Hansatech Oxygen Probe

May 1, 2012
DW 3 chamber

Prepare potassium chloride solution
Prepare sodium dithionite solution

1. Prepare oxygen electrode
    - Clean electrode with fine paper (microscope paper?).
    - Add a drop of KCl to top of probemound and another couple of drops in electrode well (everywhere there is a metallic material)
    - Cut a square of cigarette paper without touching with your hands (no oils on paper). Use forceps and a pair of scissors. Place the paper on top of the electrode mound.
    - Cut a square of membrane material (Teflon paper) without touching with hands. Make sure there are no bubbles underneath membrane. Can massage bubbles gently with forceps.
    - Place two o-rings onto electrode. One larger o-ring fits into the outer well-space on top side of electrode.
    - The second smaller o-ring will go around the mound in the center of the electrode. To do so, place o-ring on the tip of the o-ring applicator (a black, plastic cylinder about the size of a lipstick. Wrap the o-ring around the tip. Place the tip on the top of the mound (right on the membrane) and press down until oring ends up at base of the mound. The o-ring should now hold the paper+membrane in place over the mound.

2. Open the *Oxygraph Plus 1.01* software.

    a. A dialogue stating that calibration has expired appears
        →click *OK*

    b. You may or may not get a menu asks which port you are using, usually port 1 or 2 (using serial cable, USB may be port 3 or 4)
        →click *OK*

    c. A *Box ID* menu appears
        →click *OK*

3. Turn the Stirrer on.
    → click *Hardware*
        → click *stirrer speed*
            → press the *On* button, then *OK*

    This should start the magnetic stir bar to rotate. Take a look in the DW3 chamber to confirm this is actually moving.

4. Calibrate the probe with seawater

    Fill the chamber with oxygenated dH20, dH20 should be held in (60mL) falcon tubes that are placed in the water chiller to obtain appropriate temperature. Then shake the water for

30 seconds before pouring into the chamber. Make sure the bottom of the plunger is in contact with the water surface. You can adjust the vertical height of the plunger by rotating the top plate along its threads.

→ click *Calibration*
→ click *Liquid Phase Calibration*

A menu pops up, *Calibration (Liquid Phase) - Step 1 of 5*

Note: we have to run a work-around conversion here because Hansatech is designed to be calibrated with dH20, not seawater. *Work-around:* Use Table 1 to find the max O2 for your given temperature of seawater. Take that concentration and then use Table 2 to find a new temperature (to convert back to dH20 scale)

→ now input the new temperature value for whatever your water temp is, click *OK*

A menu pops up, *Calibration (Liquid Phase) - Step 2 of 5*
→ set the stirrer speed appropriately (probably 100), click *OK*

A menu pops up, *Calibration (Liquid Phase) - Step 3 of 5*
The software will read the oxygen concentration in the chamber to establish a 100% saturation point. Wait until the dialogue reads *Plateau reached, press OK to continue*. This may take several minutes.
→ click *OK*

A menu pops up, *Calibration (Liquid Phase) - Step 4 of 5*
Menu reads *Establish zero oxygen in the chamber*
Remove the plunger top from the chamber and add some **sodium dithionite** solution to the chamber. This will remove oxygen from the water. **Pubble Nitrogen! instead**
→ click *OK*

A menu pops up, *Calibration (Liquid Phase) - Step 5 of 5*
Allow for signal to plateau. The software is now reading the low point (0% saturation) for the calibration.
→ click *OK*

Two more menus appear
→ click *OK* and *save*

Rinse dithionite solution out of chamber (liquid needs to be disposed of properly)

5. Place your own organism in the chamber and measure.

Table 1. Max O2 concentrations for seawater

Solubility of oxygen in water at various temperatures and pressures
[In milligrams per liter. Values based on Weiss (1970). C, degrees Celsius; mmHg, millimeters of mercury]

Temp. C	Atmospheric pressure, mmHg																			
	760.0	750.0	740.0	730.0	720.0	710.0	700.0	690.0	680.0	670.0	660.0	650.0	640.0	630.0	620.0	610.0	600.0	590.0	580.0	570.0
0	14.6	14.4	14.2	14.0	13.8	13.6	13.4	13.2	13.0	12.8	12.7	12.5	12.3	12.1	11.9	11.7	11.5	11.3	11.1	10.9
1	14.2	14.0	13.8	13.6	13.4	13.2	13.1	12.9	12.7	12.5	12.3	12.1	11.9	11.7	11.6	11.4	11.2	11.0	10.8	10.6
2	13.8	13.6	13.4	13.3	13.1	12.9	12.7	12.5	12.3	12.2	12.0	11.8	11.6	11.4	11.2	11.1	10.9	10.7	10.5	10.3
3	13.4	13.3	13.1	12.9	12.7	12.5	12.4	12.2	12.0	11.8	11.7	11.5	11.3	11.1	10.9	10.8	10.6	10.4	10.2	10.0
4	13.1	12.9	12.7	12.6	12.4	12.2	12.0	11.9	11.7	11.5	11.3	11.2	11.0	10.8	10.7	10.5	10.3	10.1	10.0	9.8
5	12.7	12.6	12.4	12.2	12.1	11.9	11.7	11.6	11.4	11.2	11.1	10.9	10.7	10.5	10.4	10.2	10.0	9.9	9.7	9.5
6	12.4	12.3	12.1	11.9	11.8	11.6	11.4	11.3	11.1	10.9	10.8	10.6	10.4	10.3	10.1	9.9	9.8	9.6	9.5	9.3
7	12.1	12.0	11.8	11.6	11.5	11.3	11.1	11.0	10.8	10.7	10.5	10.3	10.2	10.0	9.9	9.7	9.5	9.4	9.2	9.1
8	11.8	11.7	11.5	11.3	11.2	11.0	10.9	10.7	10.6	10.4	10.2	10.1	9.9	9.8	9.6	9.5	9.3	9.1	9.0	8.8
9	11.5	11.4	11.2	11.1	10.9	10.8	10.6	10.5	10.3	10.2	10.0	9.8	9.7	9.5	9.4	9.2	9.1	8.9	8.8	8.6
10	11.3	11.1	11.0	10.8	10.7	10.5	10.4	10.2	10.1	9.9	9.8	9.6	9.5	9.3	9.2	9.0	8.9	8.7	8.6	8.4
11	11.0	10.9	10.7	10.6	10.4	10.3	10.1	10.0	9.8	9.7	9.5	9.4	9.2	9.1	9.0	8.8	8.7	8.5	8.4	8.2
12	10.8	10.6	10.5	10.3	10.2	10.0	9.9	9.8	9.6	9.5	9.3	9.2	9.0	8.9	8.7	8.6	8.5	8.3	8.2	8.0
13	10.5	10.4	10.2	10.1	10.0	9.8	9.7	9.5	9.4	9.3	9.1	9.0	8.8	8.7	8.5	8.4	8.3	8.1	8.0	7.8
14	10.3	10.1	10.0	9.9	9.7	9.6	9.5	9.3	9.2	9.0	8.9	8.8	8.6	8.5	8.4	8.2	8.1	7.9	7.8	7.7
15	10.1	9.9	9.8	9.7	9.5	9.4	9.3	9.1	9.0	8.8	8.7	8.6	8.4	8.3	8.2	8.0	7.9	7.8	7.6	7.5
16	9.8	9.7	9.6	9.5	9.3	9.2	9.1	8.9	8.8	8.7	8.5	8.4	8.3	8.1	8.0	7.9	7.7	7.6	7.5	7.3
17	9.6	9.5	9.4	9.3	9.1	9.0	8.9	8.7	8.6	8.5	8.3	8.2	8.1	8.0	7.8	7.7	7.6	7.4	7.3	7.2
18	9.4	9.3	9.2	9.1	8.9	8.8	8.7	8.6	8.4	8.3	8.2	8.0	7.9	7.8	7.7	7.5	7.4	7.3	7.2	7.0
19	9.3	9.1	9.0	8.9	8.8	8.6	8.5	8.4	8.3	8.1	8.0	7.9	7.8	7.6	7.5	7.4	7.3	7.1	7.0	6.9
20	9.1	8.9	8.8	8.7	8.6	8.5	8.3	8.2	8.1	8.0	7.8	7.7	7.6	7.5	7.4	7.2	7.1	7.0	6.9	6.7
21	8.9	8.8	8.6	8.5	8.4	8.3	8.2	8.1	7.9	7.8	7.7	7.6	7.5	7.3	7.2	7.1	7.0	6.9	6.7	6.6
22	8.7	8.6	8.5	8.4	8.2	8.1	8.0	7.9	7.8	7.7	7.5	7.4	7.3	7.2	7.1	7.0	6.8	6.7	6.6	6.5
23	8.6	8.4	8.3	8.2	8.1	8.0	7.9	7.7	7.6	7.5	7.4	7.3	7.2	7.0	6.9	6.8	6.7	6.6	6.5	6.4
24	8.4	8.3	8.2	8.0	7.9	7.8	7.7	7.6	7.5	7.4	7.3	7.1	7.0	6.9	6.8	6.7	6.6	6.5	6.3	6.2
25	8.2	8.1	8.0	7.9	7.8	7.7	7.6	7.5	7.3	7.2	7.1	7.0	6.9	6.8	6.7	6.6	6.4	6.3	6.2	6.1
26	8.1	8.0	7.9	7.8	7.6	7.5	7.4	7.3	7.2	7.1	7.0	6.9	6.8	6.7	6.5	6.4	6.3	6.2	6.1	6.0
27	7.9	7.8	7.7	7.6	7.5	7.4	7.3	7.2	7.1	7.0	6.9	6.7	6.6	6.5	6.4	6.3	6.2	6.1	6.0	5.9
28	7.8	7.7	7.6	7.5	7.4	7.3	7.2	7.1	6.9	6.8	6.7	6.6	6.5	6.4	6.3	6.2	6.1	6.0	5.9	5.8
29	7.7	7.6	7.5	7.3	7.2	7.1	7.0	6.9	6.8	6.7	6.6	6.5	6.4	6.3	6.2	6.1	6.0	5.9	5.8	5.7
30	7.5	7.4	7.3	7.2	7.1	7.0	6.9	6.8	6.7	6.6	6.5	6.4	6.3	6.2	6.1	6.0	5.9	5.8	5.7	5.6
31	7.4	7.3	7.2	7.1	7.0	6.9	6.8	6.7	6.6	6.5	6.4	6.3	6.2	6.1	6.0	5.9	5.8	5.7	5.6	5.5
32	7.3	7.2	7.1	7.0	6.9	6.8	6.7	6.6	6.5	6.4	6.3	6.2	6.1	6.0	5.9	5.8	5.7	5.6	5.5	5.4
33	7.2	7.1	7.0	6.9	6.8	6.7	6.6	6.5	6.4	6.3	6.2	6.1	6.0	5.9	5.8	5.7	5.6	5.5	5.4	5.3
34	7.0	6.9	6.8	6.7	6.7	6.6	6.5	6.4	6.3	6.2	6.1	6.0	5.9	5.8	5.7	5.6	5.5	5.4	5.3	5.2
35	6.9	6.8	6.7	6.6	6.5	6.4	6.3	6.3	6.2	6.1	6.0	5.9	5.8	5.7	5.6	5.5	5.4	5.3	5.2	5.1
36	6.8	6.7	6.6	6.5	6.4	6.3	6.2	6.2	6.1	6.0	5.9	5.8	5.7	5.6	5.5	5.4	5.3	5.2	5.1	5.0
37	6.7	6.6	6.5	6.4	6.3	6.2	6.1	6.1	6.0	5.9	5.8	5.7	5.6	5.5	5.4	5.3	5.2	5.1	5.0	4.9
38	6.6	6.5	6.4	6.3	6.2	6.1	6.0	6.0	5.9	5.8	5.7	5.6	5.5	5.4	5.3	5.2	5.1	5.0	4.9	4.8
39	6.5	6.4	6.3	6.2	6.1	6.0	6.0	5.9	5.8	5.7	5.6	5.5	5.4	5.3	5.2	5.1	5.0	4.9	4.8	4.8
40	6.4	6.3	6.2	6.1	6.0	5.9	5.9	5.8	5.7	5.6	5.5	5.4	5.3	5.2	5.1	5.0	4.9	4.9	4.8	4.7

Weiss, R.F., 1970, The solubility of nitrogen, oxygen, and argon in water and seawater: Deep Sea Research, v. 17, no. 4, p. 721-735

Table 2. Conversion Table

Temperature C	O2 concn nmol/ml	O2 concn ppm
0	442.500	14.16
1	430.427	13.7736494
2	418.823	13.4023392
3	407.678	13.0456818
4	396.978	12.7032896
5	386.712	12.374775
6	376.867	12.0597504
7	367.432	11.7578282
8	358.394	11.4686208
9	349.742	11.1917406
10	341.463	10.9268
11	333.544	10.6734114
12	325.975	10.4311872
13	318.742	10.1997398
14	311.834	9.9786816
15	305.238	9.767625
16	298.943	9.5661824
17	292.936	9.3739662
18	287.206	9.1905888
19	281.739	9.0156626
20	276.525	8.8488
21	271.550	8.6896134
22	266.804	8.5377152
23	262.272	8.3927178
24	257.945	8.2542336
25	253.809	8.121875
26	249.852	7.9952544
27	246.062	7.8739842
28	242.427	7.7576768
29	238.936	7.6459446
30	235.575	7.5384
31	232.333	7.4346554
32	229.198	7.3343232
33	226.157	7.2370158
34	223.198	7.1423456
35	220.310	7.049925
36	217.480	6.9593664
37	214.696	6.8702822
38	211.946	6.7822848
39	209.218	6.6949866
40	206.500	6.608

APPENDIX

Fw: Hansatech DW3 Return to INBOX

From: Tim Doyle <TD@ppsystems.com> Add

To: mikenish@u.washington.edu

Date: Fri, 18 Jun 2010 11:35:13 -0400 Show Full Headers

Attachments: Oxygen concentration with temp..xlsx (View | Download)

Dear Mike,

Unfortunately, we don't have a lookup table for seawater calibration built into the software at present. However, the work around is simple if you really want to calibrate using seawater. You simply put in a false value for the temperature to match the O2 value from the Salinity/Temp tables for seawater. For example, entering 26C for the temperature will make the software assume a upper calibration level of 249 nmol/mL rather than 305 nmol/mL. I have attached a spreadsheet with calculated O2 values of air-saturated water with temperature.

I am told by Hansatech that they will look to revise the software to allow the user to enter their own concentration values for the upper calibrant. By default you will be offered the value for air-saturated water at the set temperature and pressure, but then the user can override these to the values for seawater at a given temperature and salinity from the tables.

This is being discussed with Hansatech software engineers next week and we will see if we can implement something quickly. If they can, I will keep you updated.

Please let me know if you still have questions.

Sincerely,
Tim Doyle
Direct Telephone: +1 978-834-0505 - X 20

PP Systems - 110 Haverhill Road, Suite 301 - Amesbury, MA 01913 U.S.A.
Tel: +1 978.834.0505 - Fax: +1 978.834.0545 - Email: TD@ppsystems.com

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