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Appendix

A. Photographs

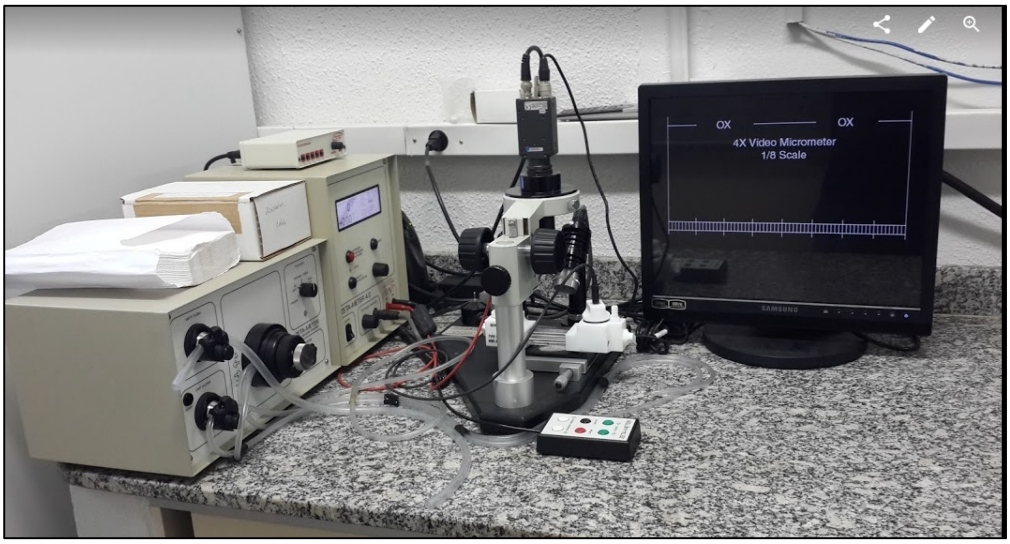

Photo A-1: Zeta Meter System 4.0.

Photo A-2: Crude biosurfactant after the drying stage.

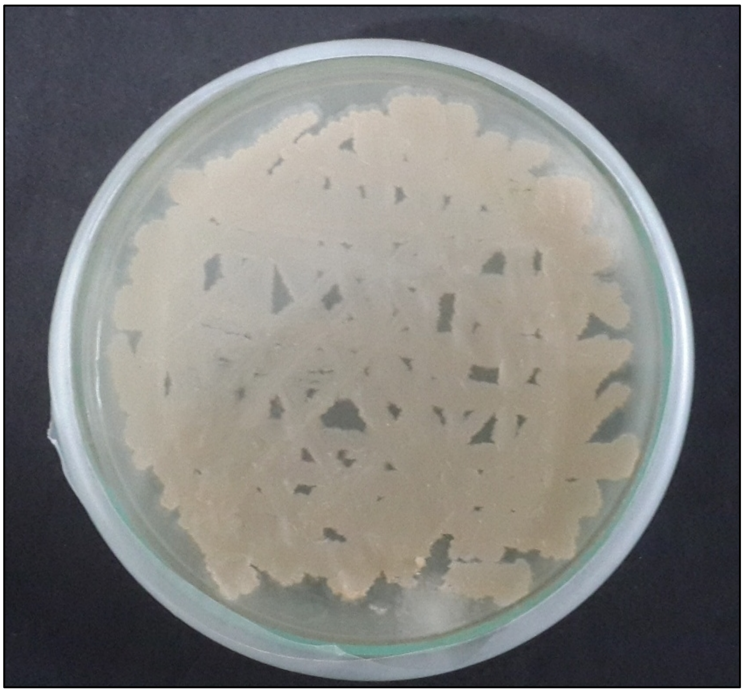

Photo A-3: R. opacus bactéria in solid media.

Photo A-4: Crude biosurfactant concentrate, 6500 ppm.

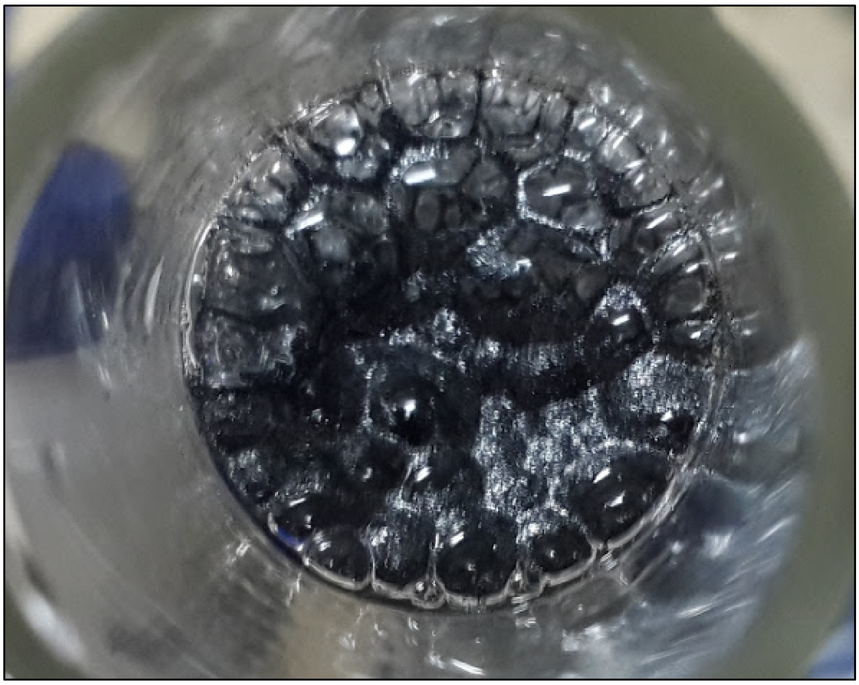

Photo A-5: Biosurfactant froth with hematite particles (+75 -150 µm), NaCl 10^{-3} M, and concentration of 125 ppm and pH 3.

Photo A-6: R.opacus froth with hematite particles (+75 -150 µm), NaCl 10^{-3} M, and concentration of 125 ppm and pH 7.

B. Recorded and estimated data

Table B-1: Zeta potential measurements of hematite before and after interaction with the biosurfactant, size -35 µm, hematite concentration 0.1 g/L.

pH	NaCl 10^{-2} M	NaCl 10^{-3} M	NaCl 10^{-4} M	With BS and NaCl 10^{-3}M
3.0	20.21	25.18	33.66	8.85
4.0	18.64	22.21	27.47	-9.5
5.3	12.9	16.3	22.29	-12.51
6.2	8.7	11.8	16.01	-18.8
7.0	3.09	6.1	10.3	-23.97
8.6	-9.9	-14.03	-22.16	-25.78
9.5	-16.94	-22.04	-28.3	-31.8
11.0	-20.2	-24.25	-30.47	-34.5

Table B-2: Estimated surface potential, based on the Nerst equation.

pH	$\cdot_0$, mV
3.0	272.182
4.0	213.012
5.3	136.091
6.2	82.838
7.0	35.502
8.6	-59.170
9.5	-112.423
11.0	-201.178

Table B-3: Potential distribution in mV, applying the simplified Poison-Boltzman equation for pH 3.

	Background electrolyte concentration (NaCl)		
x (Å)	0.01 mol.dm-3	0.001 mol.dm-3	0.0001 mol.dm-3
0.0	272.18	272.18	272.18
50.0	52.55	161.80	230.90
100.0	10.15	96.18	195.88
150.0	1.96	57.18	166.18
200.0	0.38	33.99	140.97
250.0	0.07	20.20	119.59
300.0	0.01	12.01	101.46
350.0	0.00	7.14	86.07
400.0	0.00	4.24	73.02
450.0	0.00	2.52	61.94
500.0	0.00	1.50	52.55
550.0	0.00	0.89	44.58
600.0	0.00	0.53	37.82
650.0	0.00	0.32	32.08
700.0	0.00	0.19	27.22
750.0	0.00	0.11	23.09
800.0	0.00	0.07	19.59
850.0	0.00	0.04	16.62
900.0	0.00	0.02	14.10
950.0	0.00	0.01	11.96
1000.0	0.00	0.01	10.15
1050.0	0.00	0.00	8.61
1100.0	0.00	0.00	7.30
1150.0	0.00	0.00	6.19
1200.0	0.00	0.00	5.25
1250.0	0.00	0.00	4.46
1300.0	0.00	0.00	3.78
1350.0	0.00	0.00	3.21
1400.0	0.00	0.00	2.72
1450.0	0.00	0.00	2.31

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Table B-4: Potential distribution in mV, applying the simplified Poison-Boltzman equation for pH 5.

x (Å)	Background electrolyte concentration (NaCl) 0.01 mol.dm-3	0.001 mol.dm-3	0.0001 mol.dm-3
0.0	153.84	153.84	153.84
50.0	29.70	91.45	130.51
100.0	5.73	54.36	110.72
150.0	1.11	32.32	93.92
200.0	0.21	19.21	79.68
250.0	0.04	11.42	67.60
300.0	0.01	6.79	57.34
350.0	0.00	4.04	48.65
400.0	0.00	2.40	41.27
450.0	0.00	1.43	35.01
500.0	0.00	0.85	29.70
550.0	0.00	0.50	25.20
600.0	0.00	0.30	21.38
650.0	0.00	0.18	18.13
700.0	0.00	0.11	15.38
750.0	0.00	0.06	13.05
800.0	0.00	0.04	11.07
850.0	0.00	0.02	9.39
900.0	0.00	0.01	7.97
950.0	0.00	0.01	6.76
1000.0	0.00	0.00	5.73
1050.0	0.00	0.00	4.86
1100.0	0.00	0.00	4.13
1150.0	0.00	0.00	3.50
1200.0	0.00	0.00	2.97
1250.0	0.00	0.00	2.52
1300.0	0.00	0.00	2.14
1350.0	0.00	0.00	1.81
1400.0	0.00	0.00	1.54
1450.0	0.00	0.00	1.30

Table B-5: Potential distribution in mV, applying the simplified Poison-Boltzman equation for pH 7.

x (Å)	Background electrolyte concentration (NaCl) 0.01 mol.dm-3	0.001 mol.dm-3	0.0001 mol.dm-3
0.0	35.50	35.50	35.50
50.0	6.85	21.10	30.12
100.0	1.32	12.55	25.55
150.0	0.26	7.46	21.68
200.0	0.05	4.43	18.39
250.0	0.01	2.64	15.60
300.0	0.00	1.57	13.23
350.0	0.00	0.93	11.23
400.0	0.00	0.55	9.52
450.0	0.00	0.33	8.08
500.0	0.00	0.20	6.85
550.0	0.00	0.12	5.81
600.0	0.00	0.07	4.93
650.0	0.00	0.04	4.18
700.0	0.00	0.02	3.55
750.0	0.00	0.01	3.01
800.0	0.00	0.01	2.55
850.0	0.00	0.01	2.17
900.0	0.00	0.00	1.84
950.0	0.00	0.00	1.56
1000.0	0.00	0.00	1.32
1050.0	0.00	0.00	1.12
1100.0	0.00	0.00	0.95
1150.0	0.00	0.00	0.81
1200.0	0.00	0.00	0.69
1250.0	0.00	0.00	0.58
1300.0	0.00	0.00	0.49
1350.0	0.00	0.00	0.42
1400.0	0.00	0.00	0.35
1450.0	0.00	0.00	0.30

Table B-6: Potential distribution in mV, applying the simplified Poison-Boltzman equation for pH 9.

x (Å)	Background electrolyte concentration (NaCl) 0.01 mol.dm-3	0.001 mol.dm-3	0.0001 mol.dm-3
0.0	-82.84	-82.84	-82.84
50.0	-15.99	-49.24	-70.27
100.0	-3.09	-29.27	-59.62
150.0	-0.60	-17.40	-50.58
200.0	-0.12	-10.34	-42.91
250.0	-0.02	-6.15	-36.40
300.0	0.00	-3.66	-30.88
350.0	0.00	-2.17	-26.20
400.0	0.00	-1.29	-22.22
450.0	0.00	-0.77	-18.85
500.0	0.00	-0.46	-15.99
550.0	0.00	-0.27	-13.57
600.0	0.00	-0.16	-11.51
650.0	0.00	-0.10	-9.76
700.0	0.00	-0.06	-8.28
750.0	0.00	-0.03	-7.03
800.0	0.00	-0.02	-5.96
850.0	0.00	-0.01	-5.06
900.0	0.00	-0.01	-4.29
950.0	0.00	0.00	-3.64
1000.0	0.00	0.00	-3.09
1050.0	0.00	0.00	-2.62
1100.0	0.00	0.00	-2.22
1150.0	0.00	0.00	-1.89
1200.0	0.00	0.00	-1.60
1250.0	0.00	0.00	-1.36
1300.0	0.00	0.00	-1.15
1350.0	0.00	0.00	-0.98
1400.0	0.00	0.00	-0.83
1450.0	0.00	0.00	-0.70

Table B-7: Estimated charge density as function of the pH applying the Grahame equation simplified for low potentials.

pH	σ (C/m^2)
3.0	20.05
4.0	15.69
5.3	10.02
6.2	6.10
7.0	2.61
8.6	-4.36
9.5	-8.28
11.0	-14.82

Table B-8: Estimated shear plane distance as function of the pH before and after biosurfactant interaction.

	Shear plane distance, Å	
pH	Only NaCl 10-3 M	Crude BS adsorption
3.0	228.84	93.01
4.0	217.34	100.42
5.3	204.01	104.97
6.2	187.34	117.95
7.0	169.32	161.04
8.6	138.36	54.25
9.5	156.64	68.94
11.0	203.40	79.65

Table B-9: Potential distribution applying the plate capacitor and the Gouy-Chapman model for pH 3, 5, 7, 9 and 11; background electrolyte NaCl 10 -3 M.

pH 3		*pH 5*		*pH 7*		*pH 9*		*pH 11*	
x (Å)	ψ (mV)	x (Å)	ψ (mV)	x (Å)	ψ (mV)	x (Å)	ψ (mV)	x (Å)	ψ (mV)
0	272.18	0.00	153.84	0.00	35.50	0.00	-82.83	0.00	-201.18
93	8.85	104.97	-12.51	161.04	-23.97	60.78	-28.46	79.65	-34.50
143	5.26	154.97	-7.44	211.04	-14.25	110.78	-16.92	129.65	-20.51
193	3.13	204.97	-4.42	261.04	-8.47	160.78	-10.06	179.65	-12.19
243	1.86	254.97	-2.63	311.04	-5.04	210.78	-5.98	229.65	-7.25
293	1.11	304.97	-1.56	361.04	-2.99	260.78	-3.55	279.65	-4.31
343	0.66	354.97	-0.93	411.04	-1.78	310.78	-2.11	329.65	-2.56
393	0.39	404.97	-0.55	461.04	-1.06	360.78	-1.26	379.65	-1.52
443	0.23	454.97	-0.33	511.04	-0.63	410.78	-0.75	429.65	-0.90
493	0.14	504.97	-0.20	561.04	-0.37	460.78	-0.44	479.65	-0.54
543	0.08	554.97	-0.12	611.04	-0.22	510.78	-0.26	529.65	-0.32
593	0.05	604.97	-0.07	661.04	-0.13	560.78	-0.16	579.65	-0.19
643	0.03	654.97	-0.04	711.04	-0.08	610.78	-0.09	629.65	-0.11
693	0.02	704.97	-0.02	761.04	-0.05	660.78	-0.06	679.65	-0.07
743	0.01	754.97	-0.01	811.04	-0.03	710.78	-0.03	729.65	-0.04
793	0.01	804.97	-0.01	861.04	-0.02	760.78	-0.02	779.65	-0.02
843	0.00	854.97	-0.01	911.04	-0.01	810.78	-0.01	829.65	-0.01
893	0.00	904.97	0.00	961.04	-0.01	860.78	-0.01	879.65	-0.01
943	0.00	954.97	0.00	1011.04	0.00	910.78	0.00	929.65	0.00
993	0.00	1004.97	0.00	1061.04	0.00	960.78	0.00	979.65	0.00
1043	0.00	1054.97	0.00	1111.04	0.00	1010.78	0.00	1029.65	0.00
1093	0.00	1104.97	0.00	1161.04	0.00	1060.78	0.00	1079.65	0.00
1143	0.00	1154.97	0.00	1211.04	0.00	1110.78	0.00	1129.65	0.00
1193	0.00	1204.97	0.00	1261.04	0.00	1160.78	0.00	1179.65	0.00
1243	0.00	1254.97	0.00	1311.04	0.00	1210.78	0.00	1229.65	0.00
1293	0.00	1304.97	0.00	1361.04	0.00	1260.78	0.00	1279.65	0.00
1343	0.00	1354.97	0.00	1411.04	0.00	1310.78	0.00	1329.65	0.00
1393	0.00	1404.97	0.00	1461.04	0.00	1360.78	0.00	1379.65	0.00
1443	0.00	1454.97	0.00	1511.04	0.00	1410.78	0.00	1429.65	0.00
1493	0.00	1504.97	0.00	1561.04	0.00	1460.78	0.00	1479.65	0.00
1543	0.00	1554.97	0.00	1611.04	0.00	1510.78	0.00	1529.65	0.00

Table B-10: Hematite recovery in function of pH and R.O. concentration.

N°	pH	Biomass concentration, ppm	Hematite recovery, %
1	3	0	1.50
2	3	25	2.97
3	3	50	2.91
4	3	75	4.95
5	3	100	5.83
6	3	125	6.50
7	3	150	7.80
8	3	175	8.08
9	5	0	5.00
10	5	25	8.91
11	5	50	15.60
12	5	75	21.00
13	5	100	28.10
14	5	125	29.41
15	5	150	33.30
16	5	175	28.57
17	7	0	2.00
18	7	25	25.00
19	7	50	35.40
20	7	75	47.62
21	7	100	44.50
22	7	125	47.62
23	7	150	42.00
24	7	175	33.33
25	9	0	1.00
26	9	25	14.85
27	9	50	17.50
28	9	75	17.82
29	9	100	25.24
30	9	125	20.95
31	9	150	25.71
32	9	175	25.40
33	11	0	2.00
34	11	25	2.86
35	11	50	5.77
36	11	75	7.40
37	11	100	8.10
38	11	125	13.00
39	11	150	17.65
40	11	175	18.40

Table B-11: Hematite recovery in function of pH and BS concentration.

N°	pH	BS concentration, ppm	Hematite recovery %
1	3	0	1.5
2	3	25	26.2
3	3	50	80.7
4	3	75	92.3
5	3	100	91.0
6	3	125	89.0
7	3	150	96.2
8	5	0	4.8
9	5	25	26.1
10	5	50	67.6
11	5	75	74.0
12	5	100	93.4
13	5	125	84.3
14	5	150	89.2
15	7	0	2.4
16	7	25	26.2
17	7	50	73.6
18	7	75	79.2
19	7	100	86.8
20	7	125	81.1
21	7	150	73.0
22	9	0	1.5
23	9	25	6.6
24	9	50	11.7
25	9	75	20.1
26	9	100	43.8
27	9	125	47.2
28	9	150	66.0
29	11	0	2.0
30	11	25	5.7
31	11	50	9.9
32	11	75	13.0
33	11	100	14.8
34	11	125	18.3
35	11	150	22.8

C. Matlab scripts – Diffuse layer modeling

```
%Function that estimates the potential distribution
%of hematite particles -35 um
%x is the distance in A
%y is the pH
%c0 is the background electrolyte concentration
function PotDis=f(x,y,c0)
    PotDis=59.17*(7.6-y).*exp(-x/(3.04/sqrt(c0)));
end
```

```
%Function that shows the potential distribution
%in a 3d graph
%c0 is the background electrolyte concentration
%xmax is the limit of the X axis
%xint is the interval of the distance points
function PlotPot=f(c0,xmax,xint,TitleGraph)
    %plotting 3d
    %graph of diffuse layer
    %defining the x range
    x=0:xint:xmax;
    %defining the pH range
    y=3:11;
    %creating the mesh
    [X, Y] = meshgrid(x,y);
    Z=PotDis(X,Y,c0);
    %plotting 3D
    %subplot(2,2,1);
    surf(X,Y,Z), title(TitleGraph);
    axis([0 xmax 3 11 -300 +300]);
    xlabel('Distance Å'), ylabel('pH'), zlabel('Potential, mV');
    %Plotting the contour
    %subplot(2,2,2);
    figure;
    contour(X,Y,Z,500), xlabel('Distance Å'), ylabel('pH'),
title(TitleGraph);
    H=contourcbar, ylabel(H,'Potential mV');
    PlotPot=0;
end
```

```
%a function that plots the potential distribution
function OnlyCon=f(c0,xmax,xint,TitleGraph)
    %plotting 3d
    %graph of diffuse layer
    %defining the x range
    x=0:xint:xmax;
    %defining the pH range
    y=3:11;
    %creating the mesh
    [X, Y] = meshgrid(x,y);
    Z=PotDis(X,Y,c0);
    %plotting 3D
    %subplot(2,2,1); bad idea!!
%     surf(X,Y,Z), title(TitleGraph);
%     axis([0 xmax 3 11 -300 +300]);
%     xlabel('Distance Å'), ylabel('pH'), zlabel('Potential,
mV');
    %Plotting the contour
    %subplot(2,2,2); bad idea!!
```

```
%    figure;
    contour(X,Y,Z,500), xlabel('Distance Å'), ylabel('pH'),
title(TitleGraph);
    H=contourcbar, ylabel(H,'Potential mV');
    OnlyCon=0;
end
```

```
%function that runs the simulation
%of the electric double layer compression
%for hematite particles where the results
% is shown in a contour graph
function SimPlotPot=f(c0,cf,xmax,xint,n)
    %plotting 3d
    %graph of diffuse layer
    %defining the x range
    x=0:xint:xmax;
    %defining the pH range
    y=3:11;
    %creating the mesh
    [X, Y] = meshgrid(x,y);
    %creating intervals
    ci=linspace(c0,cf,n);
    for i=ci
        Z=PotDis(X,Y,i);
            %plotting 3D
            %subplot(2,2,1); bad idea!!
        %     surf(X,Y,Z), title(TitleGraph);
        %     axis([0 xmax 3 11 -300 +300]);
        %     xlabel('Distance Å'), ylabel('pH'),
zlabel('Potential, mV');
            %Plotting the contour
            %subplot(2,2,2); bad idea!!
        %    figure;

        contour(X,Y,Z,500), xlabel('Distance Å'), ylabel('pH'),
title([num2str(i) ' M of NaCl']);
        H=contourcbar, ylabel(H,'Potential mV');
        pause(0.000009);
    end

    PlotPot=0;
end
```

```
%function that runs the simulation
%of the electric double layer compression
%for hematite particles where the results
% is shown in a 3D graph
function Plot3DPot=f(c0,cf,xmax,xint,n)
    %plotting 3d
    %graph of diffuse layer
    %defining the x range
    x=0:xint:xmax;
    %defining the pH range
    y=3:11;
    %creating the mesh
    [X, Y] = meshgrid(x,y);
    %creating intervals
    ci=linspace(c0,cf,n);
    for i=ci
        Z=PotDis(X,Y,i);
        %plotting 3D
```

```
        %subplot(2,2,1); bad idea!!
        surf(X,Y,Z), title([num2str(i) ' M of NaCl']);
        axis([0 xmax 3 11 -300 +300]);
        xlabel('Distance Å'), ylabel('pH'), zlabel('Potential,
mV');
            %Plotting the contour
            %subplot(2,2,2); bad idea!!
        %    figure;

%         contour(X,Y,Z,500), xlabel('Distance Å'),
ylabel('pH'), title([num2str(i) ' M of NaCl']);
%         H=contourcbar, ylabel(H,'Potential mV');
        pause(0.00009);
    end

    PlotPot=0;
end
```

```
%Final script that creates the 3D graphs
PlotPot(1e-4,200,5,'A) 10-2 M NaCl');
PlotPot(1e-3,500,10,'B) 10-3 M NaCl');
PlotPot(1e-4,1500,50,'C) 10-4 M NaCl');

%Final script that creates the contour graphs
OnlyCon(1e-4,200,5,'A) 10-2 M NaCl');
OnlyCon(1e-3,500,10,'B) 10-3 M NaCl');
OnlyCon(1e-4,1500,50,'C) 10-4 M NaCl');
```

```
% Animation of the electric double layer compression
% in a contour plot
c0=1e-4;
cf=1e-2
%number of intervals
n=30
% create a vector linearly separated
ci=linspace(c0,cf,n);
for i=ci
    LabelS= [num2str(i) ' M NaCl'];
    OnlyCon(i,500,10,LabelS);
    pause(0.000000009);
end
```

```
%Electric double layer compression
figure('units','normalized','position',[0 0 1 1]);
%in a 3d plot
MovPlot(1e-4,1e-2,200,10,30);
%in a contour graph
MovPlot2(1e-4,1e-2,500,10,500);
```

D. FTIR spectras

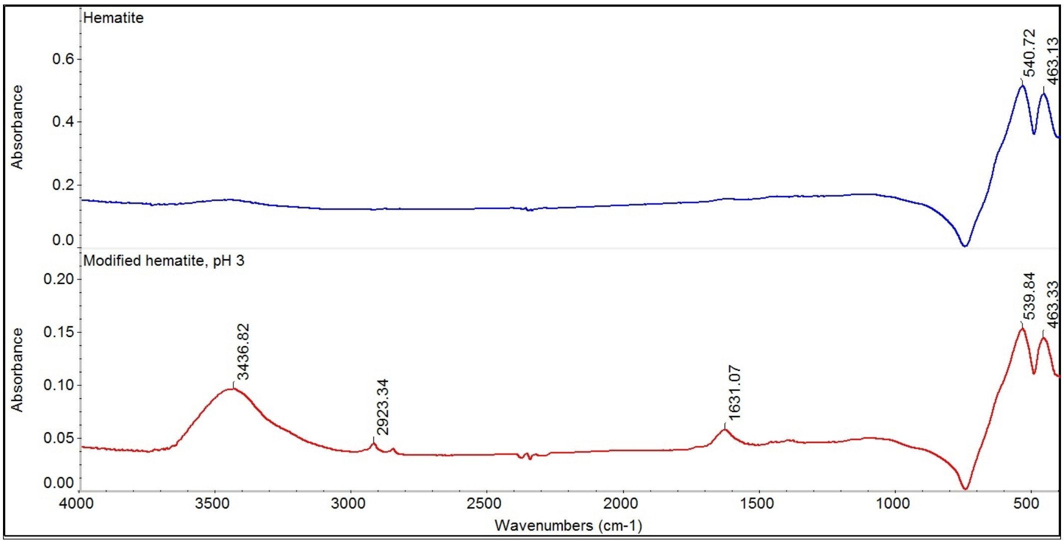

Fig. D.1: FTIR spectra of hematite particles (-35 µm) before and after interaction with the Rhodococcus opacus' crude biosurfactant at pH 3.

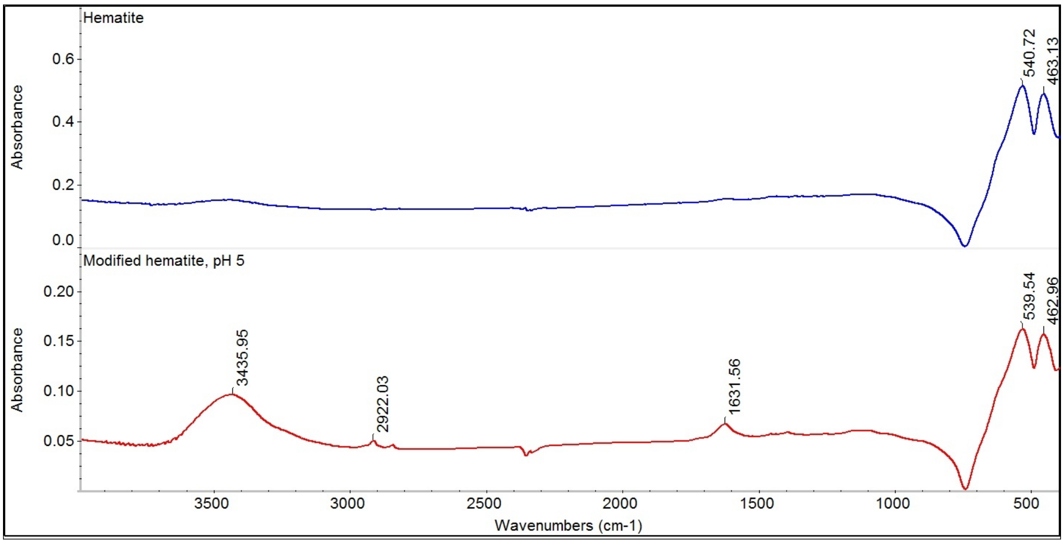

Fig. D.2: FTIR spectra of hematite particles (-35 µm) before and after interaction with the Rhodococcus opacus' crude biosurfactant at pH 5.

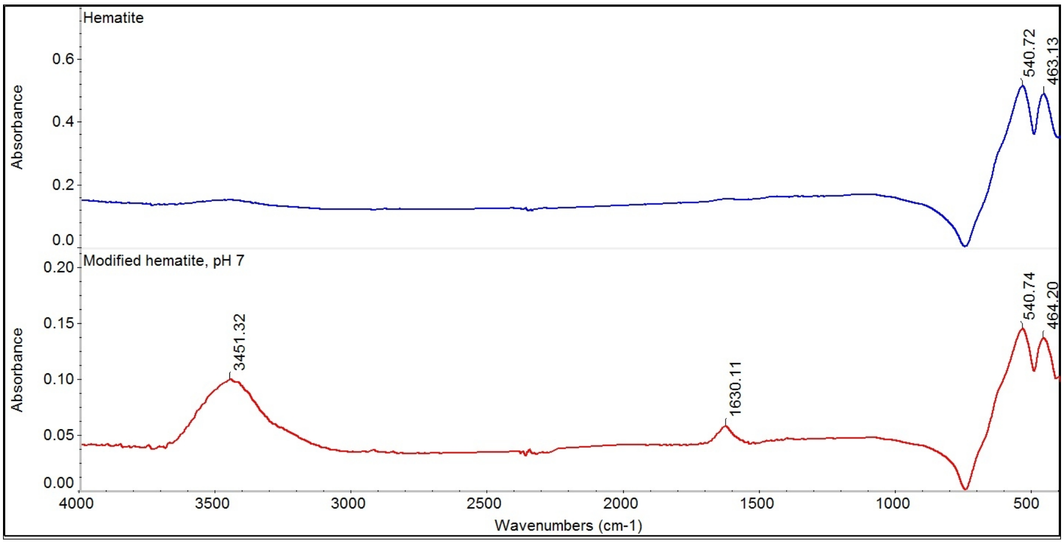

Fig. D.3: FTIR spectra of hematite particles (-35 µm) before and after interaction with the Rhodococcus opacus' crude biosurfactant at pH 7.

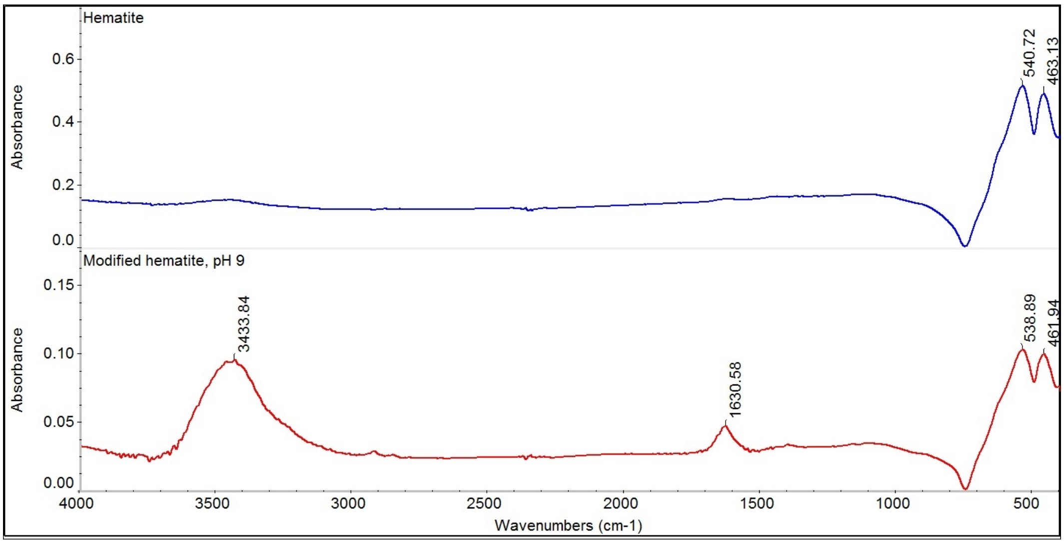

Fig. D.4: FTIR spectra of hematite particles (-35 µm) before and after interaction with the Rhodococcus opacus' crude biosurfactant at pH 9.