Table 1. Isochrysis galbana. Composition of main culture in terms of culture volume. Data for ammonium and intracellular amino acids (InAA) are given in terms of nitrogen. Extracted chlorophylls were measured by fluorometry (fl) or spectrophotometry (sp).

-: data missing due to instrument malfunction or, for spectrophotometric pigment analysis (sp), because the excessive volumes of culture required for first half of the experiment meant that samples were not taken

| Time (d) | Cells (10 ⁶ ml ⁻¹) | Biovolume (nl ml ⁻¹) | Ammonium (μg N ml ⁻¹) | Cell C (µg C ml ⁻¹) | Cell N (μg N ml ⁻¹) | InAA (ng N ml ⁻¹) | Chlorophyll (ng nl ⁻¹) | | | Carotenoid |
|--------------|--|-------------------------------------|-----------------------------------|------------------------------------|------------------------------------|----------------------------------|------------------------------------|--------|--------|--------------------------|
| | | | | | | | a (fl) | a (sp) | c (sp) | (mg N nl ⁻¹) |
| 0.0 | 0.04 | 2.0 | 1.411 | 0.60 | 0.03 | 0.48 | 4 | 4 | 1 | 13 |
| 0.5 | 0.04 | 2.2 | 1.400 | 0.61 | 0.08 | 0.86 | 3 | - | _ | _ |
| 1.0 | 0.04 | 2.1 | 1.383 | 0.58 | 0.08 | 0.72 | 3 | _ | | _ |
| 1.5 | 0.04 | 2.1 | 1.355 | - | 0.12 | 1.02 | 3 | _ | **** | _ |
| 2.0 | 0.04 | 2.1 | 1.355 | 0.68 | 0.12 | 1.13 | 3 | _ | _ | _ |
| 2.5 | 0.05 | 2.6 | 1.350 | 0.66 | _ | 1.25 | 4 | | _ | _ |
| 3.0 | 0.05 | - | 1.355 | 0.75 | _ | 1.93 | 6 | | _ | _ |
| 3.5 | 0.05 | 2.5 | 1.333 | 2.29 | 0.38 | 2.27 | 9 | _ | _ | - |
| 4.0 | 0.04 | 2.1 | 1.316 | 2.91 | 0.92 | 2.29 | 9 | _ | - | _ |
| 4.5 | 0.06 | 2.9 | 1.288 | 1.75 | 0.61 | 2.49 | 14 | _ | _ | _ |
| 5.0 | 0.05 | 2.3 | 1.288 | 1.49 | 0.26 | 2.77 | 14 | - | _ | _ |
| 5.5 | 0.05 | 3.1 | 1.260 | 2.14 | 0.27 | 3.90 | 17 | - | | _ |
| 6.0 | 0.10 | 4.4 | 1.257 | 1.58 | 0.64 | 4.14 | 18 | _ | _ | _ |
| 6.5 | 0.10 | 5.8 | 1.226 | 2.07 | 0.36 | 5.07 | 41 | _ | _ | _ |
| 7.0 | 0.15 | 7.4 | 1.226 | 2.04 | 0.36 | 5.40 | 41 | | _ | _ |
| 7.5 | 0.15 | 9.5 | 1.092 | 2.66 | 0.45 | 7.81 | 50 | 53 | 52 | 35 |
| 8.0 | 0.23 | 9.8 | 1.092 | 2.25 | 0.52 | 8.14 | 53 | 67 | 70 | 54 |
| 8.5 | 0.22 | 13.4 | 0.935 | 4.12 | 0.57 | 11.67 | 68 | 78 | 43 | 75 |
| 9.0 | 0.28 | 13.9 | 0.932 | 3.93 | 0.64 | 11.12 | 84 | - | _ | _ |
| 9.5 | 0.29 | 18.6 | 0.806 | 5.03 | 0.72 | 15.76 | 133 | 148 | 46 | 134 |
| 10.0 | 0.45 | 18.9 | 0.795 | 4.42 | 0.60 | 16.14 | 133 | 152 | 55 | 134 |
| 10.5 | 0.46 | 31.3 | 0.577 | 6.06 | 0.74 | 24.39 | 178 | 205 | 75 | 192 |
| 11.0 | 0.66 | 28.1 | 0.566 | 5.69 | 0.79 | 23.16 | 210 | 227 | 80 | 201 |
| 11.5 | 0.66 | 41.0 | 0.274 | 8.42 | 1.01 | 32.66 | 255 | 283 | 104 | 240 |
| 12.0 | 0.89 | 38.6 | 0.291 | 8.46 | 1.29 | 34.13 | 255 | 290 | 115 | 243 |
| 12.5 | 0.89 | 54.8 | 0 | 11.29 | 1.37 | 41.36 | 323 | 376 | 129 | 320 |
| 13.0 | 1.11 | 51.6 | 0 | 11.51 | 1.56 | 39.02 | 336 | 410 | 152 | 369 |
| 13.5 | 1.13 | 59.8 | 0 | 13.83 | 1.53 | 32.42 | 377 | 434 | 160 | 408 |
| 14.0 | 1.33 | 62.2 | Ö | 13.91 | 1.61 | 34.73 | 399 | 385 | 148 | 387 |
| 14.5 | 1.33 | 67.8 | 0 | 15.95 | 1.52 | 32.07 | 377 | 468 | 164 | 486 |
| 15.0 | 1.47 | 70.1 | 0 | 15.06 | 1.53 | 33.43 | 413 | 493 | 181 | 533 |
| | 1.47 | 76.1 76.2 | 0 | 20.03 | 1.60 | 36.44 | 440 | 518 | 206 | 597 |
| 15.5 | 1.50 | 76.2 76.5 | 0 | 16.40 | 1.43 | 35.46 | 395 | 478 | 167 | 557 |
| 16.0 | | 85.2 | 0 | 18.74 | 1.44 | 39.91 | 439 | 515 | 195 | 644 |
| 16.5 17.0 | 1.64 1.82 | 93.3 | 0 | 18.03 | 1.54 | 34.12 | 399 | 468 | 136 | 594 |
| | | | 0 | 16.03 | 1.54 | J4.12 — | <i></i> | | _ | _ |
| 18.0 | 1.98 2.00 | 87.9 105.2 | 0 | 23.20 | 1.59 | 34.83 | 395 | 481 | 164 | 693 |
| 18.5 | | 105.2 | 0 | 22.02 | 1.32 | 41.05 | 386 | 441 | 151 | 657 |
| 19.0 | 1.93 | 100.4 | 0 | 22.02 | 1.32 — | - | | | - | - |
| 20.0 | 1.95 | 101 1 | | 23.69 | 1.30 | 31.72 | 377 | 442 | 146 | 674 |
| 20.5 | 1.99 | 101.1 | 0 | 23.66 | 1.31 | 30.11 | 368 | 439 | 144 | 693 |
| 21.0 | 1.93 | 93.4 | 0 | 23.00 | 1.31 | 50.11 - | : - | 439 | , , , | - - |
| 21.5 | 1.95 | - 00.2 | 0 | 25.80 | _ 1 31 | 30.10 | 368 | 415 | 156 | 717 |
| 22.0 | 2.00 | 90.3 | 0 | 25.80 | 1.31 | 34.24 | 296 | 476 | 153 | 815 |
| 23.0 | 2.00 | 85.8 | 0 | 27.49 | 1.46 | 29.62 | 296 268 | 375 | 119 | 645 |
| 23.5 | 2.00 | 90.4 | 0 | 25.58 | 1.38 | 29.62 27.20 | | 407 | 142 | 753 |
| 24.0 | 1.94 | 84.2 | 0 | 25.53 | 1.38 | | 341 | | | |
| 24.5 | 1.90 | 86.7 | 0 | 27.05 | 1.44 | 30.83 | 341 | 407 | 150 | 792 731 |
| 25.0 | 1.99 | 90.8 | 0 | 26.02 | 1.36 | 31.36 | 323 | 375 | 126 | /31 |

into 5-litre and 3-litre flasks. Cultures were grown at $18\,^{\circ}$ C, illuminated at $100\,\mu\text{mol m}^{-2}\,\text{s}^{-1}$, with a $12\,\text{h}$ light: $12\,\text{h}$ dark cycle, and occasional shaking. No aeration was provided. Samples were withdrawn aseptically through a syphon tube with the inlet near to the bottom of the flask.

The main culture (initially of 5 litres) was inoculated by the aseptic addition of stationary-phase culture grown on the same medium under the same conditions to give an addition of 3% (v:v inoculum: medium). This culture was sampled at least every 12 h (30 min before the end of the light or dark phase), and more fre-

quently following refeeding during the light (Day 0), for a 24 h period during the exponential phase of growth (Day 9), and whilst entering the stationary phase (Day 15). At the end of the stationary phase (Day 24), a portion of the culture was used to inoculate 2 litres of fresh medium, and the response to refeeding a stationary-phase culture in darkness was examined.

An additional 2-litre volume was also inoculated at Day 0, again with a 3% inoculum from the same initial culture. This culture was sampled occasionally until it had just exhausted the nitrogen source, at which time portions were taken and used to inoculate fresh