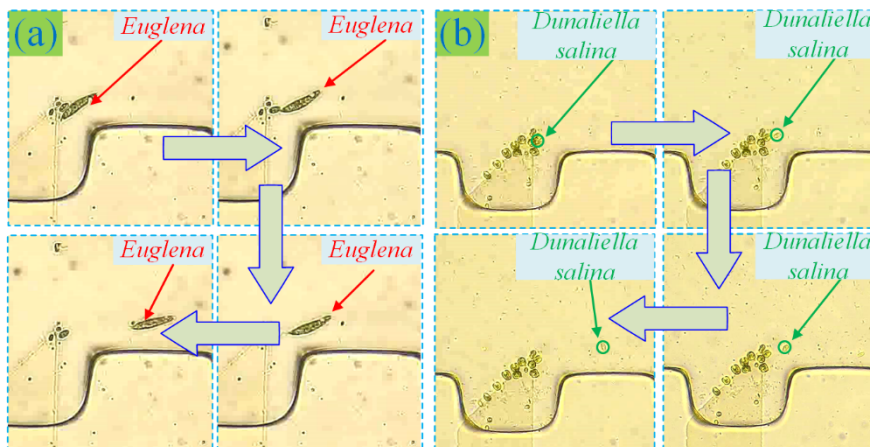


Fig. S1 Escape behaviors of flagellate algae. Escape of (a) *Euglena*, (b) *Dunaliella salina*.

### Section 3: Migration of captured flagellate microalgae between electrodes

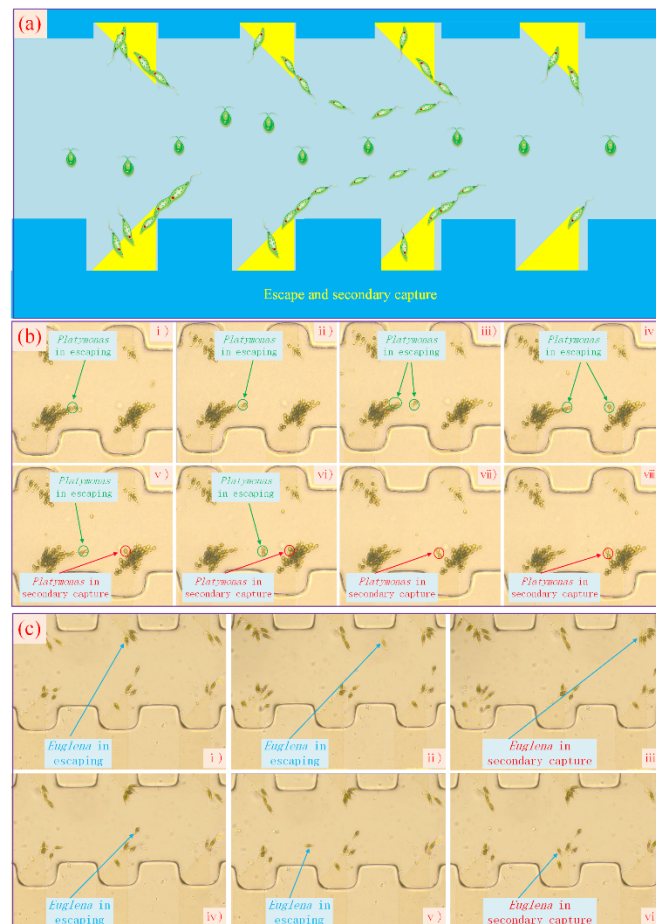


Fig. S2 Migration of captured flagellate microalgae between front and backward bipolar electrodes. (a) The sketch map demonstrating the escape and secondary capture of trapped flagellate microalgae. (b/c) Migration of captured *Platymonas/Euglena* between front and backward bipolar electrodes.

#### Section 4: The capture of *Euglena* with different aspect ratios (ARs)

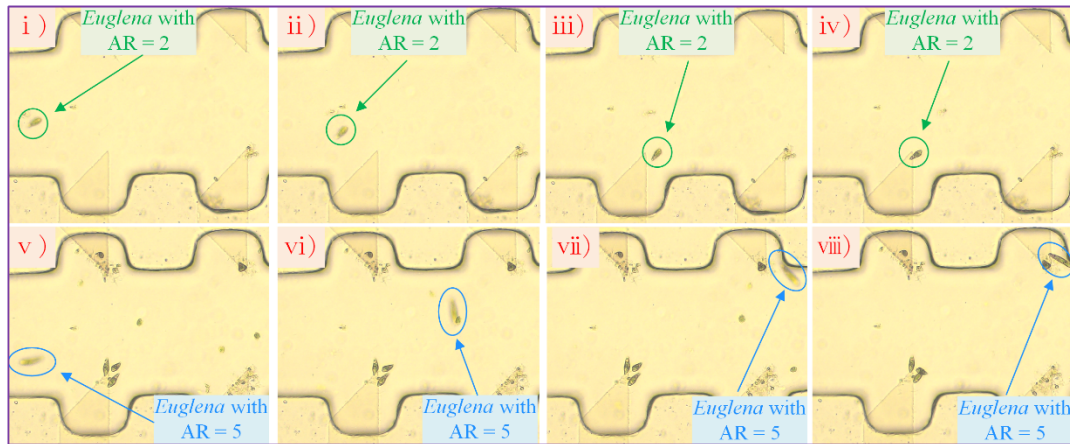


Fig. S3 The capture of *Euglena* with AR=2 and 5.

## Section 5: Comparison with conventional microfluidic separation methods of microalgae

Table S1 Comparison with conventional microfluidic separation methods of microalgae

Author	Samples	Methods	Recovery Purity
Y Wang, et al <sup>3</sup>	<i>Platymonas</i> , <i>Closterium</i> , and polystyrene particles	Dielectrophoresis aroused by 3-dimensional electrode	About 90% for <i>Platymonas</i> , <i>Closterium</i>
Y Wang, et al <sup>4</sup>	<i>Platymonas</i> and impurities	Deterministic lateral displacement	89.4% for <i>Platymonas</i>
D Jiang, et al <sup>5</sup>	<i>H. pluvialis</i> and ciliate	dean-coupled inertial microfluidics	87.1% for <i>H. pluvialis</i>
H Hadady, et al <sup>6</sup>	<i>C. reinhardtii</i> of different lipid content	Oblique interdigitated electrode array	74% for the high-lipid <i>C. reinhardtii</i>
The manuscript	<i>Platymonas</i> and <i>D. salina</i> , <i>H. pluvialis</i> and <i>Euglena</i>	Right-angle bipolar electrode array	92.78% for <i>D. salina</i> and 92.06% for <i>H. pluvialis</i>

## Section 6: Statistical Significance Testing

(a) Separation of *Chlamydomonas reinhardtii* and *Haematococcus pluvialis*

Table S2 Confidence intervals of capture rate(CR) under different frequencies  
(SD:standard deviation, CI: confidence interval at the confidence level of 95%)

Frequency (MHz)	<i>Chlamydomonas reinhardtii</i>				<i>Haematococcus pluvialis</i>			
	Mean of CR (%)	SD(%)	Lower limit of CI(%)	Upper limit of CI (%)	Mean of CR (%)	SD(%)	Lower limit of CI(%)	Upper limit of CI (%)
0.5	35.00	0.307	34.71	35.30	20.20	1.318	18.94	21.46
1.0	33.00	1.439	31.63	34.38	17.41	0.585	16.85	17.97
2.0	10.16	0.619	9.57	10.75	0	0	0	0
5.0	0	0	0	0	0	0	0	0

Table S3 Confidence intervals of CR with different flow rates

Flow rate( $\mu$ L/h)	<i>Chlamydomonas reinhardtii</i>				<i>Haematococcus pluvialis</i>			
	Mean of CR (%)	SD(%)	Lower limit of CI(%)	Upper limit of CI (%)	Mean of CR (%)	SD(%)	Lower limit of CI(%)	Upper limit of CI (%)
21.6	17.79	0.463	17.35	18.23	21.50	1.08	20.47	22.53
36.0	15.59	0.870	14.76	16.42	0	0	0	0
54.0	14.35	0.460	13.91	14.79	0	0	0	0

Table S4 Confidence intervals of CR under different voltage amplitudes

Voltage( $V_{pp}$ )	<i>Chlamydomonas reinhardtii</i>				<i>Haematococcus pluvialis</i>			
	Mean of CR (%)	SD(%)	Lower limit of CI(%)	Upper limit of CI (%)	Mean of CR (%)	SD(%)	Lower limit of CI(%)	Upper limit of CI (%)
10.00	14.21	0.475	13.76	14.67	0	0	0	0
11.25	17.62	0.659	16.99	18.25	0	0	0	0
12.50	24.40	0.593	23.83	24.96	7.28	0.625	6.68	7.87
13.75	25.48	0.508	24.99	25.97	14.40	0.900	13.54	15.26
15.00	28.80	1.024	27.82	29.77	17.42	0.540	16.90	17.93

(b) separation of *Euglena* and *Haematococcus pluvialis*

Table S5 Confidence intervals of CR under different voltage amplitudes

Voltage( V <sub>pp</sub> )	<i>Euglena</i>				<i>Haematococcus pluvialis</i>			
	Mean of CR (%)	SD(%)	Lower limit of CI(%)	Upper limit of CI (%)	Mean of CR (%)	SD(%)	Lower limit of CI(%)	Upper limit of CI (%)
13.75	60.45	1.412	59.11	61.80	3.94	0.24	3.71	4.17
15.00	66.21	1.636	64.65	67.77	5.76	0.499	5.28	6.23
16.25	83.51	1.271	82.30	84.73	9.78	0.536	9.28	10.30
17.50	85.34	0.657	84.71	85.96	11.79	0.342	11.46	12.12
18.00	89.59	0.663	88.96	90.22	13.45	0.776	12.71	14.19

Table S6 Confidence intervals of escape rate(ER) under different voltage amplitudes

Voltage( V <sub>pp</sub> )	<i>Euglena</i>				<i>Haematococcus pluvialis</i>			
	Mean of ER (%)	SD(%)	Lower limit of CI(%)	Upper limit of CI (%)	Mean of CR (%)	SD(%)	Lower limit of CI(%)	Upper limit of CI (%)
13.75	42.80	1.934	40.95	44.64	100	0	100	100
15.00	38.44	2.032	36.50	40.38	100	0	100	100
16.25	24.83	1.808	23.10	26.55	33.12	3.337	29.94	36.30
17.50	8.61	0.520	8.11	9.10	0	0	0	0
18.00	0	0	0	0	0	0	0	0

Table S7 Confidence intervals of separation purity under different voltage amplitudes

Voltage(V <sub>pp</sub> )	Mean of Purity (%)	SD(%)	Lower limit of CI(%)	Upper limit of CI (%)
13.75	68.63	1.103	67.58	69.68
15.00	70.53	1.468	69.13	71.93
16.25	81.99	0.492	81.52	82.46
17.50	85.53	0.888	84.69	86.38
18.00	92.06	1.054	91.06	93.07



(c) Separation of *Platymonas* and *Dunaliella salina*

Table S8 Confidence intervals of CR under different voltage amplitudes

Voltage( V <sub>PP</sub> )	<i>Platymonas</i>				<i>Dunaliella salina</i>			
	Mean of CR (%)	SD(%)	Lower limit of CI(%)	Upper limit of CI (%)	Mean of CR (%)	SD(%)	Lower limit of CI(%)	Upper limit of CI (%)
15.00	14.57	0.949	13.66	15.47	5.01	0.359	4.66	5.35
17.50	32.69	1.267	31.48	33.90	5.65	0.913	4.78	6.52
20.00	39.44	0.488	38.97	39.90	6.55	0.325	6.24	6.86
22.50	41.84	1.665	40.26	43.43	9.10	0.373	8.75	9.46
25.00	44.24	0.665	43.60	44.87	9.49	0.456	9.05	9.92
27.50	55.50	0.664	54.87	56.13	9.93	0.481	9.47	10.39
32.50	56.96	1.280	55.74	58.18	10.53	0.648	9.91	11.15
35.00	72.39	1.599	70.86	73.91	11.01	0.648	10.39	11.63

Table S9 Confidence intervals of separation purity under different voltage amplitudes

Voltage(V <sub>PP</sub> )	Mean of Purity (%)	SD(%)	Lower limit of CI(%)	Upper limit of CI (%)
15.00	73.61	1.690	72.00	75.22
17.50	84.97	1.700	83.35	86.59
20.00	88.48	0.792	87.72	89.23
22.50	88.28	0.656	87.66	88.91
25.0	88.16	1.572	86.67	89.66
27.50	91.13	0.926	90.24	92.01
32.50	91.39	1.008	90.42	92.35
35.00	92.78	1.027	91.80	93.76

(d) Separation of Live and Dead *Euglena*

Table S10 Confidence intervals of CR under different voltage amplitudes

Status	Mean of CR(%)	SD(%)	Lower limit of CI(%)	Upper limit of CI (%)
Live	99.06	0.880	98.22	99.90
Dead	0	0	0	0

Table S11 Confidence intervals of separation purity under different voltage amplitudes

Status	Mean of Purity (%)	SD(%)	Lower limit of CI(%)	Upper limit of CI (%)
Live	100	0	100	100
Dead	97.12	2.647	94.60	99.65

## Section 7: Videos

Video S1 Dielectrophoretic assembly of *Euglena* and *H. pluvialis* at  $A=17.5 V_{pp}$  and  $f=1$  MHz

Video S2 Separation of *H. pluvialis* and *C. reinhardtii* at  $A=12.5 V_{pp}$  and  $f=1$  MHz

Video S3 Separation of *Euglena* and *Platymonas* at  $A=50 V_{pp}$  and  $f=1$  MHz

Video S4 Separation of *Platymonas* and *Dunaliella salina* at  $A=27.5 V_{pp}$  and  $f=1$  MHz

Video S5 Separation of live and dead *Euglena* at  $A=50 V_{pp}$  and  $f=1$  MHz

## Reference

1. C. Lu, J. Xu, J. Han, X. Li, N. Xue, J. Li, W. Wu, X. Sun, Y. Wang, Q. Ouyang, G. Yang and C. Luo, *Lab Chip*, 2020, **20**, 4094-4105.
2. D. Yin, A. Shi, B. Zhou, M. Wang, G. Xu, M. Shen, X. Zhu and X. Shi, *Langmuir*, 2022, **38**, 11080-11086.
3. Y. Wang, J. Wang, X. Wu, Z. Jiang and W. Wang, *Electrophoresis*, 2019, **40**, 969-978.
4. Y. Wang, J. Wang, Y. Wu and J. Dong, *Journal of Chemical Technology & Biotechnology*, 2021, **96**, 2228-2237.
5. D. Jiang, L. Wang, Y. Liu, X. Huo, J. Lin and L. Li, *J Sep Sci*, 2022, **45**, 3900-3908.
6. H. Hadady, D. Redelman, S. R. Hiibel and E. J. Geiger, *AIMS Biophysics*, 2016, **3**, 398-414.