Figure 1

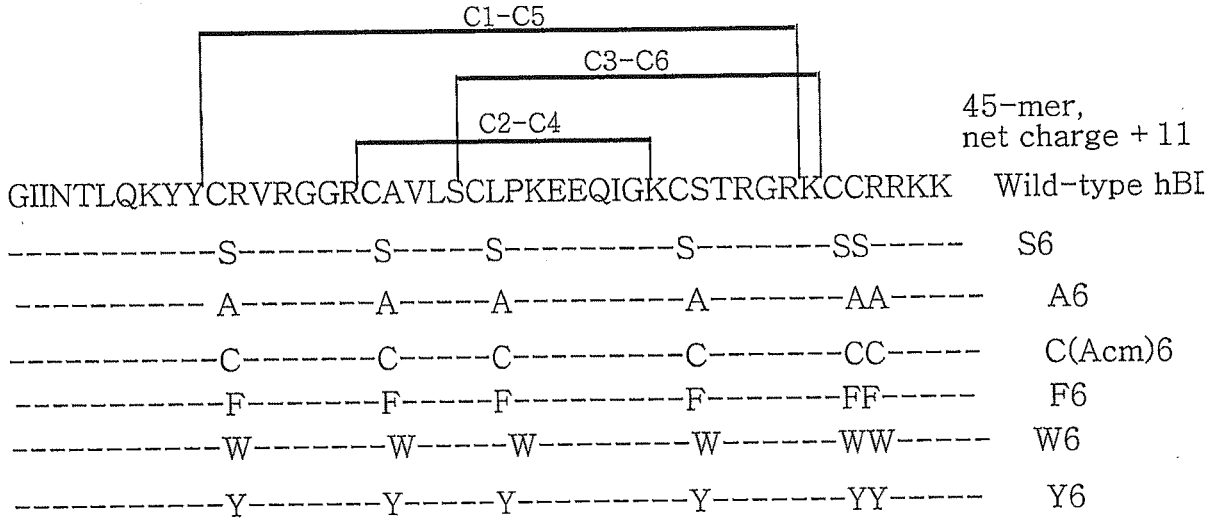

C1-C5
C3-C6
C2-C4
45-mer,
net charge + 11
GIINTLQKYYCRVRGGRCAVLSCLPKEEQIGKCSTRGRKCCRRKK Wild-type hBI
----------S-------S-----S---------S-------SS----- S6
---------- A-------A-----A---------A-------AA----- A6
----------C-------C-----C---------C-------CC----- C(Acm)6
----------F-------F-----F---------F-------FF----- F6
----------W-------W-----W--------W------WW----- W6
----------Y-------Y-----Y---------Y-------YY----- Y6

Figure 2A

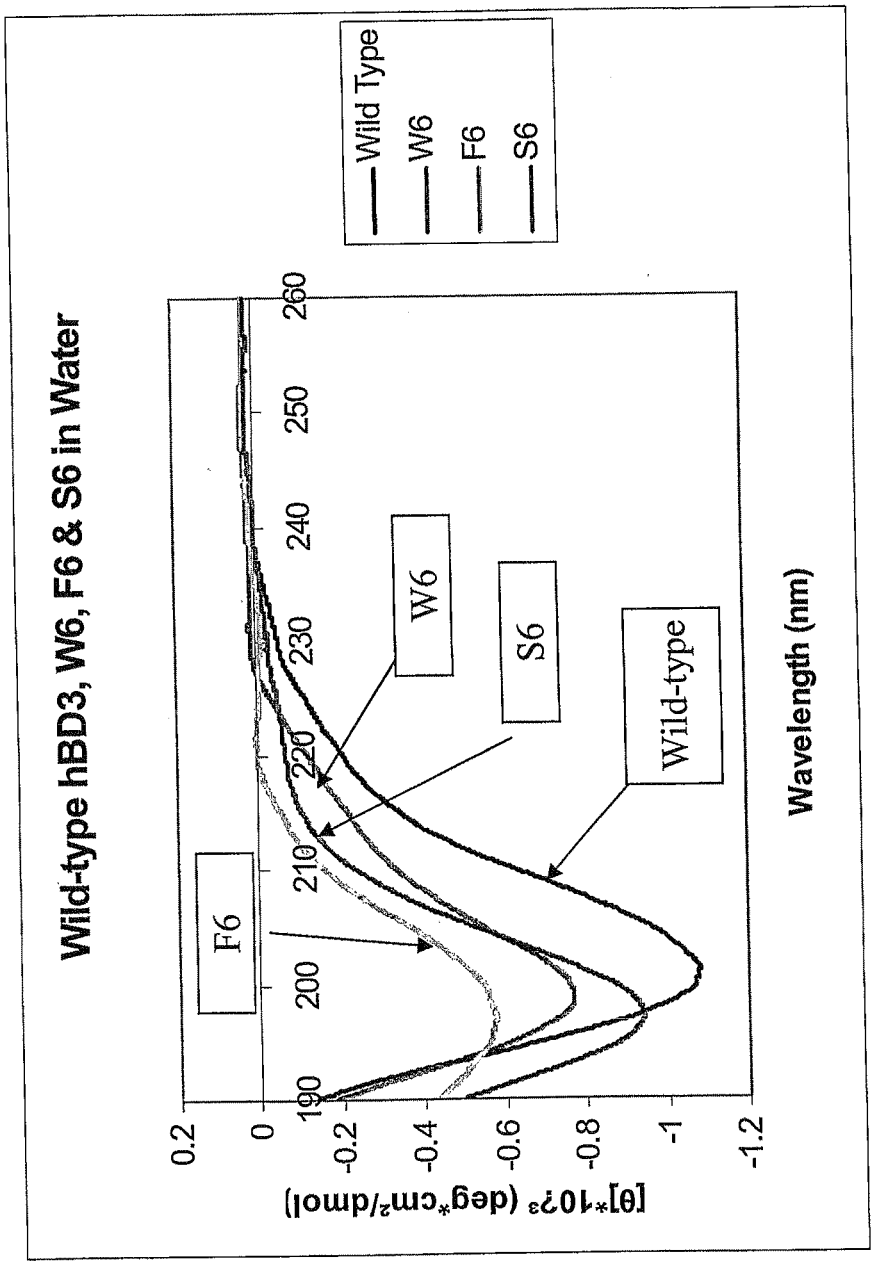

Wild-type hBD3, W6, F6 & S6 in Water
Wild Type
W6
F6
S6
F6
W6
S6
Wild-type
[θ]*10?³ (deg*cm²/dmol)
Wavelength (nm)
0.2
0
-0.2
-0.4
-0.6
-0.8
-1
-1.2
190
200
210
220
230
240
250
260

Figure 2B

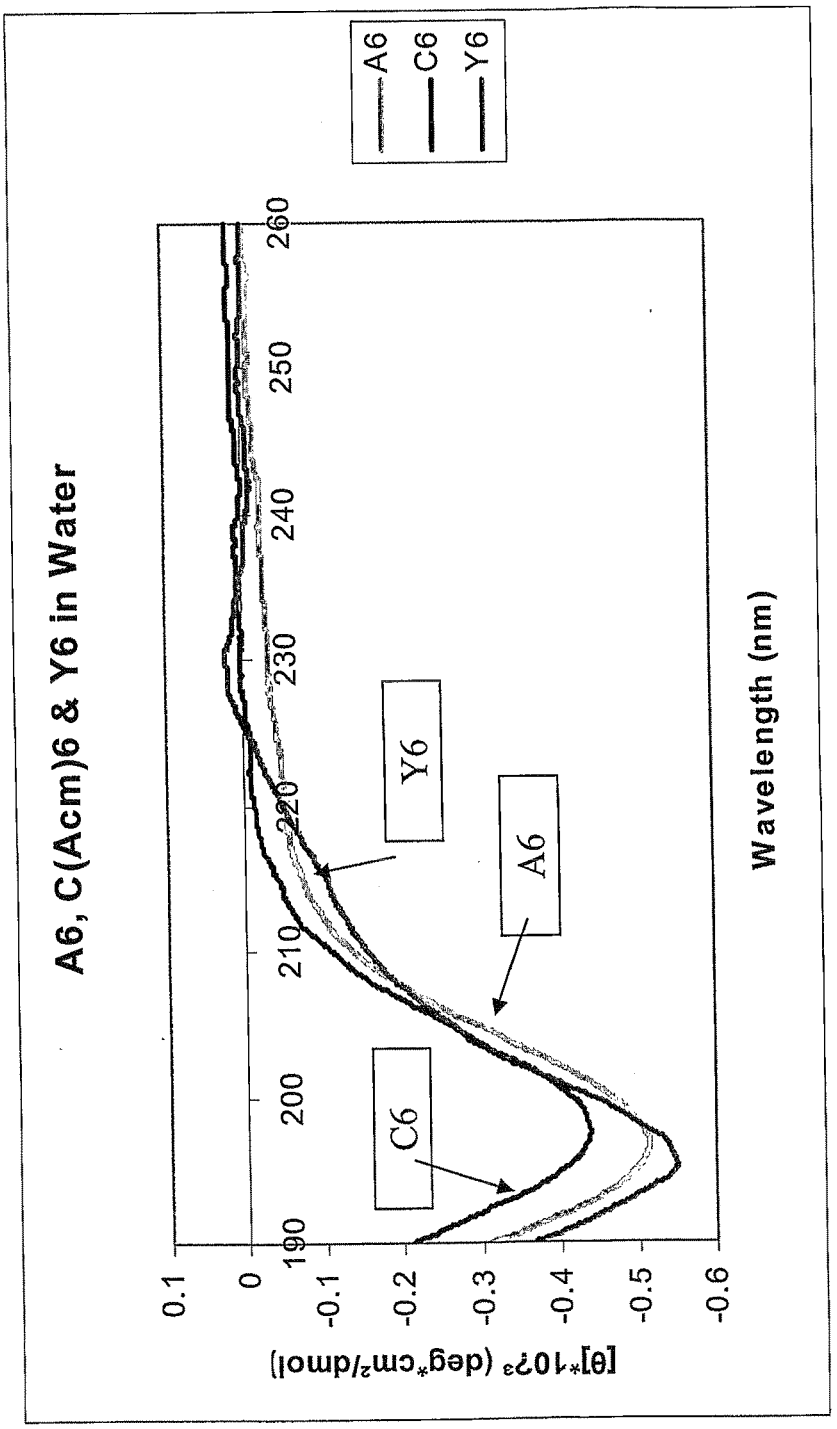

A6, C(Acm)6 & Y6 in Water
A6
C6
Y6
Y6
A6
C6
Wavelength (nm)
[θ]*10?³ (deg*cm²/dmol)
0.1
0
-0.1
-0.2
-0.3
-0.4
-0.5
-0.6
190
200
210
220
230
240
250
260

Figure 3A

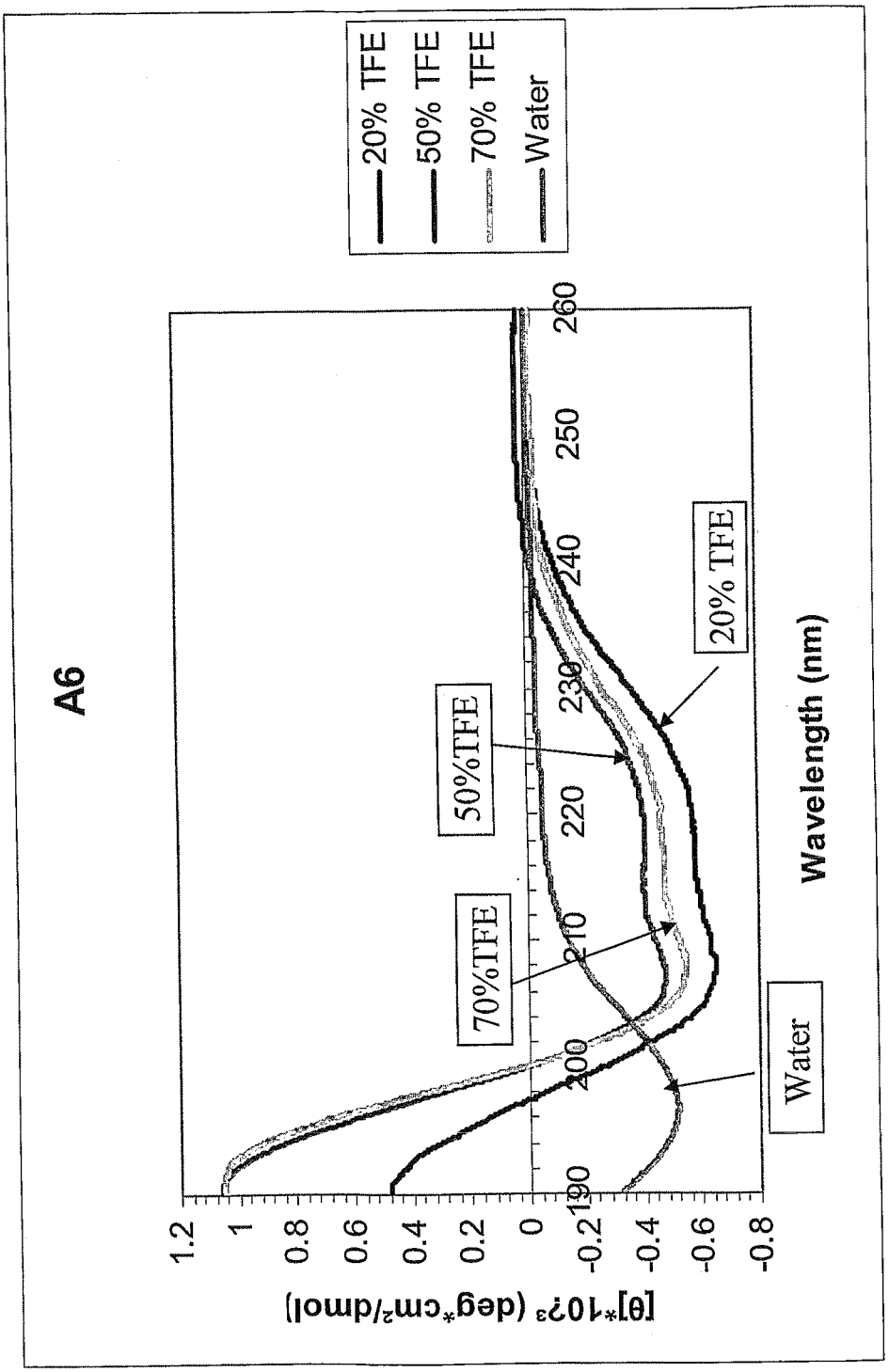

A6
20% TFE
50% TFE
70% TFE
Water
50%TFE
70%TFE
20% TFE
Water
Wavelength (nm)
[θ]*10?³ (deg*cm²/dmol)
1.2
1
0.8
0.6
0.4
0.2
0
-0.2
-0.4
-0.6
-0.8
190
200
210
220
230
240
250
260

Figure 3B

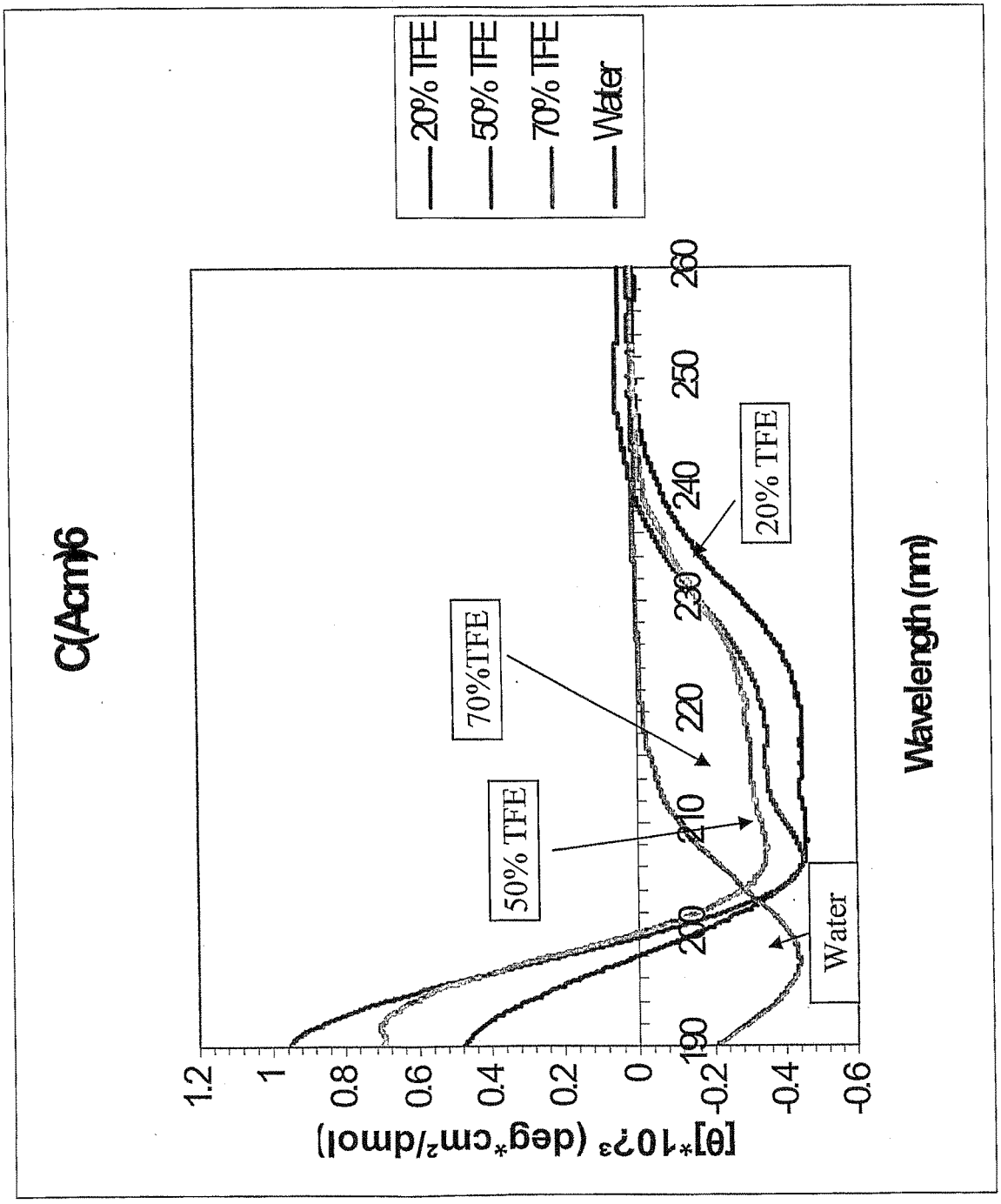

20% TFE
50% TFE
70% TFE
Water
C(Acm)6
[θ]*10?3 (deg*cm2/dmol)
Wavelength (nm)
1.2
1
0.8
0.6
0.4
0.2
0
-0.2
-0.4
-0.6
190
200
210
220
230
240
250
260
70%TFE
50% TFE
20% TFE
Water

Figure 3C

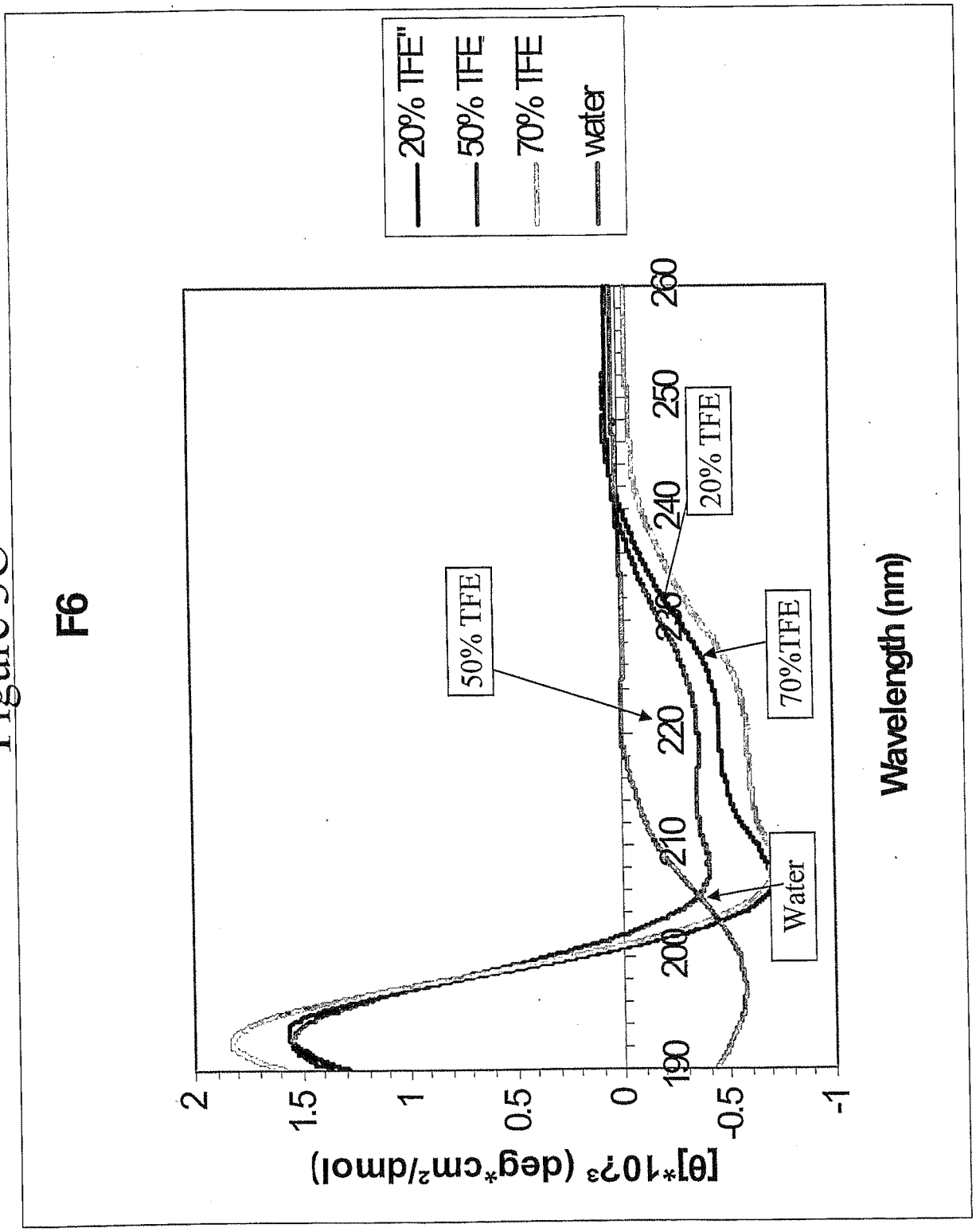

F6
20% TFE"
50% TFE
70% TFE
water
20% TFE
50% TFE
70%TFE
Water
[θ]*10?³ (deg*cm²/dmol)
Wavelength (nm)
2
1.5
1
0.5
0
-0.5
-1
190
200
210
220
230
240
250
260

Figure 3D

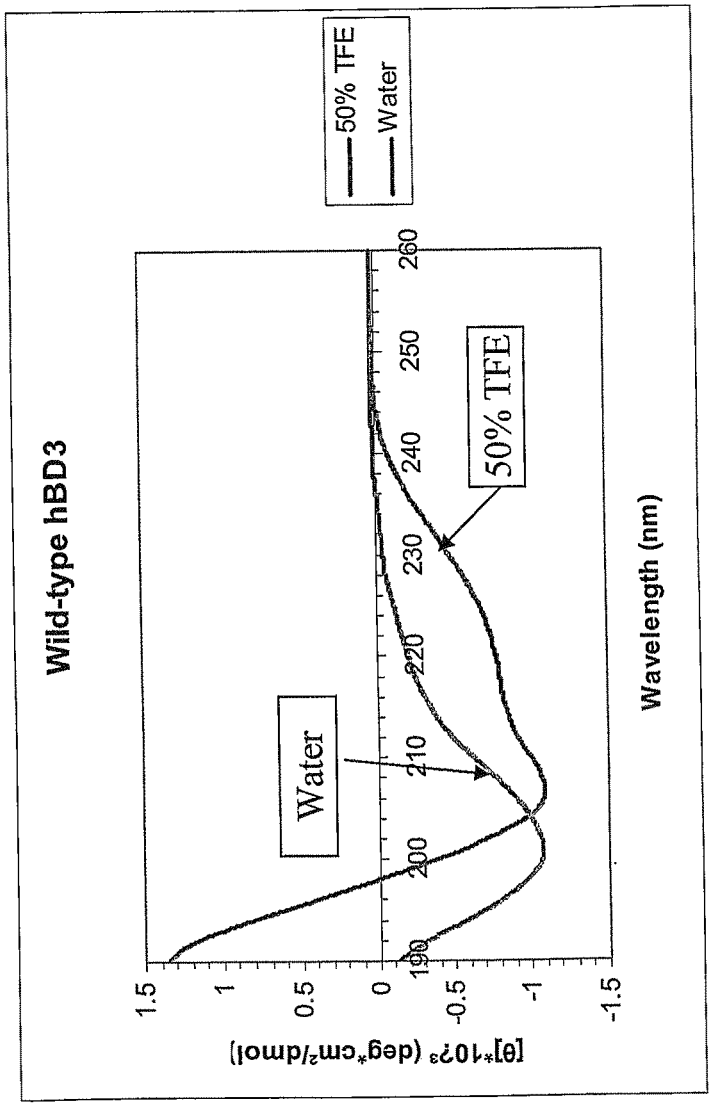

Wild-type hBD3
50% TFE
Water
Water
50% TFE
[θ]*10?³ (deg*cm²/dmol)
Wavelength (nm)
1.5
1
0.5
0
-0.5
-1
-1.5
190
200
210
220
230
240
250
260

Figure 3E

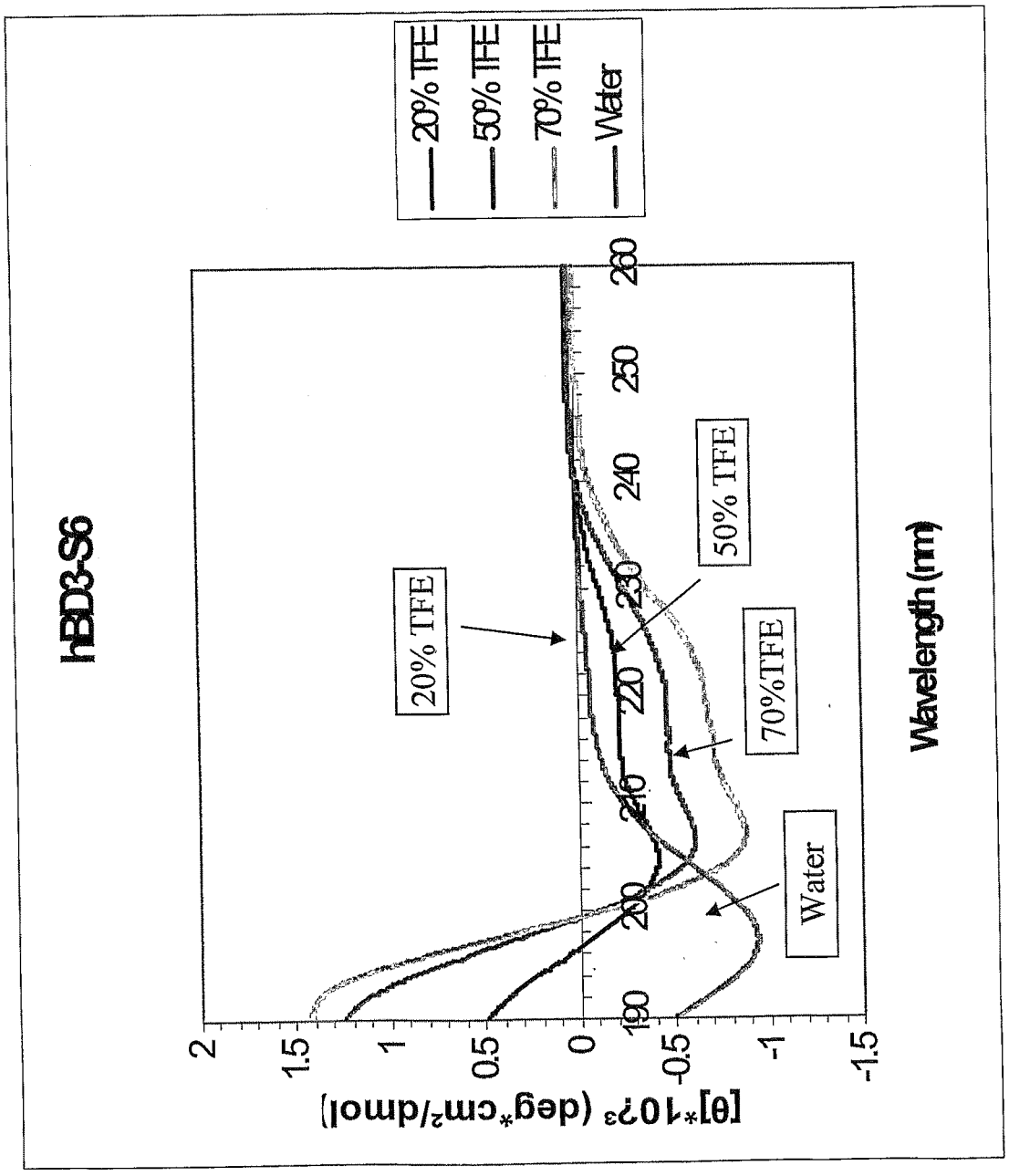

hBD3-S6
20% TFE
50% TFE
70% TFE
Water
[θ]*10?³ (deg*cm²/dmol)
Wavelength (nm)
2
1.5
1
0.5
0
-0.5
-1
-1.5
190
200
210
220
230
240
250
260

Figure 3F

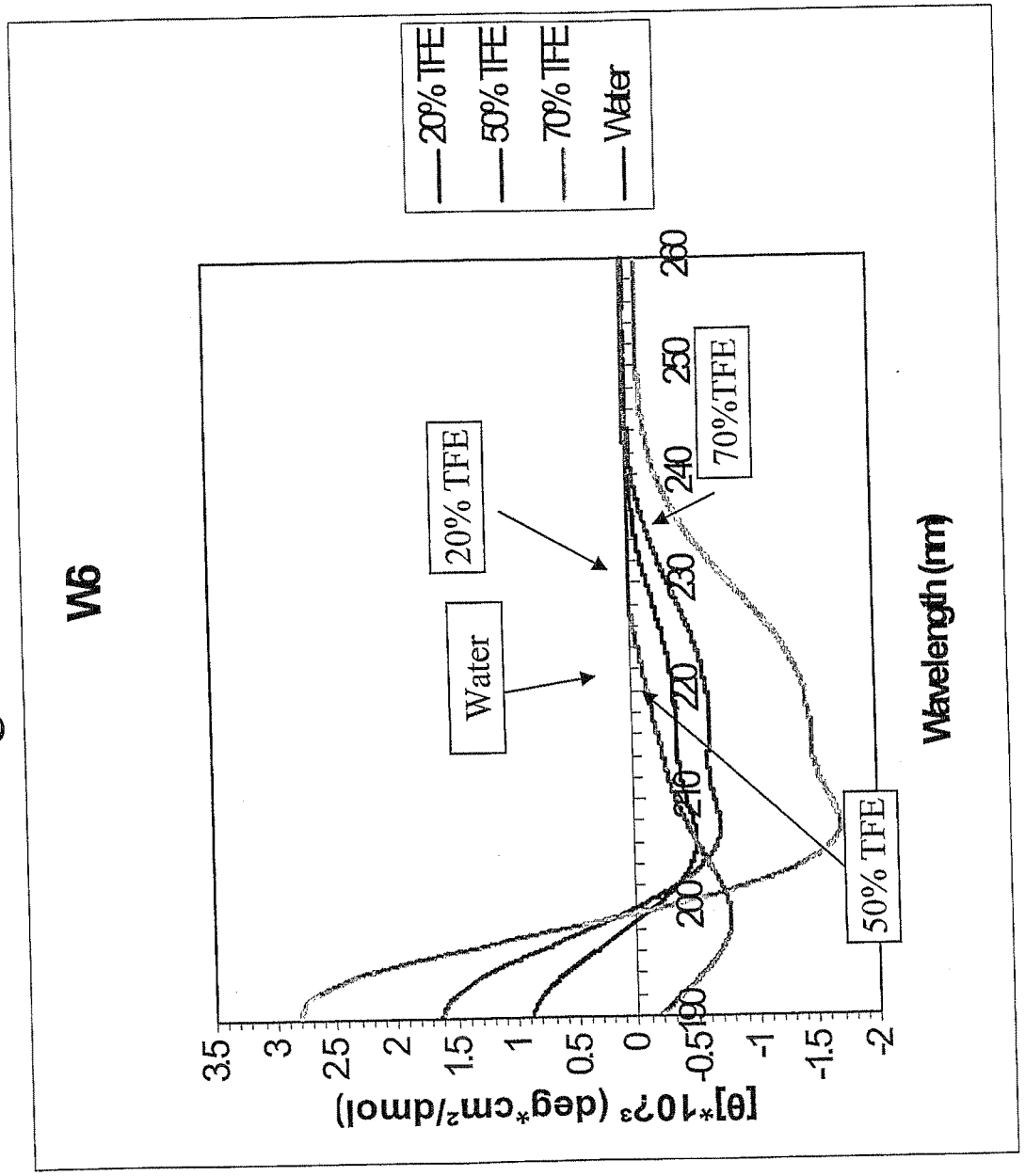

W6
20% TFE
50% TFE
70% TFE
Water
20% TFE
Water
70%TFE
50% TFE
[θ]*10?³ (deg*cm²/dmol)
Wavelength (nm)
3.5
3
2.5
2
1.5
1
0.5
0
-0.5
-1
-1.5
-2
260
250
240
230
220
210
200
190

Figure 3G

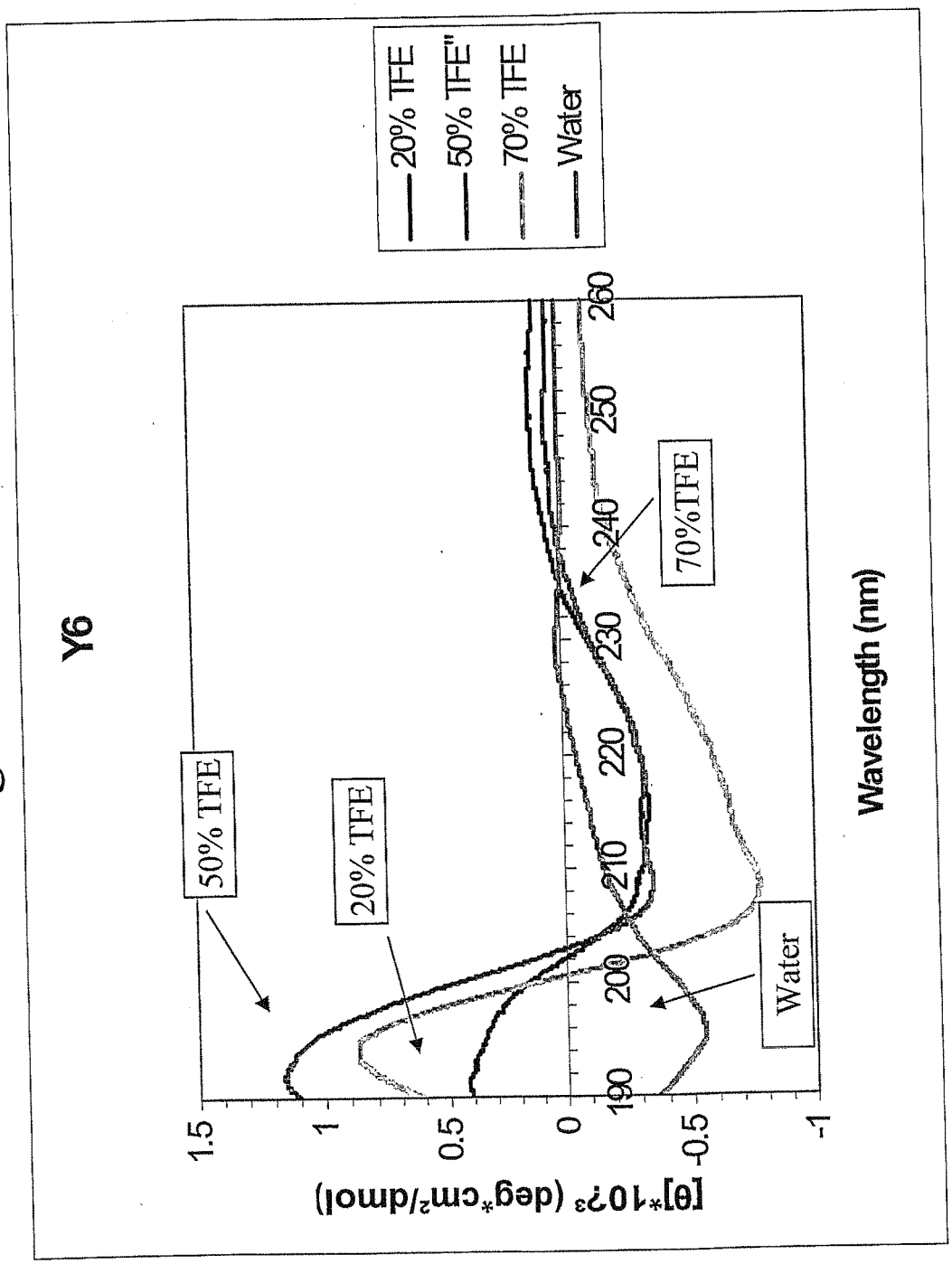

Y6
20% TFE
50% TFE"
70% TFE
Water
50% TFE
20% TFE
70%TFE
Water
[θ]*10?³ (deg*cm²/dmol)
Wavelength (nm)
1.5
1
0.5
0
-0.5
-1
190
200
210
220
230
240
250
260

Figure 4A

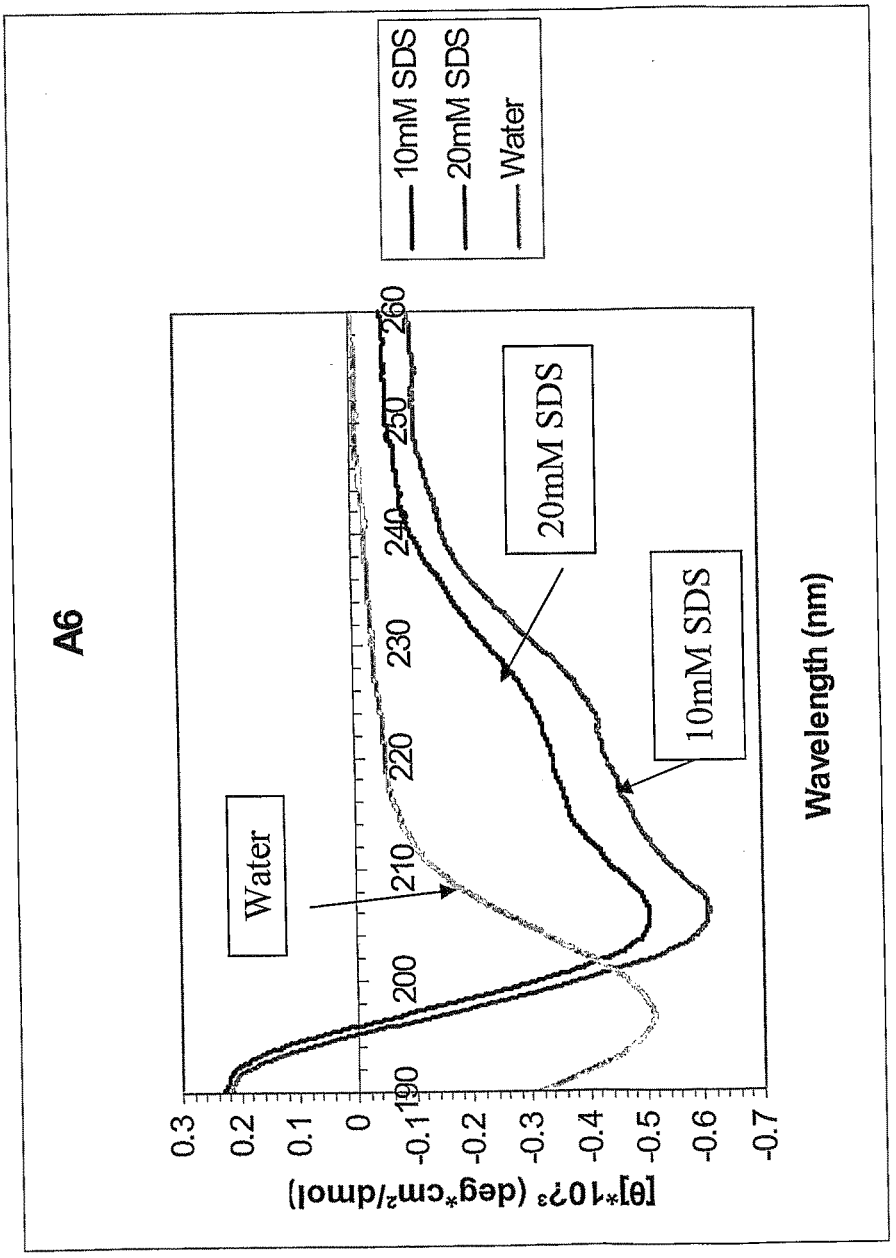

A6
10mM SDS
20mM SDS
Water
Water
20mM SDS
10mM SDS
[θ]*10?³ (deg*cm²/dmol)
Wavelength (nm)
0.3
0.2
0.1
0
-0.1
-0.2
-0.3
-0.4
-0.5
-0.6
-0.7
190
200
210
220
230
240
250
260

Figure 4B

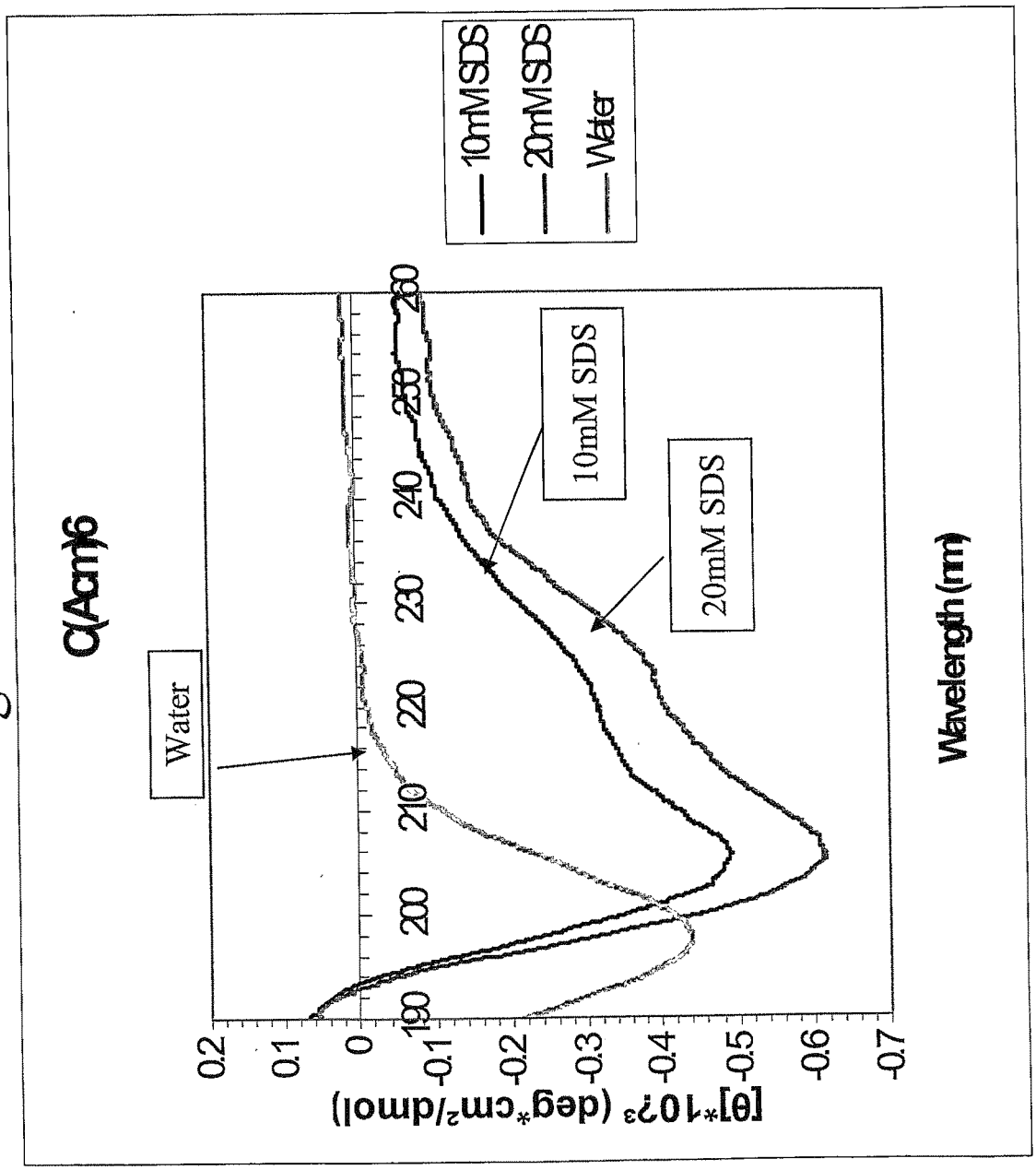

10mMSDS
20mMSDS
Water
Water
10mM SDS
20mM SDS
Wavelength (nm)
[θ]*10?³ (deg*cm²/dmol)
0.2
0.1
0
-0.1
-0.2
-0.3
-0.4
-0.5
-0.6
-0.7
190
200
210
220
230
240
250
260

Figure 4C

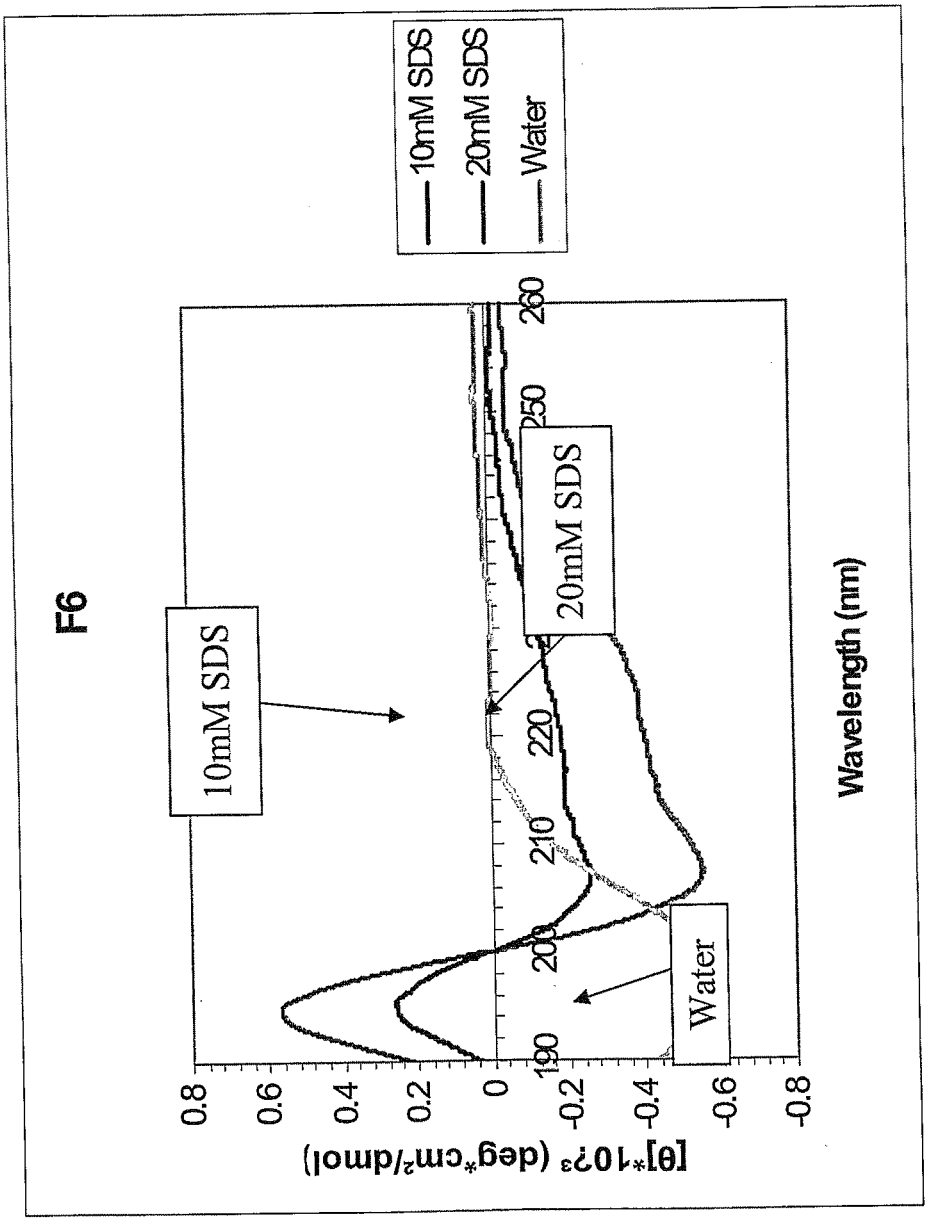

F6
10mM SDS
20mM SDS
Water
[θ]*10?³ (deg*cm²/dmol)
Wavelength (nm)
0.8
0.6
0.4
0.2
0
-0.2
-0.4
-0.6
-0.8
190
200
210
220
250
260

Figure 4D

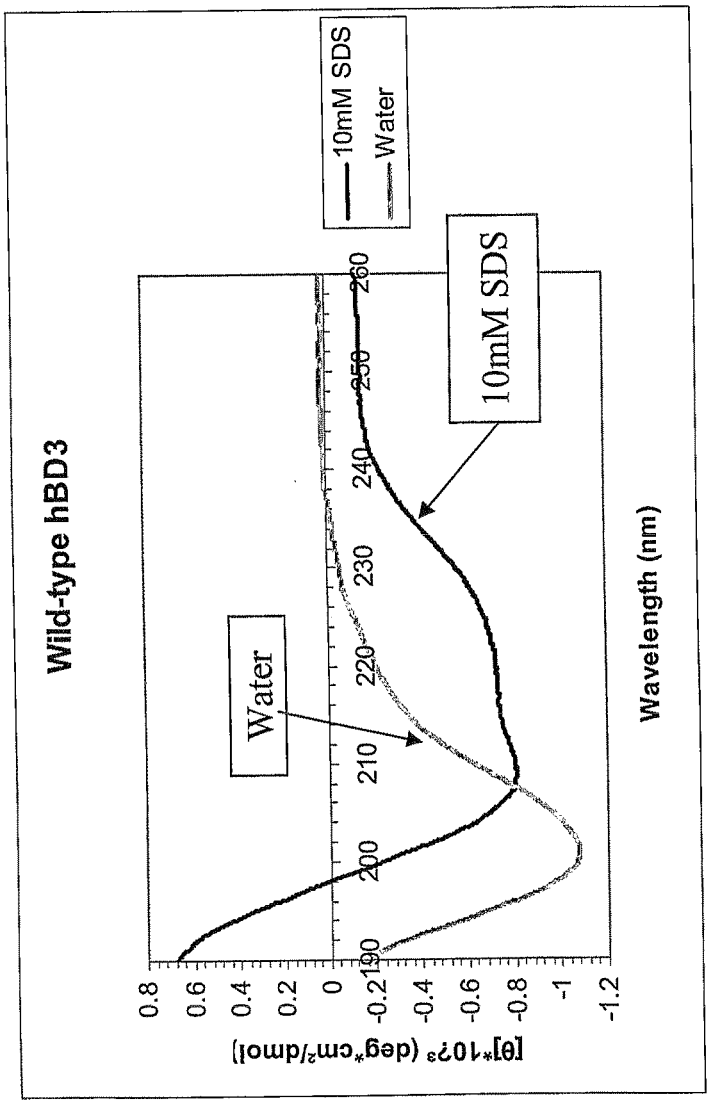

Wild-type hBD3
10mM SDS
Water
10mM SDS
Water
Wavelength (nm)
[θ]*10?³ (deg*cm²/dmol)
0.8
0.6
0.4
0.2
0
-0.2
-0.4
-0.6
-0.8
-1
-1.2
190
200
210
220
230
240
250
260

Figure 4E

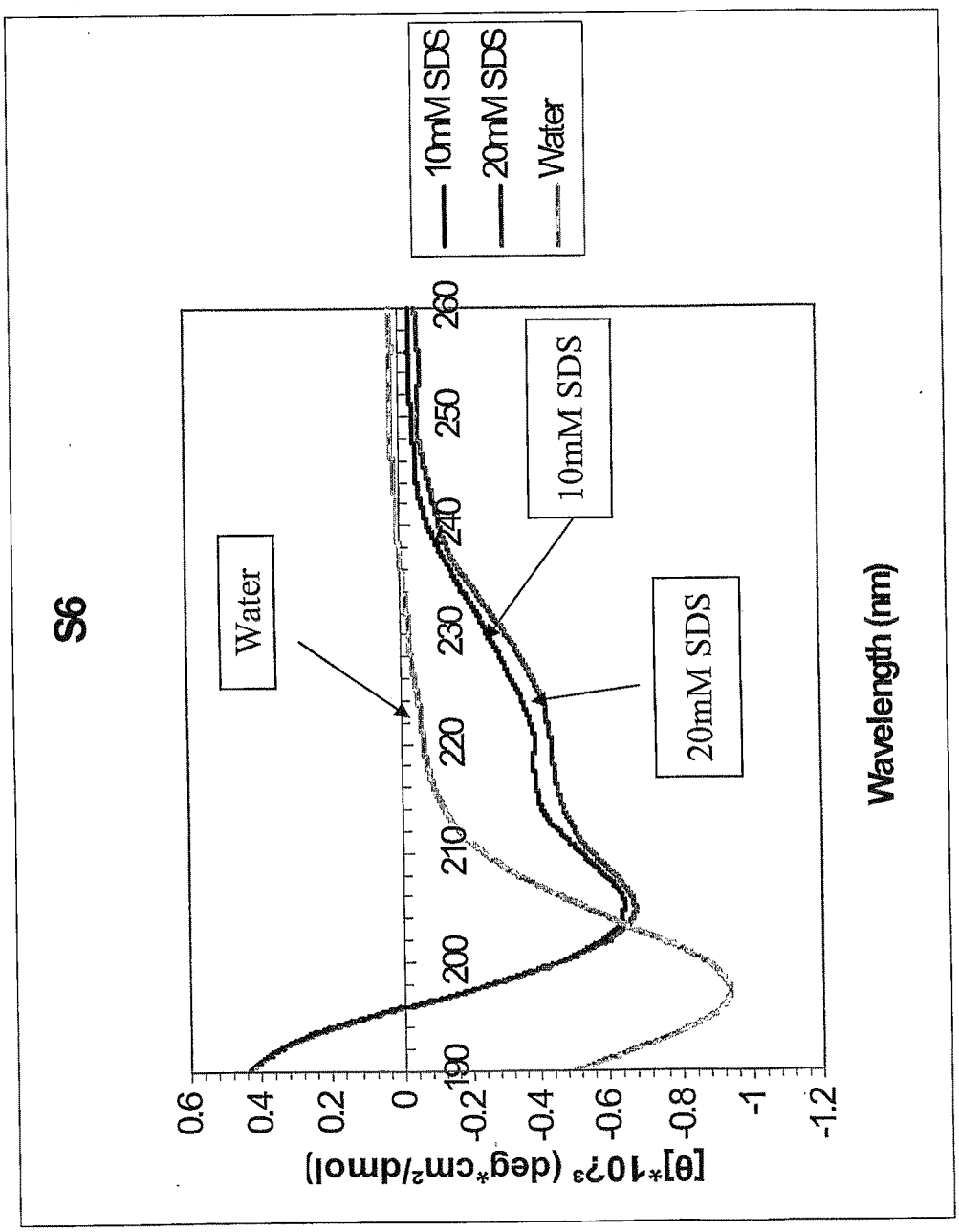

S6
10mM SDS
20mM SDS
Water
Water
10mM SDS
20mM SDS
[θ]*10?³ (deg*cm²/dmol)
Wavelength (nm)
0.6
0.4
0.2
0
-0.2
-0.4
-0.6
-0.8
-1
-1.2
190
200
210
220
230
240
250
260

Figure 4F

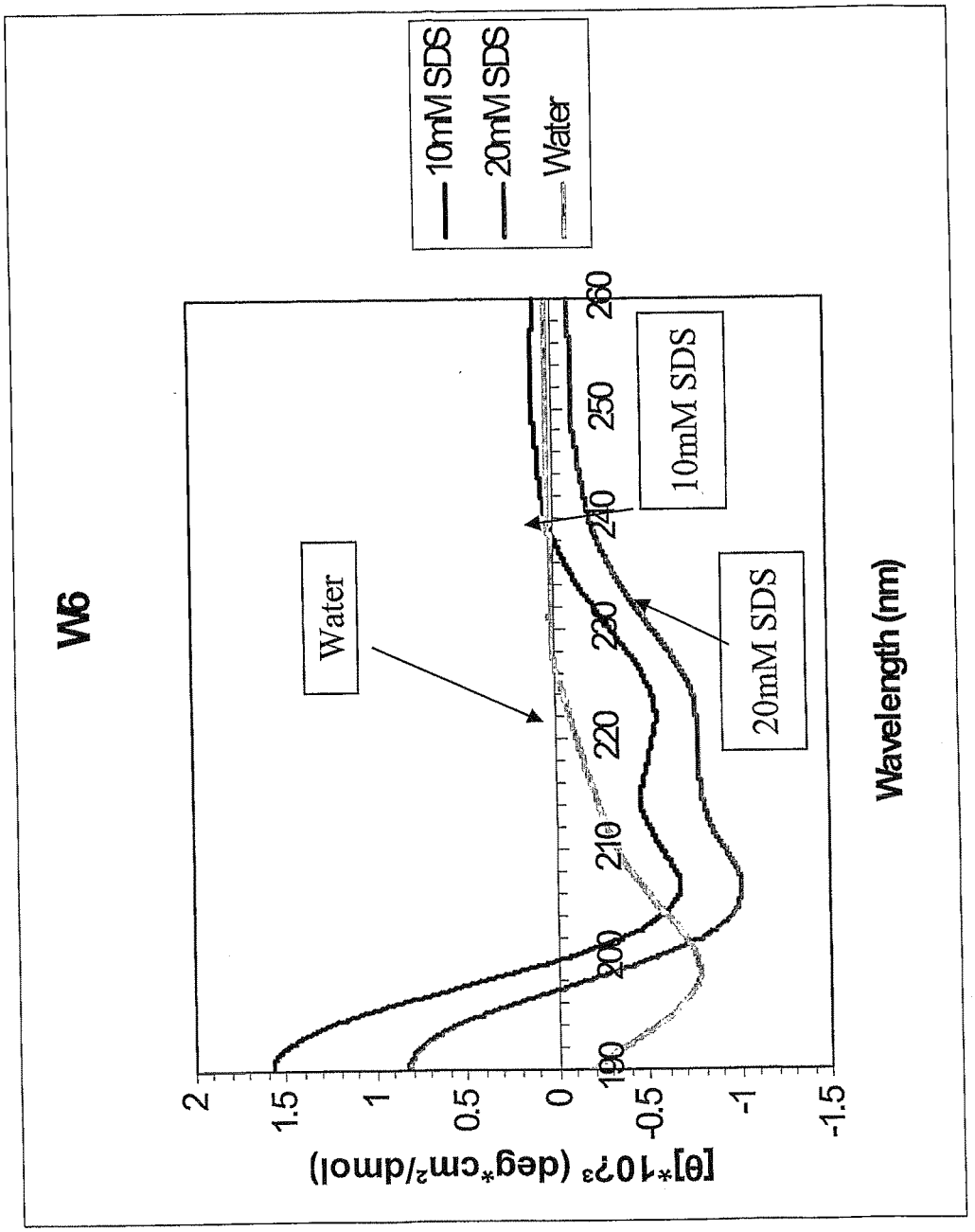

W6
10mM SDS
20mM SDS
Water
Water
10mM SDS
20mM SDS
[θ]*10?³ (deg*cm²/dmol)
Wavelength (nm)
2
1.5
1
0.5
0
-0.5
-1
-1.5
190
200
210
220
230
240
250
260

Figure 4G

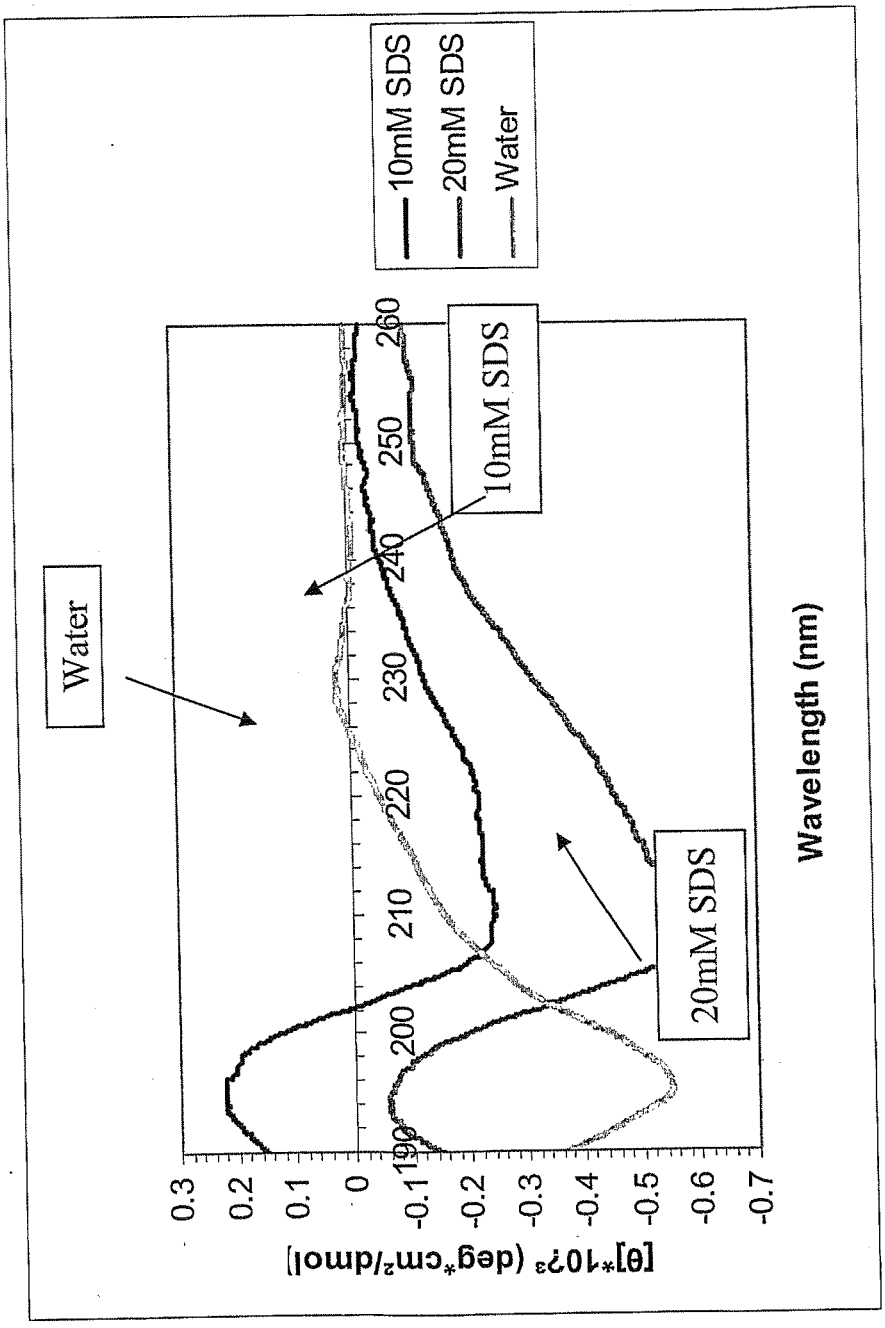

10mM SDS
20mM SDS
Water
Water
10mM SDS
20mM SDS
Wavelength (nm)
[θ]*10?³ (deg*cm²/dmol)
0.3
0.2
0.1
0
-0.1
-0.2
-0.3
-0.4
-0.5
-0.6
-0.7
190
200
210
220
230
240
250
260

Figure 5

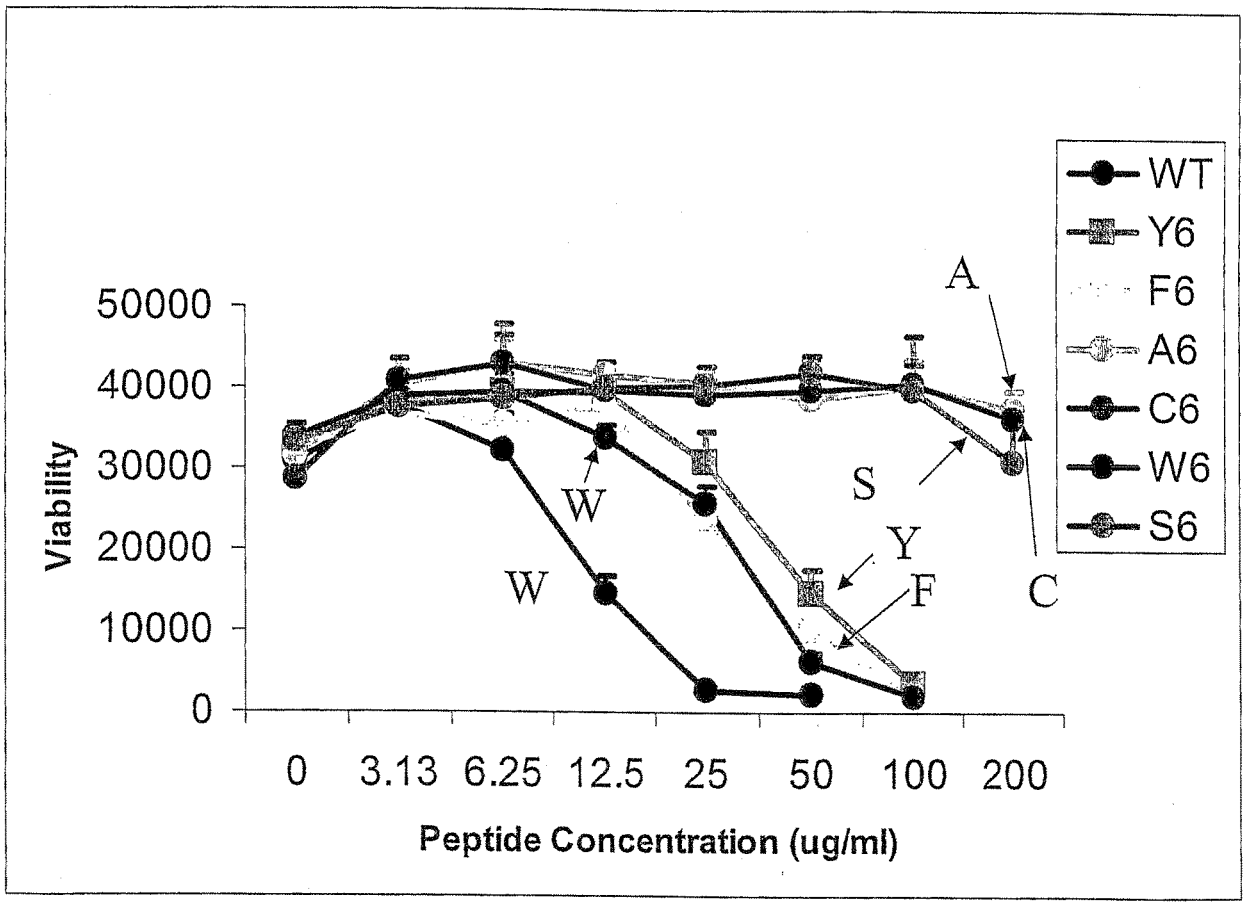

WT
Y6
F6
A6
C6
W6
S6
Viability
50000
40000
30000
20000
10000
0
0
3.13
6.25
12.5
25
50
100
200
Peptide Concentration (ug/ml)
A
W
W
S
Y
F
C

Figure 6

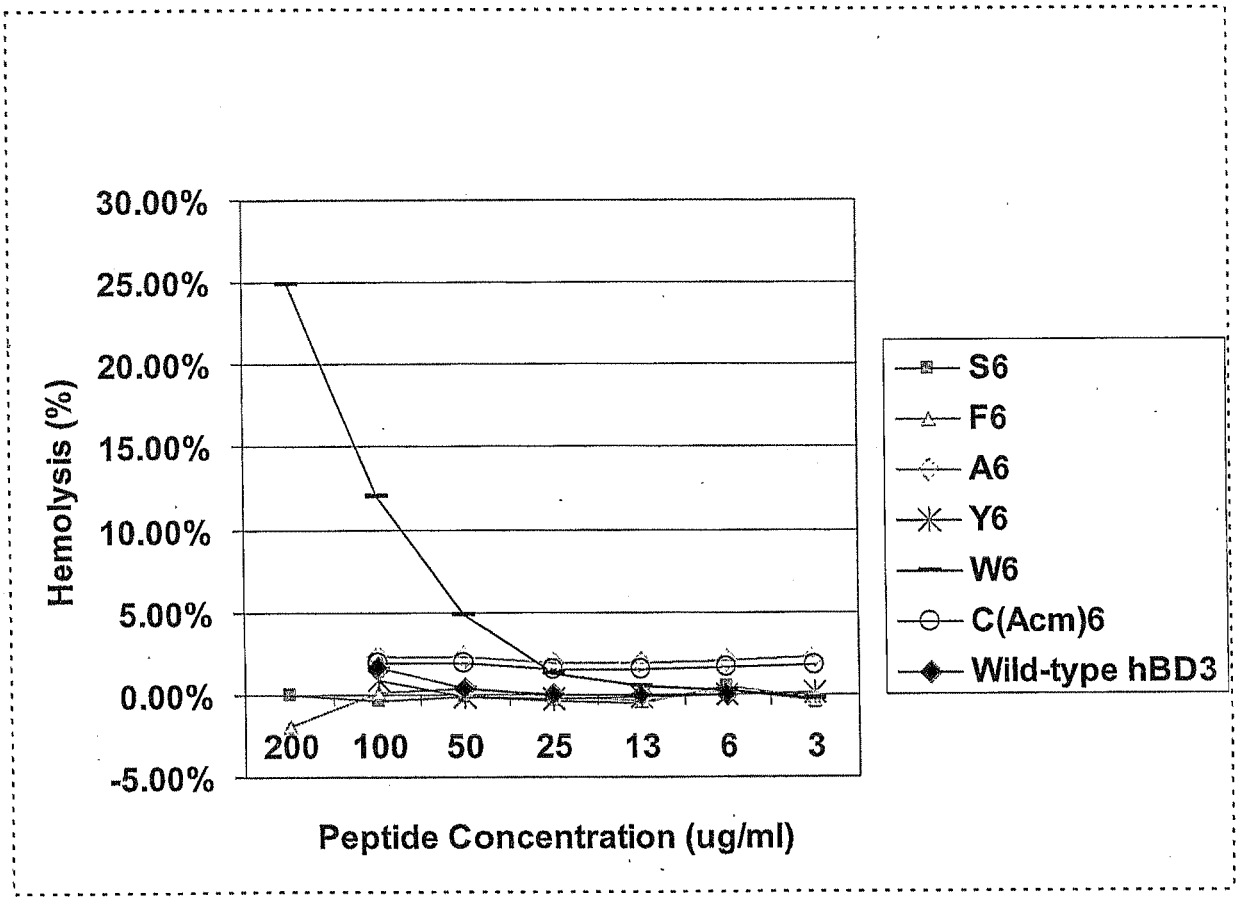

30.00%
25.00%
20.00%
15.00%
10.00%
5.00%
0.00%
-5.00%
Hemolysis (%)
200
100
50
25
13
6
3
Peptide Concentration (ug/ml)
S6
F6
A6
Y6
W6
C(Acm)6
Wild-type hBD3

Figure 7A

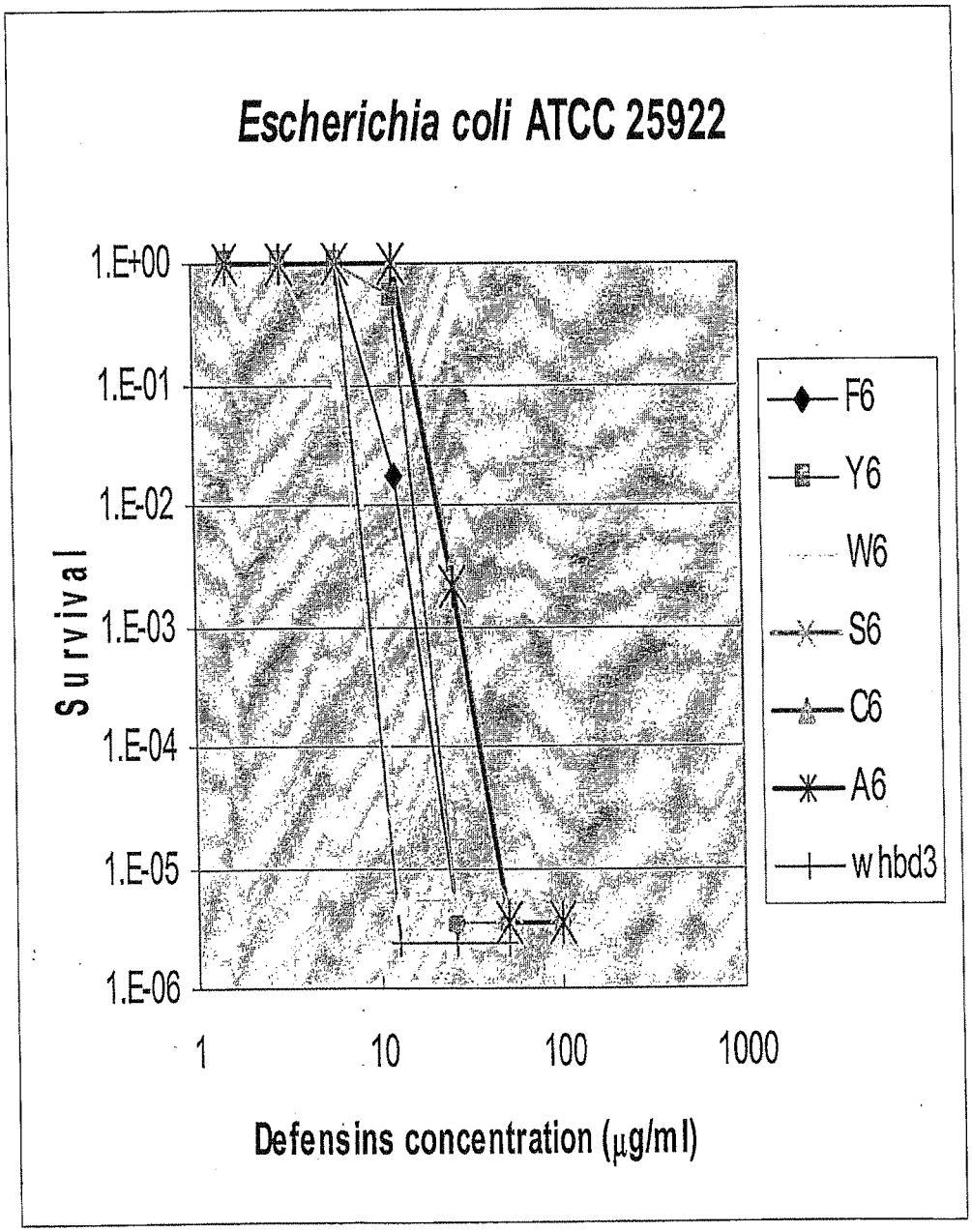

Escherichia coli ATCC 25922
Survival
1.E+00
1.E-01
1.E-02
1.E-03
1.E-04
1.E-05
1.E-06
1
10
100
1000
Defensins concentration (μg/ml)
F6
Y6
W6
S6
C6
A6
w hbd3

Figure 7B

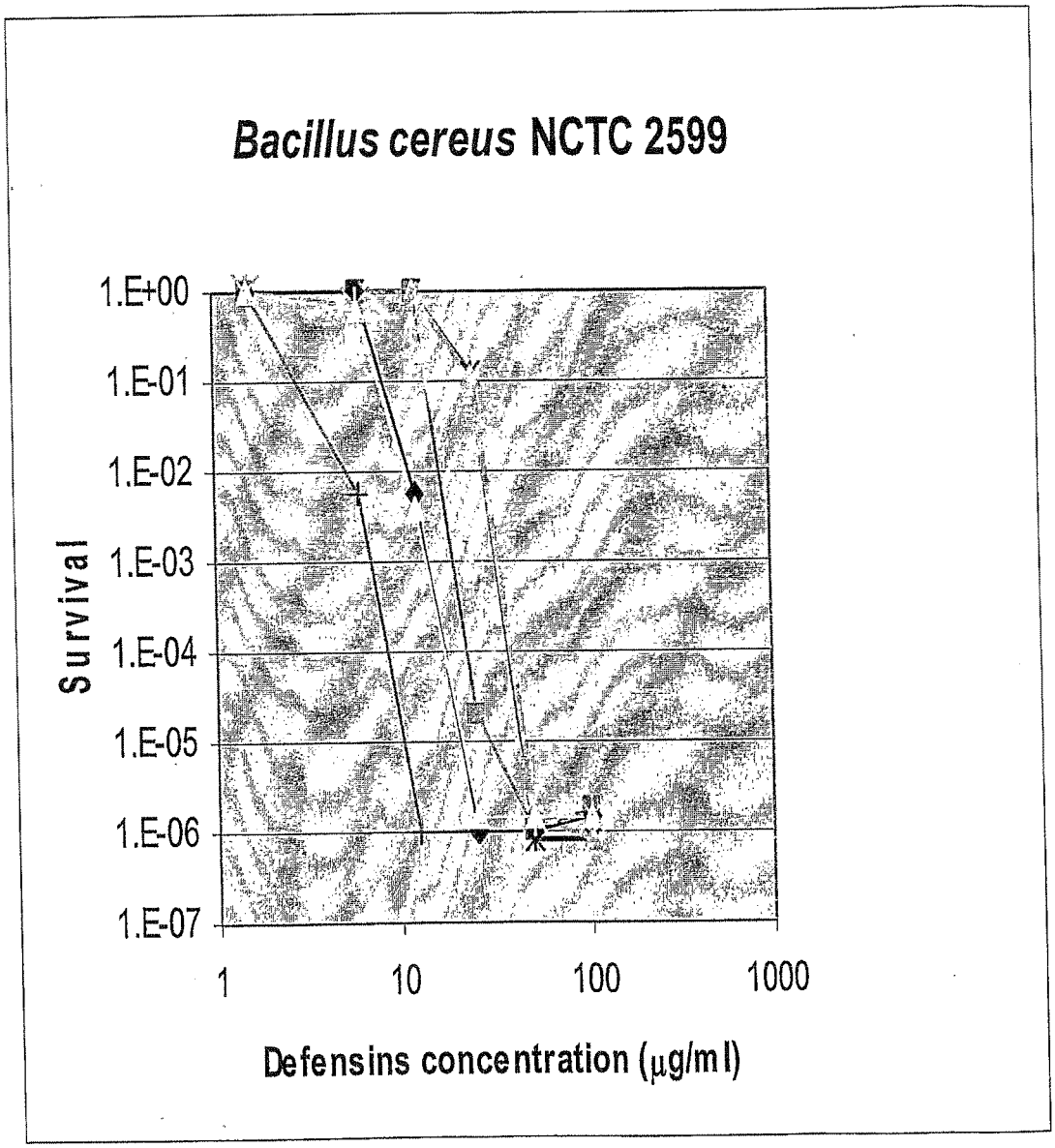

Bacillus cereus NCTC 2599
Survival
1.E+00
1.E-01
1.E-02
1.E-03
1.E-04
1.E-05
1.E-06
1.E-07
1
10
100
1000
Defensins concentration (µg/ml)

Figure 7C

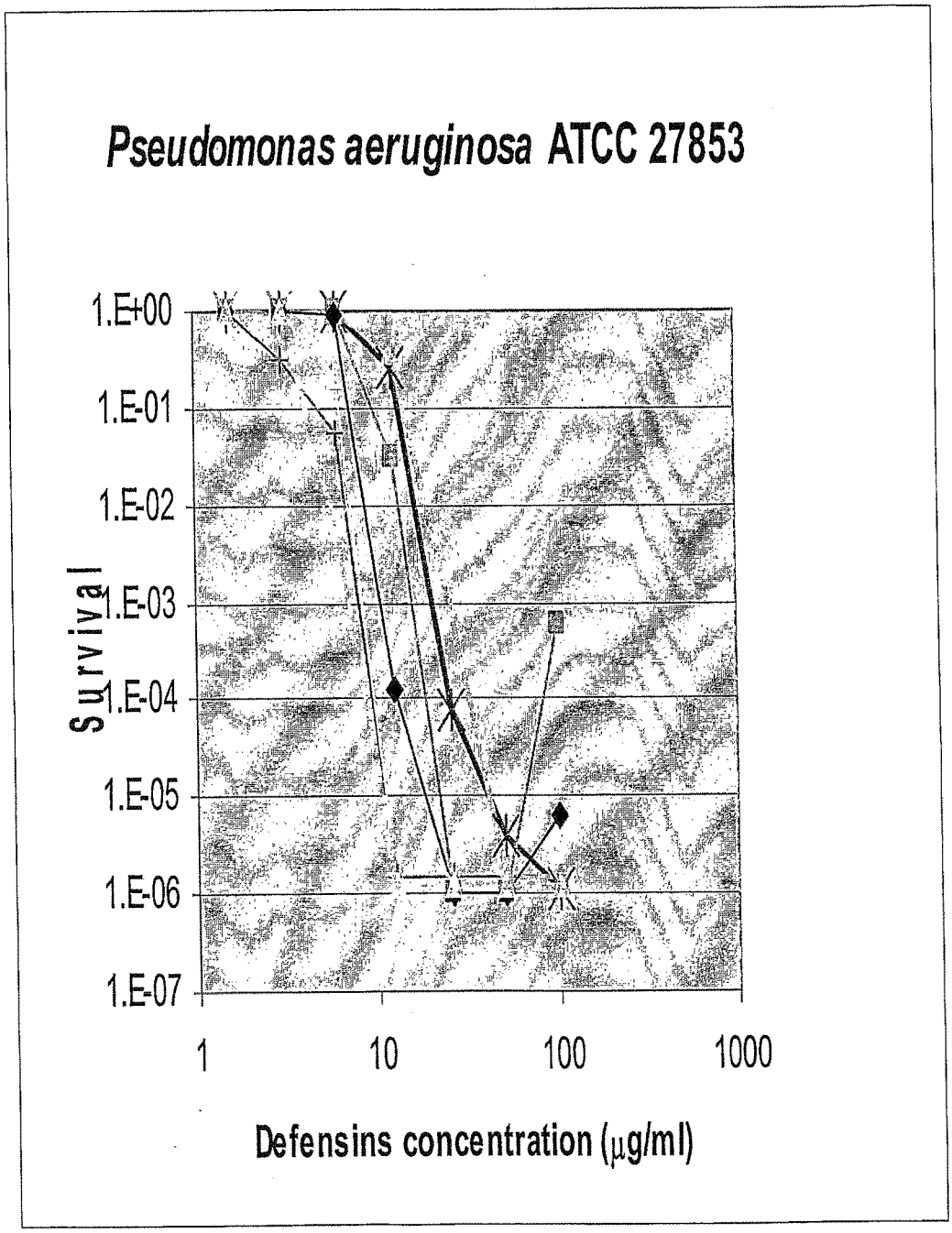

Pseudomonas aeruginosa ATCC 27853
Survival
1.E+00
1.E-01
1.E-02
1.E-03
1.E-04
1.E-05
1.E-06
1.E-07
1
10
100
1000
Defensins concentration (μg/ml)

Figure 7D

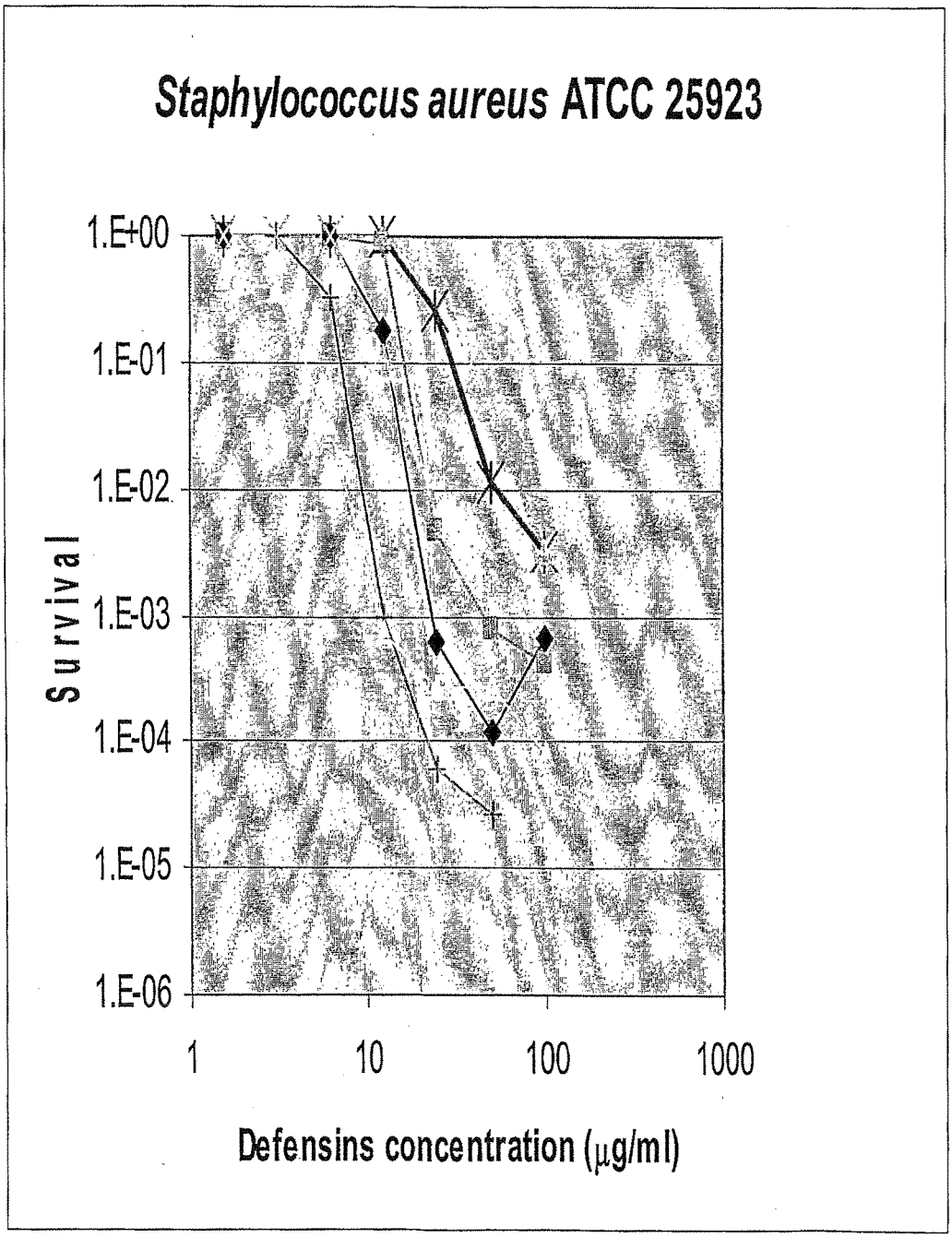
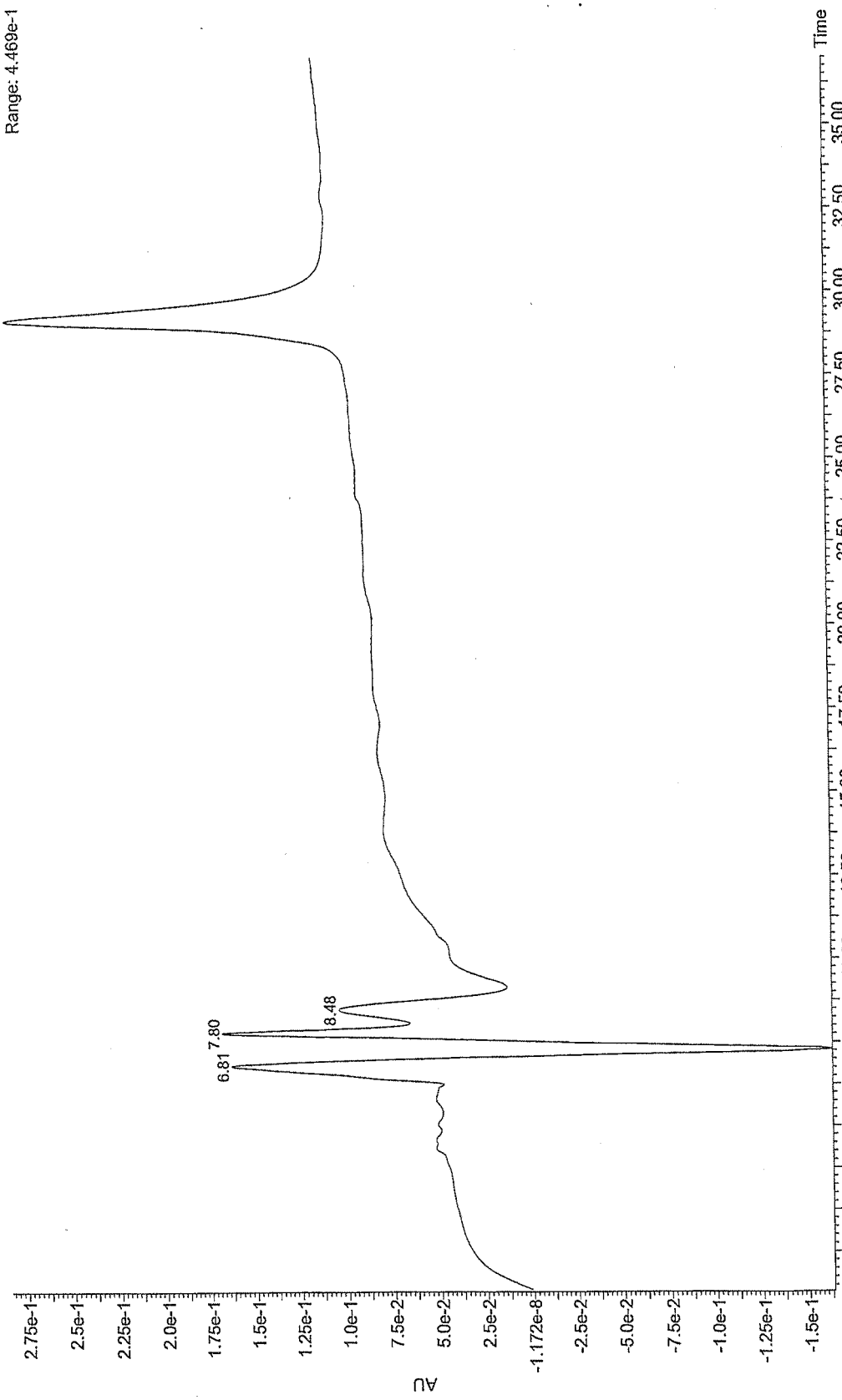

Staphylococcus aureus ATCC 25923
1.E+00
1.E-01
1.E-02
1.E-03
1.E-04
1.E-05
1.E-06
Survival
1
10
100
1000
Defensins concentration (μg/ml)

Figure 8A RP-HPLC-UV chromatogram and MS spectrum of W6



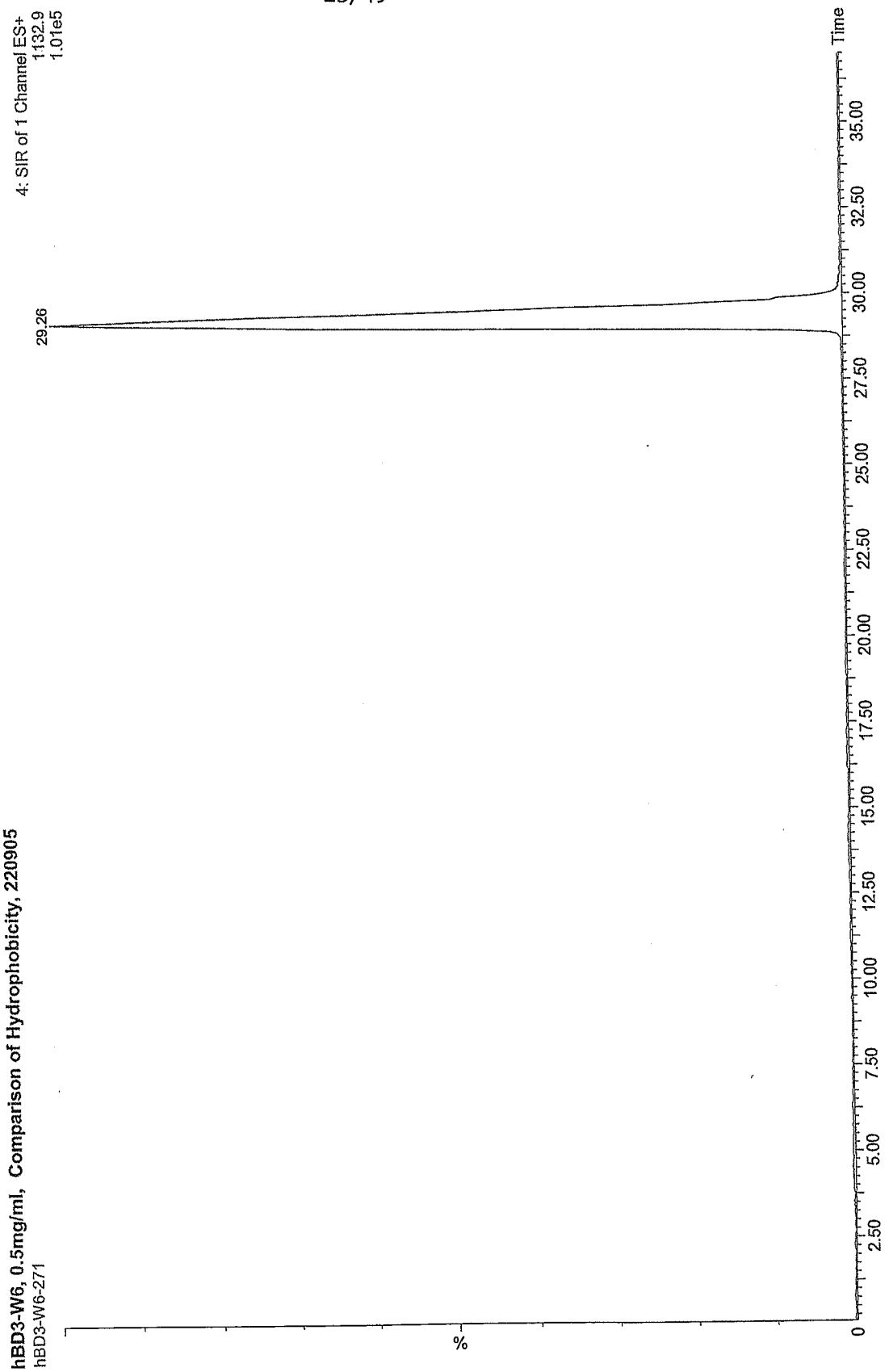


Figure 8B SIR chromatogram at m/z = 1132.9 ($[M+5H]^{5+}$)

Figure 8C ES+ MS SPECTRUM

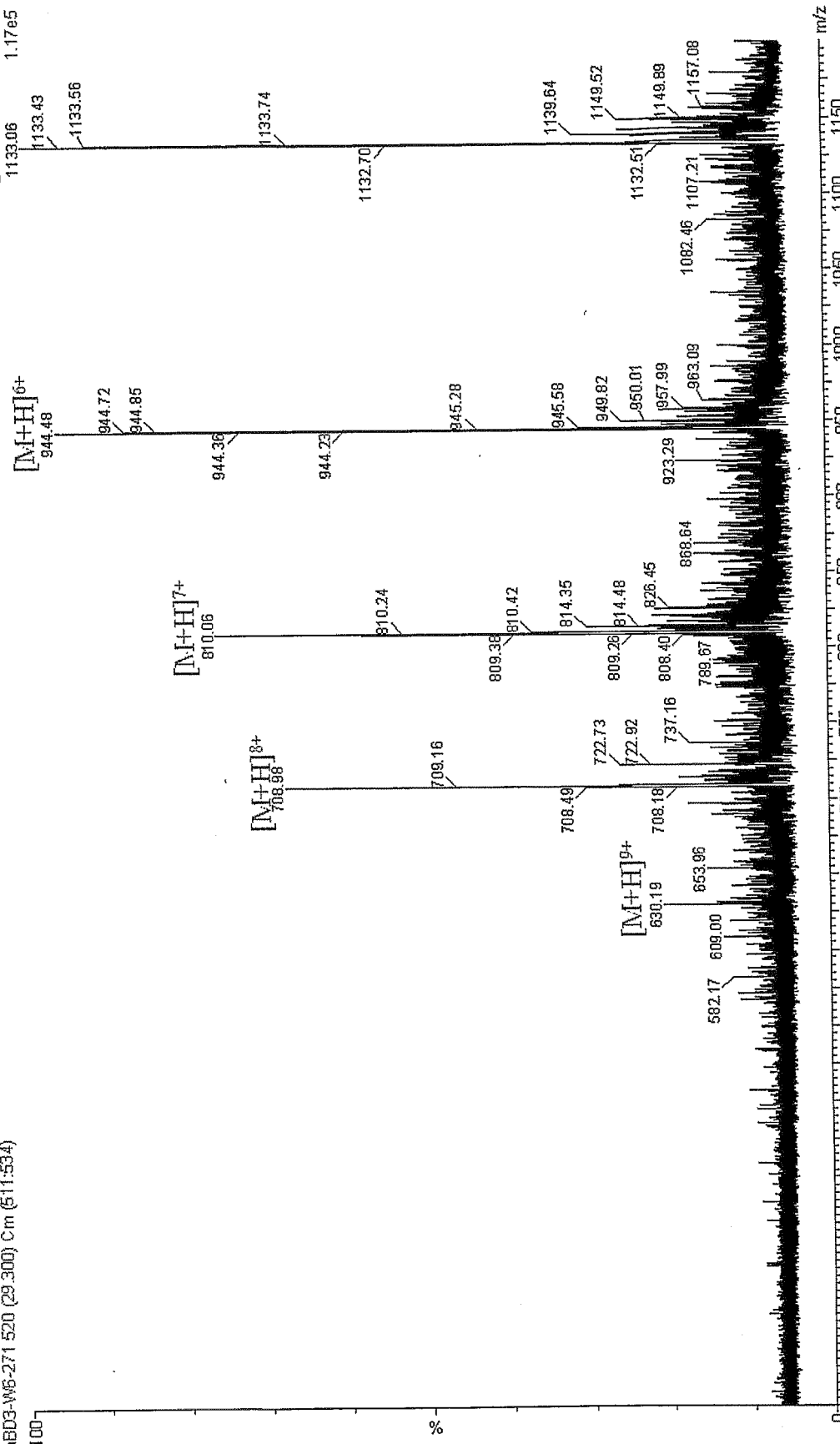


Figure 8D Deconvoluted MS spectrum

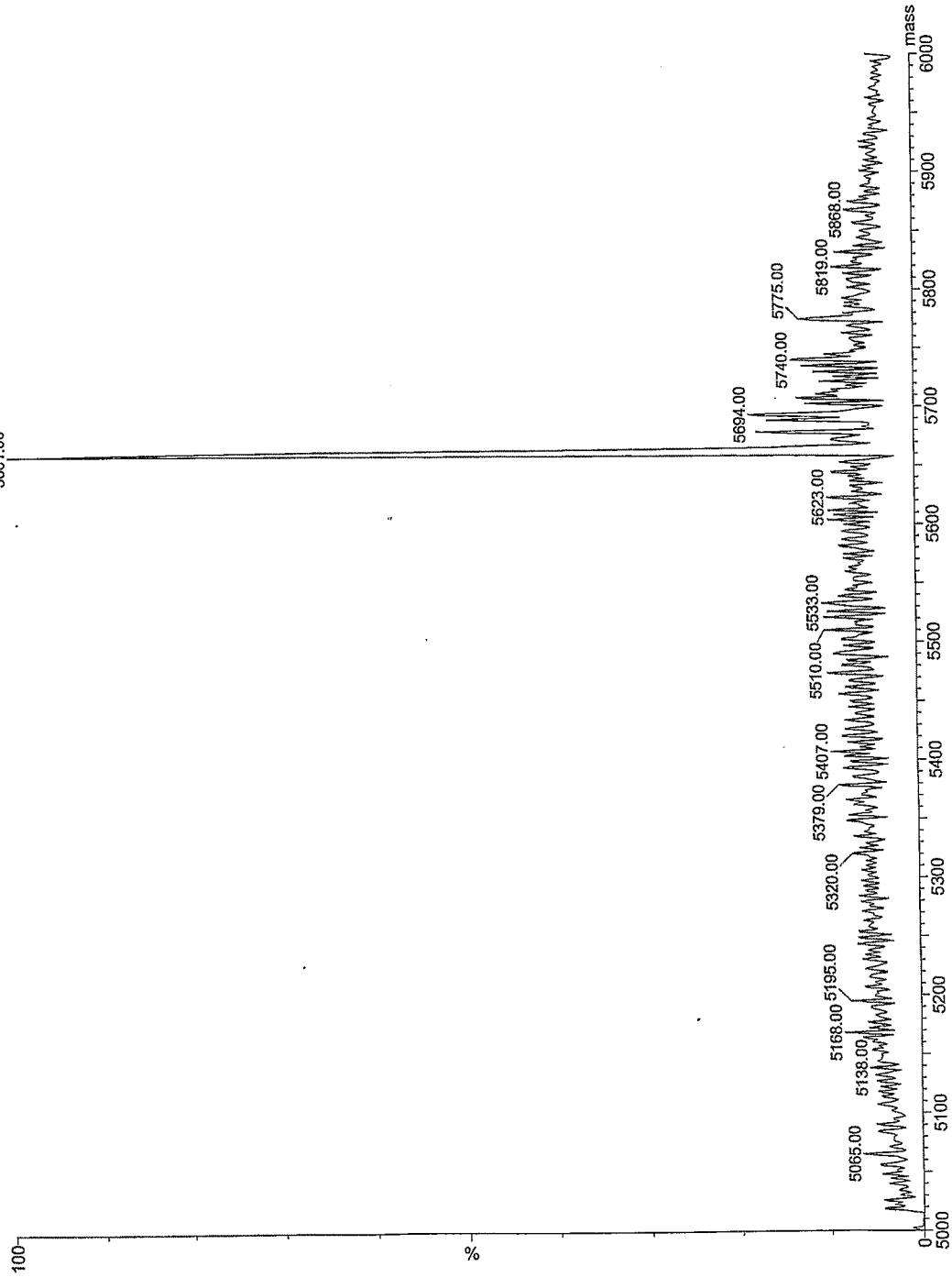
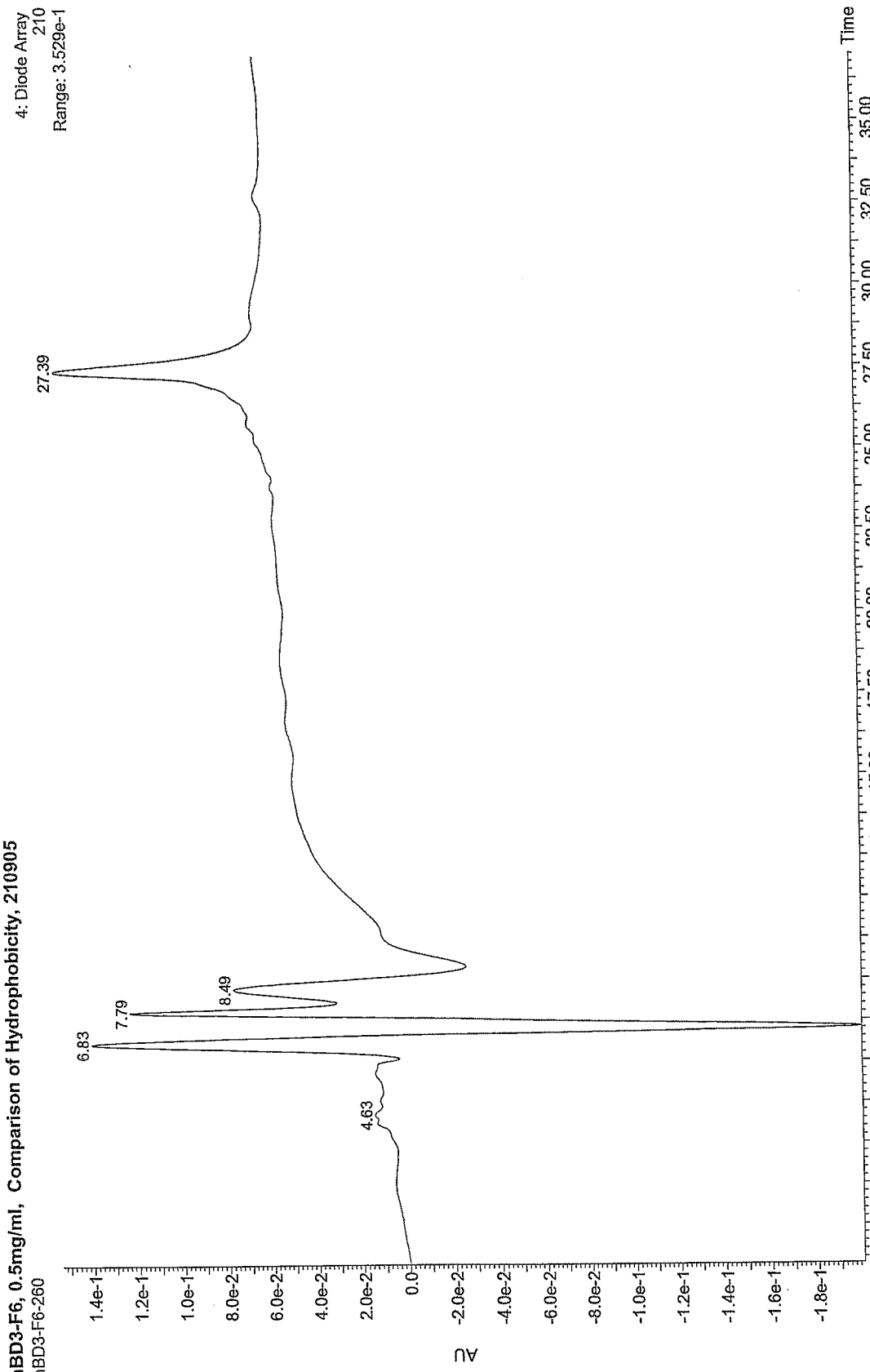


Figure 9A RP-HPLC-UV chromatogram and MS spectrum of F6



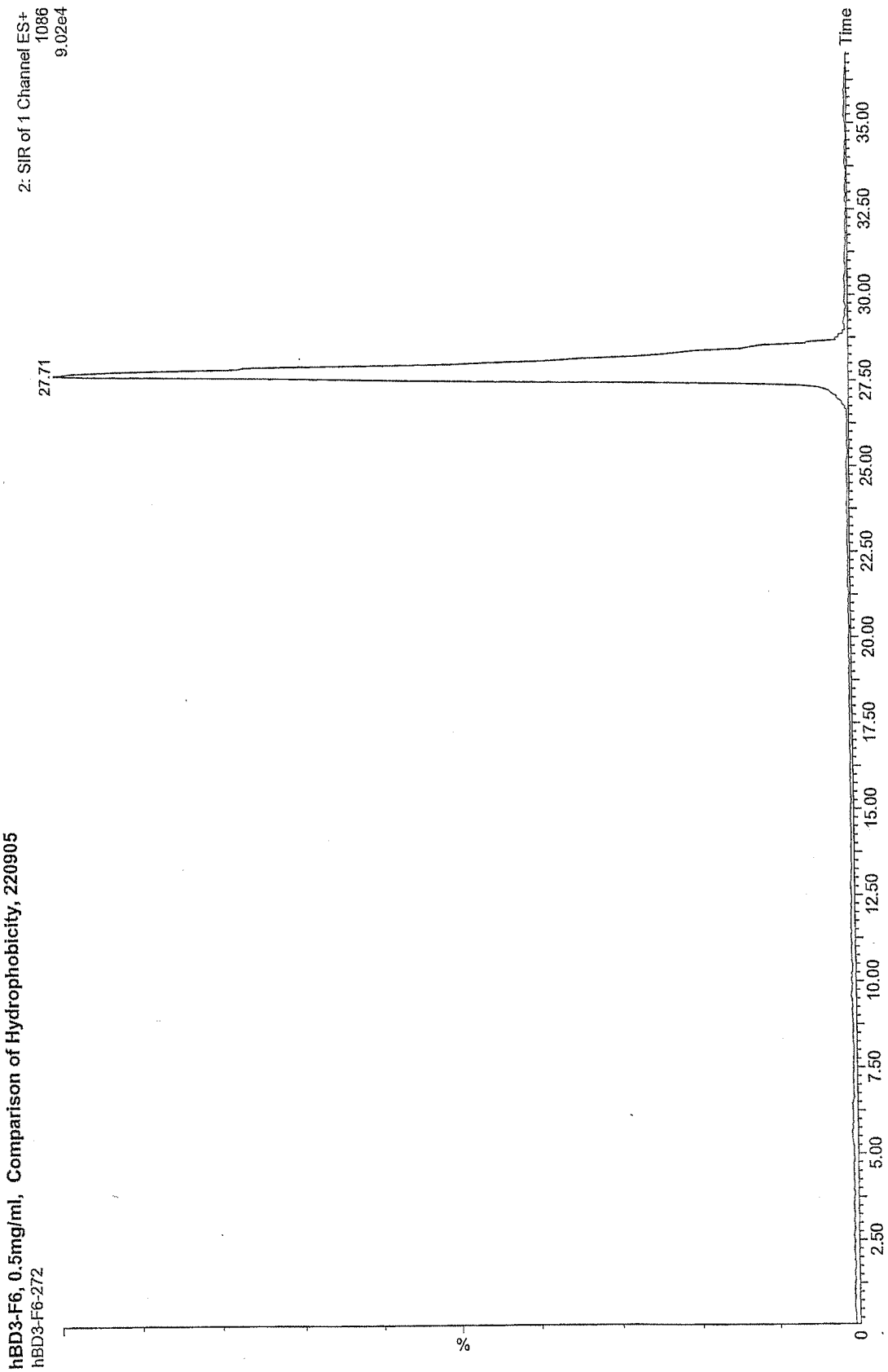


Figure 9B SIR chromatogram at m/z = 1086 ($[M+5H]^{5+}$)

Figure 9C ES+ MS Spectrum

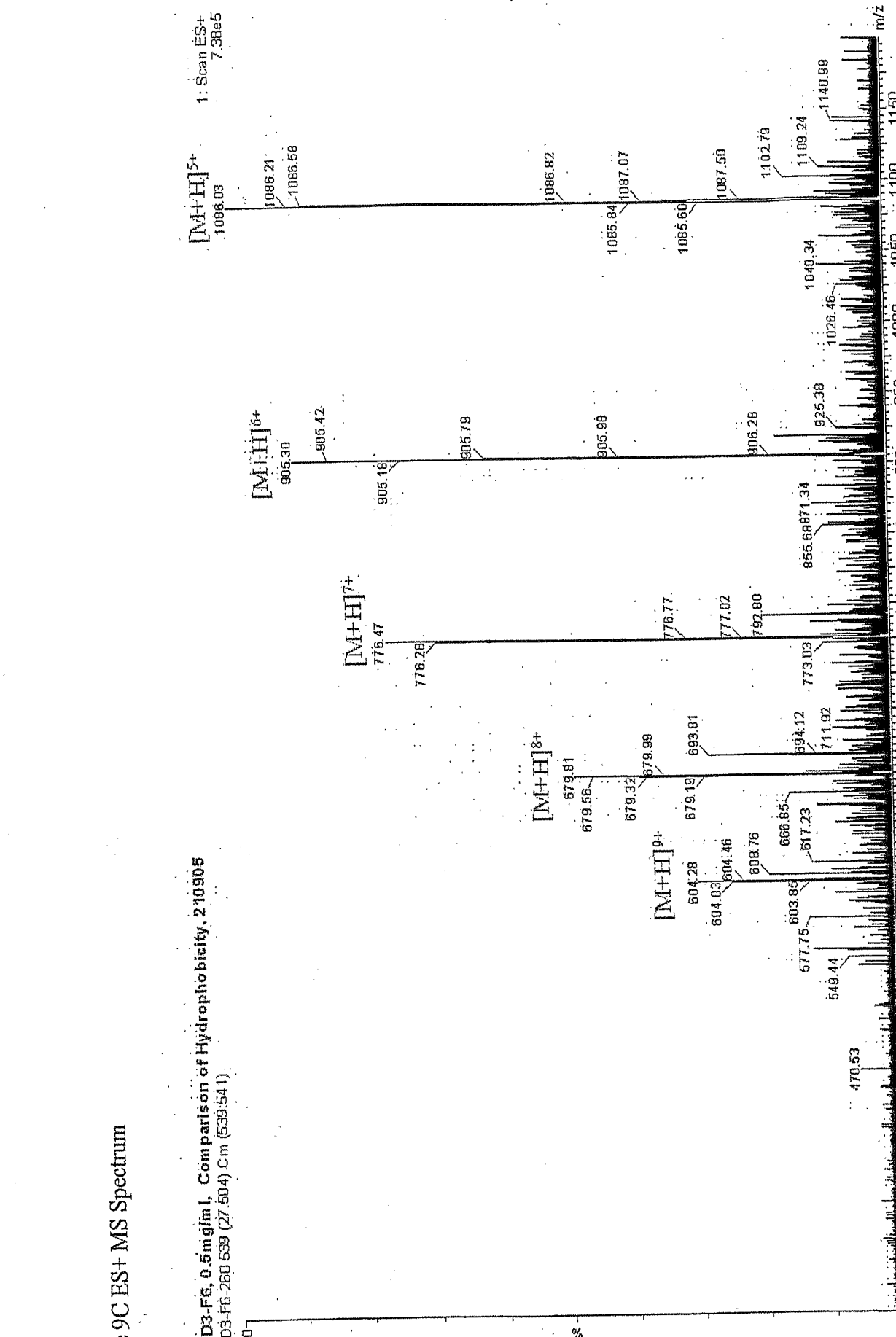


Figure 9D Deconvoluted MS spectrum

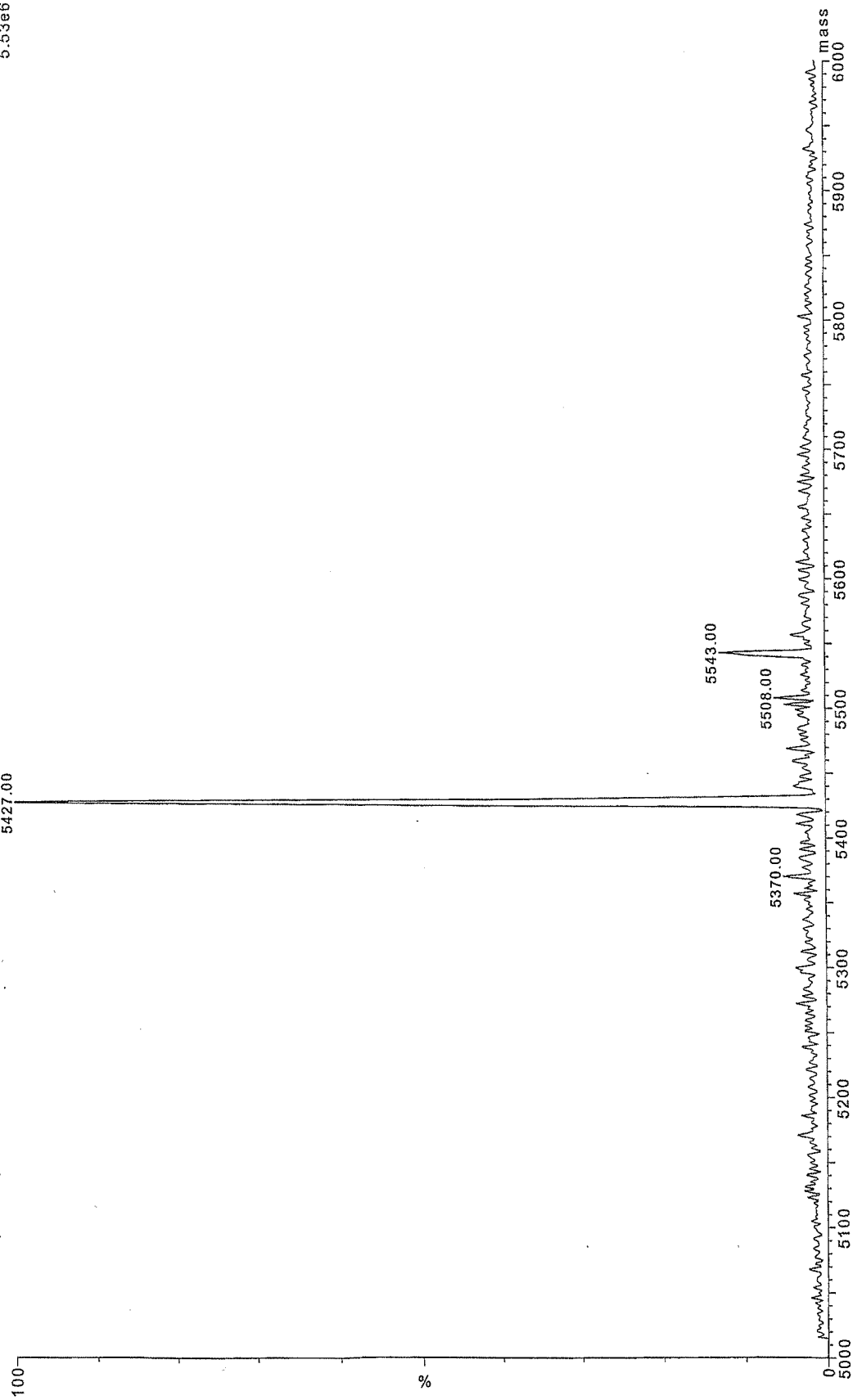
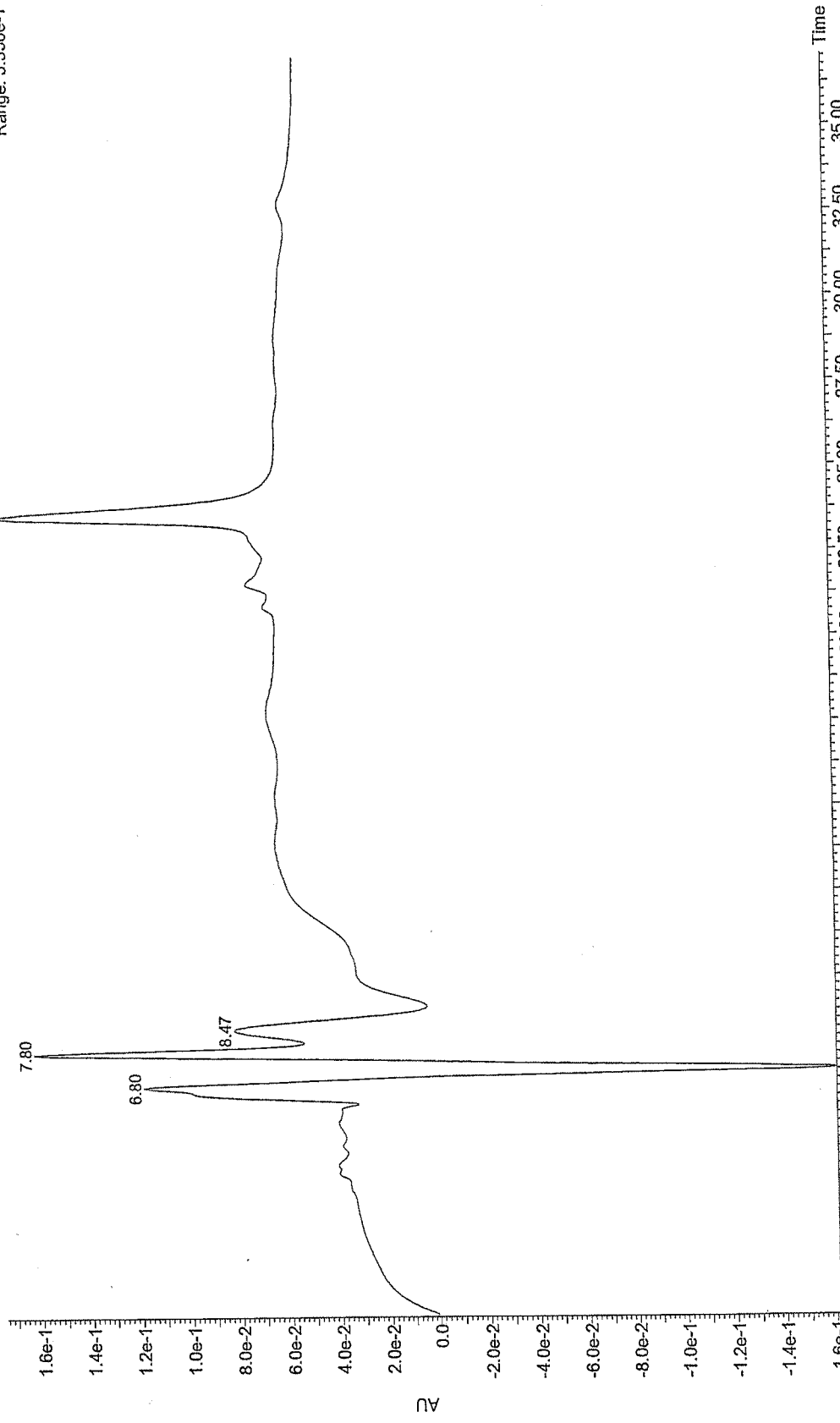


Figure 10A RP-HPLC-UV chromatogram and MS spectrum of Y6



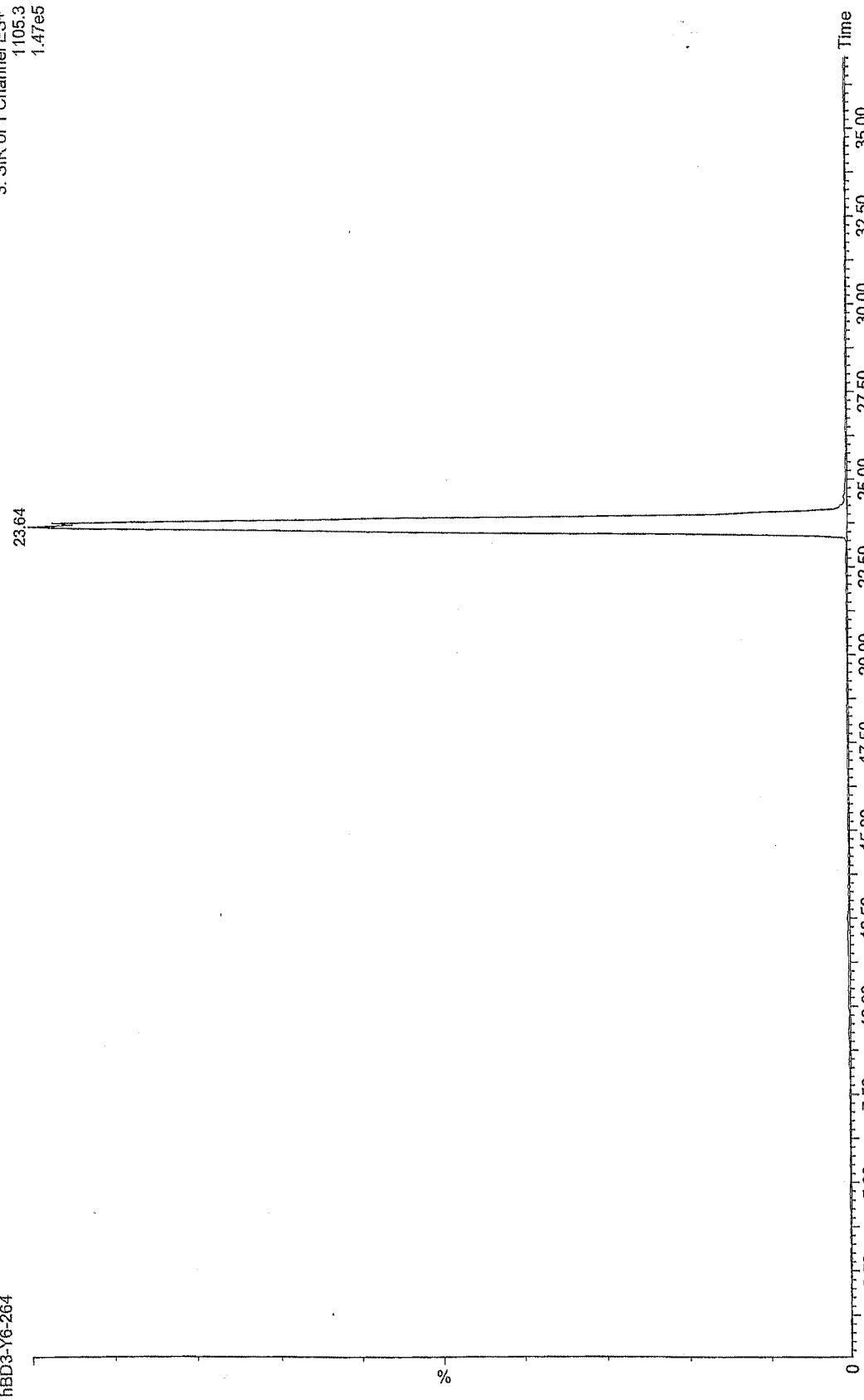


Figure 10B SIR chromatogram at m/z = 1105.3 ($[M+5H]^{5+}$)

Figure 10C ES + MS Spectrum

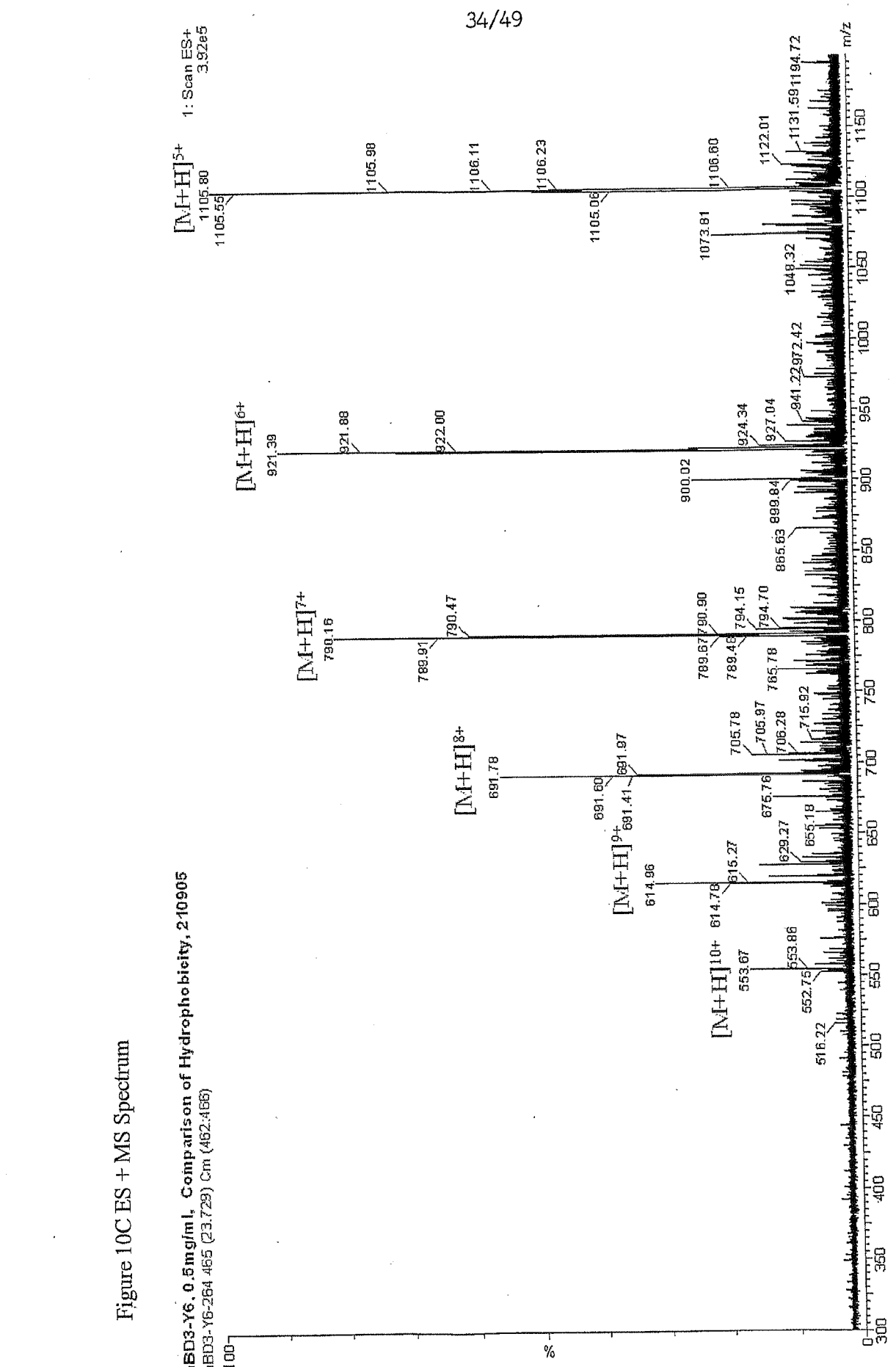


Figure 10D Deconvoluted MS

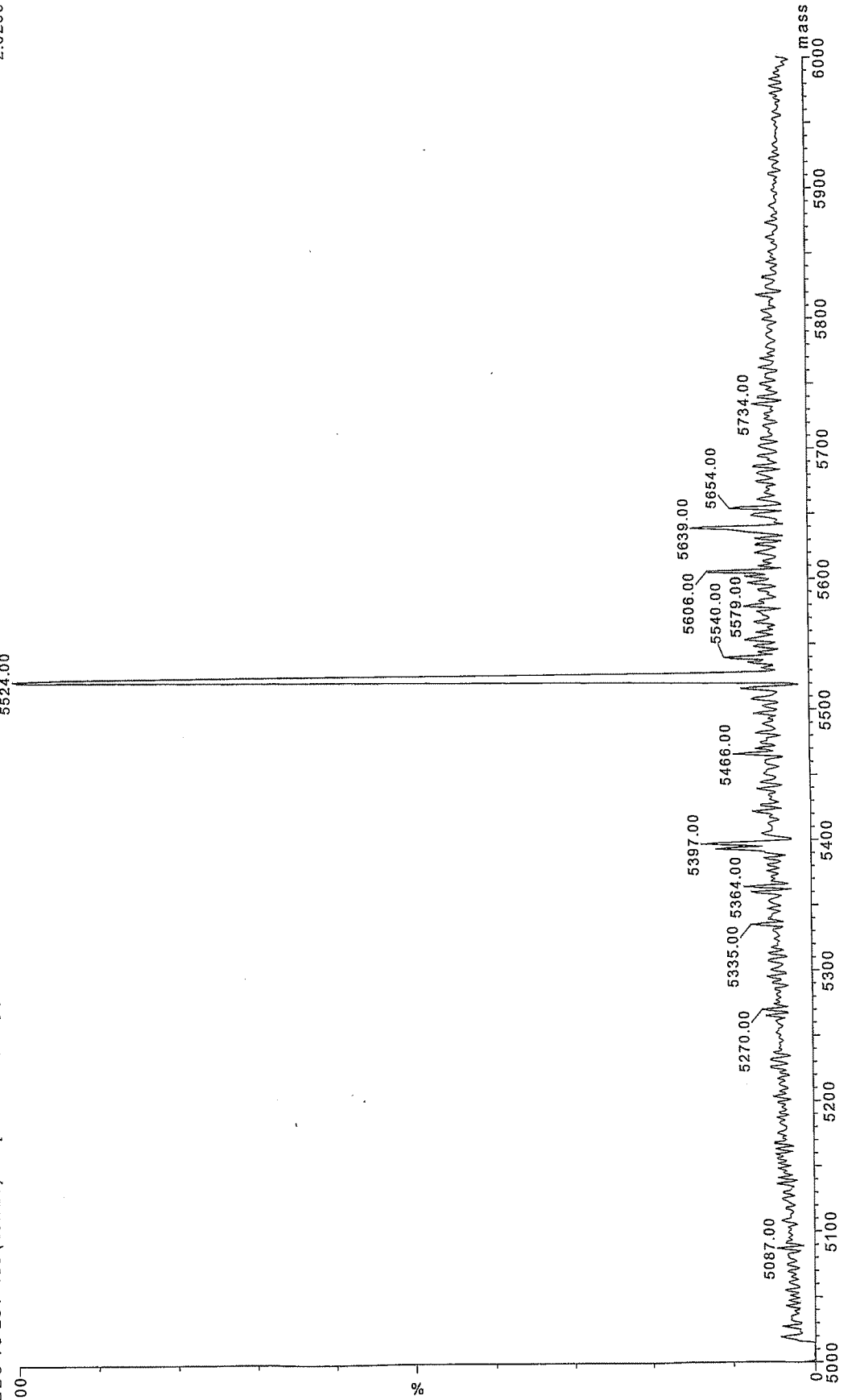
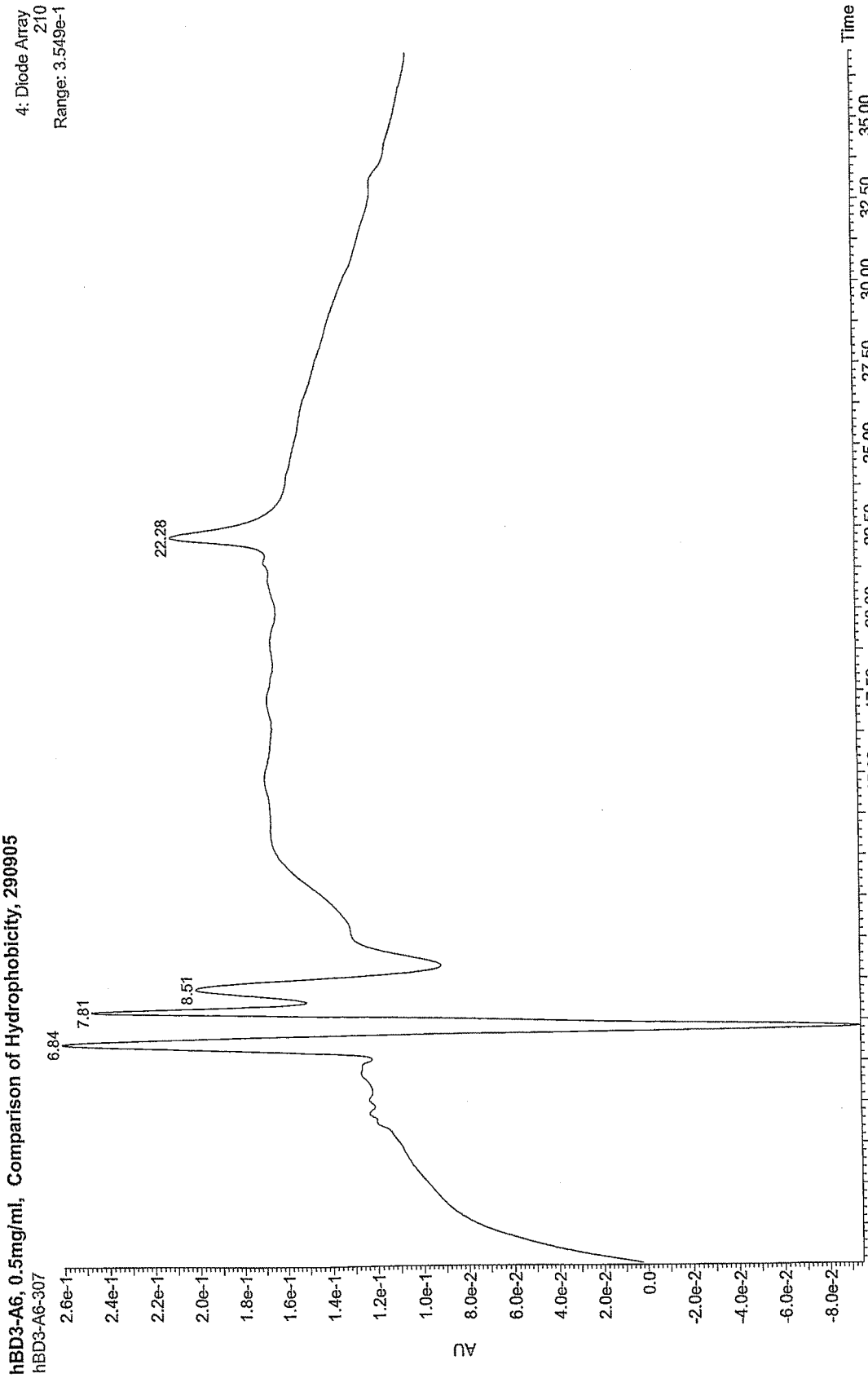


Figure 11A RP-HPLC-UV chromatogram and MS spectrum of A6



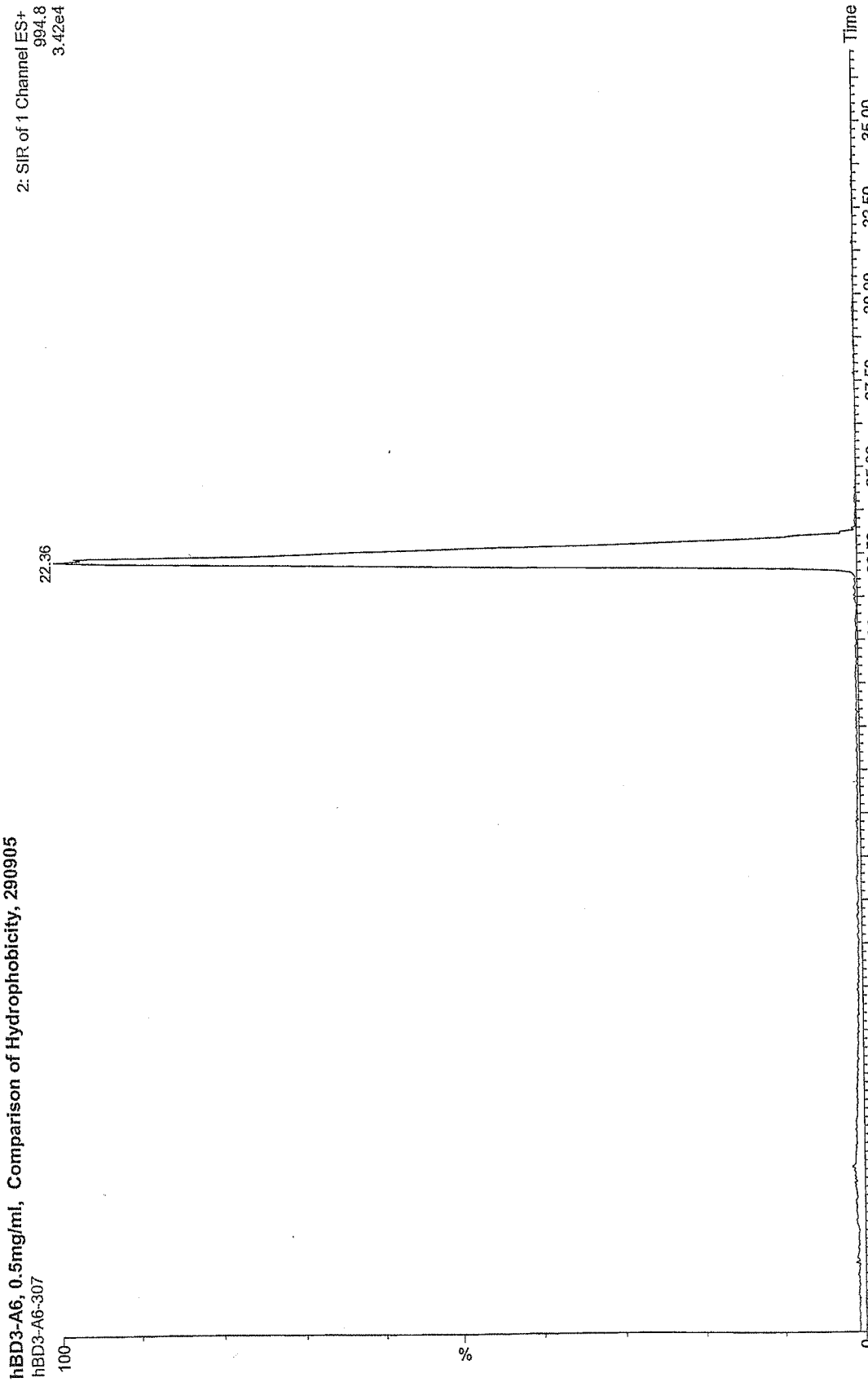


Figure 11B SIR chromatogram at m/z = 994.8 ($[M+5H]^{5+}$)

Figure 11C ES + MS specturm

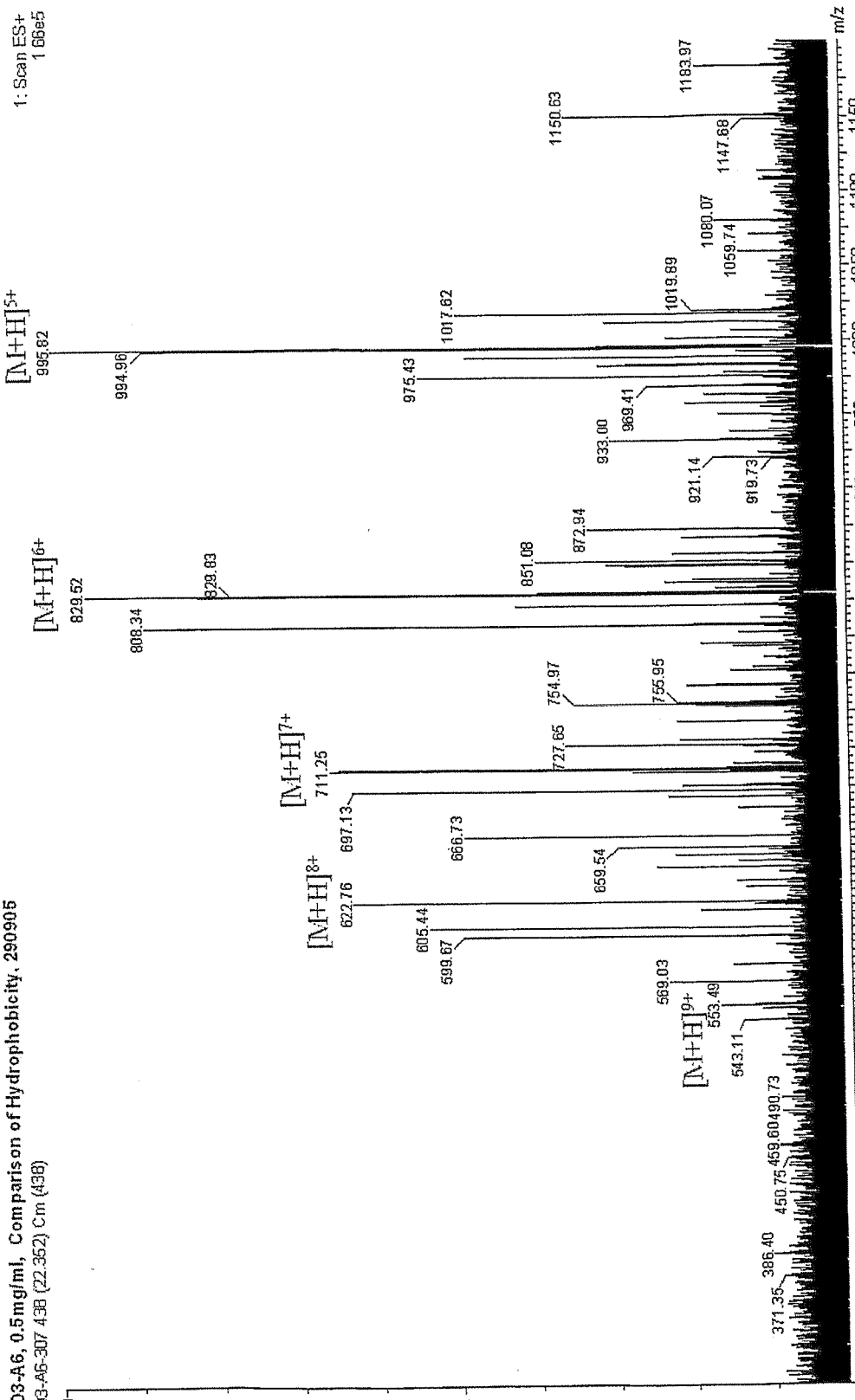


Figure 11D Deconvoluted MS spectrum

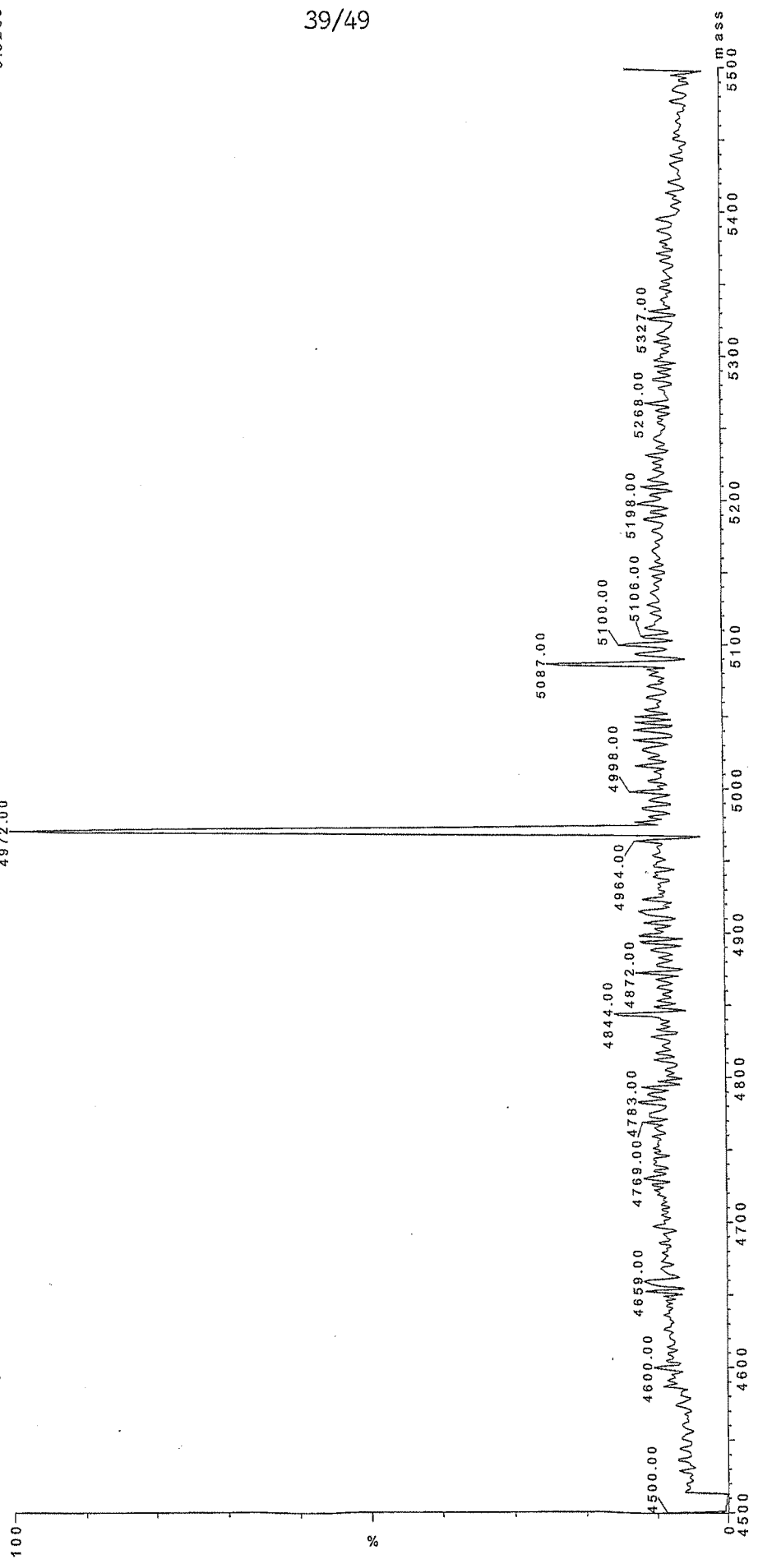
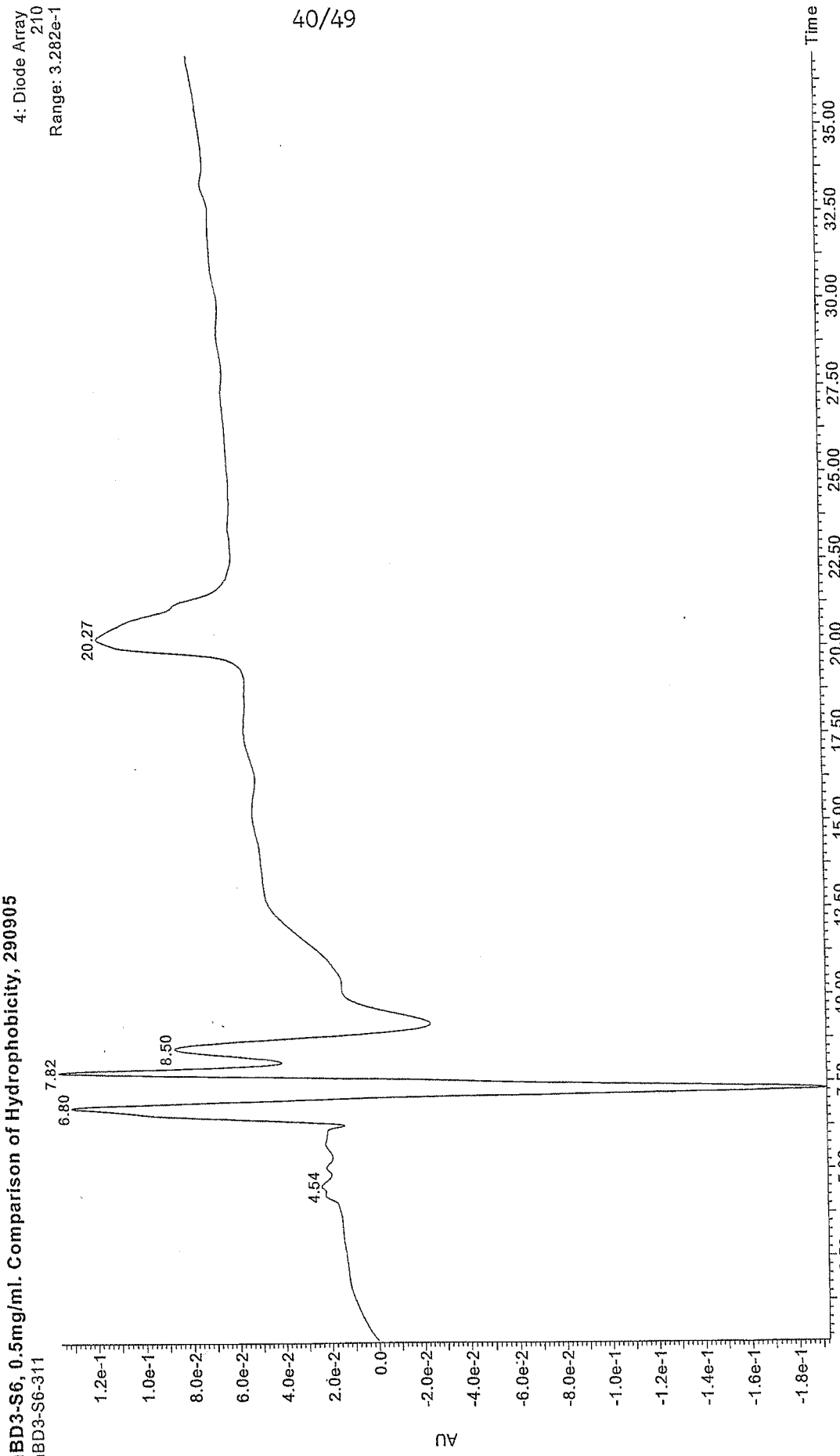


Figure 12A RP-HPLC-UV chromatogram and MS spectrum of S6

UV at 210 nm



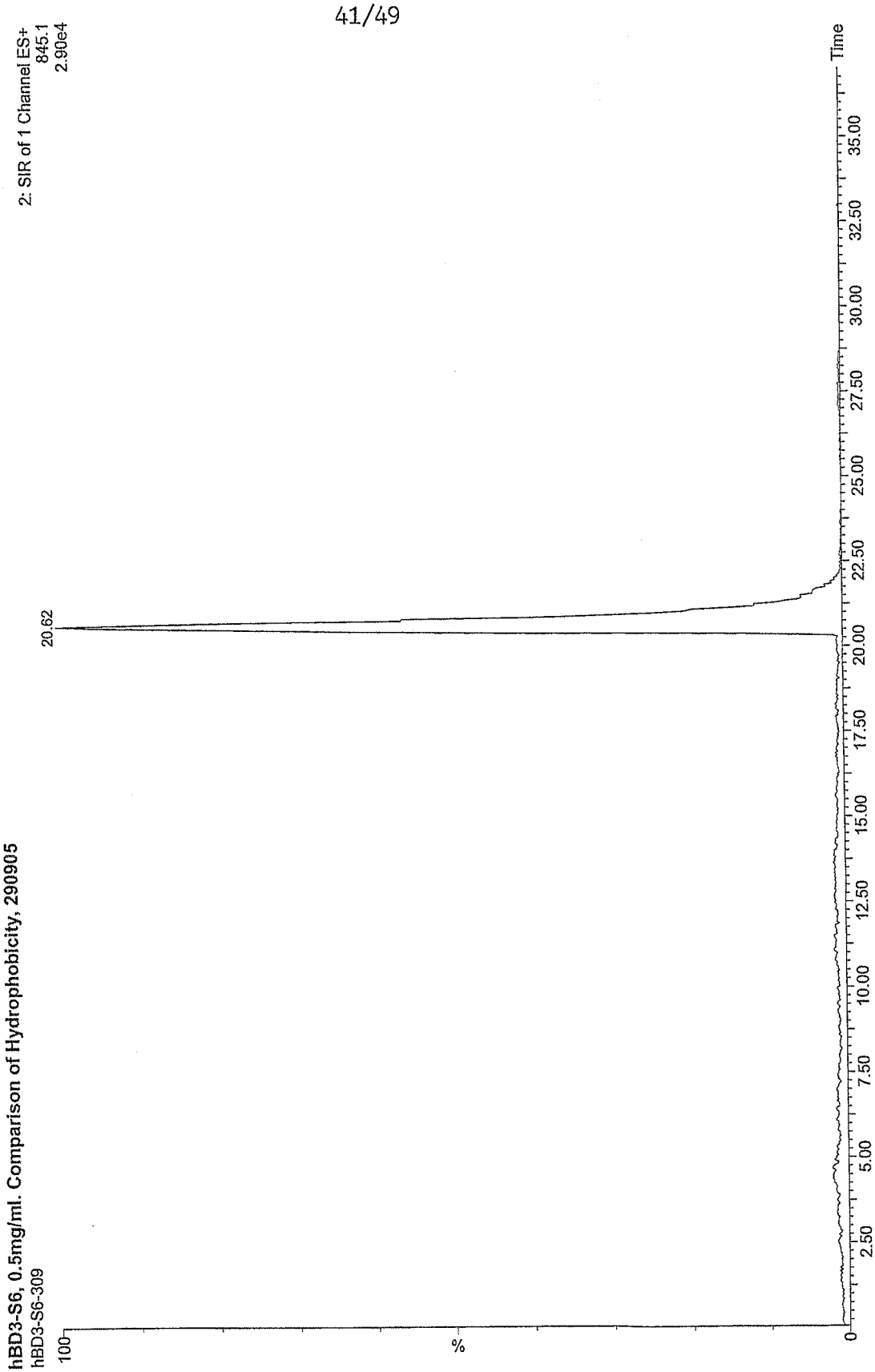
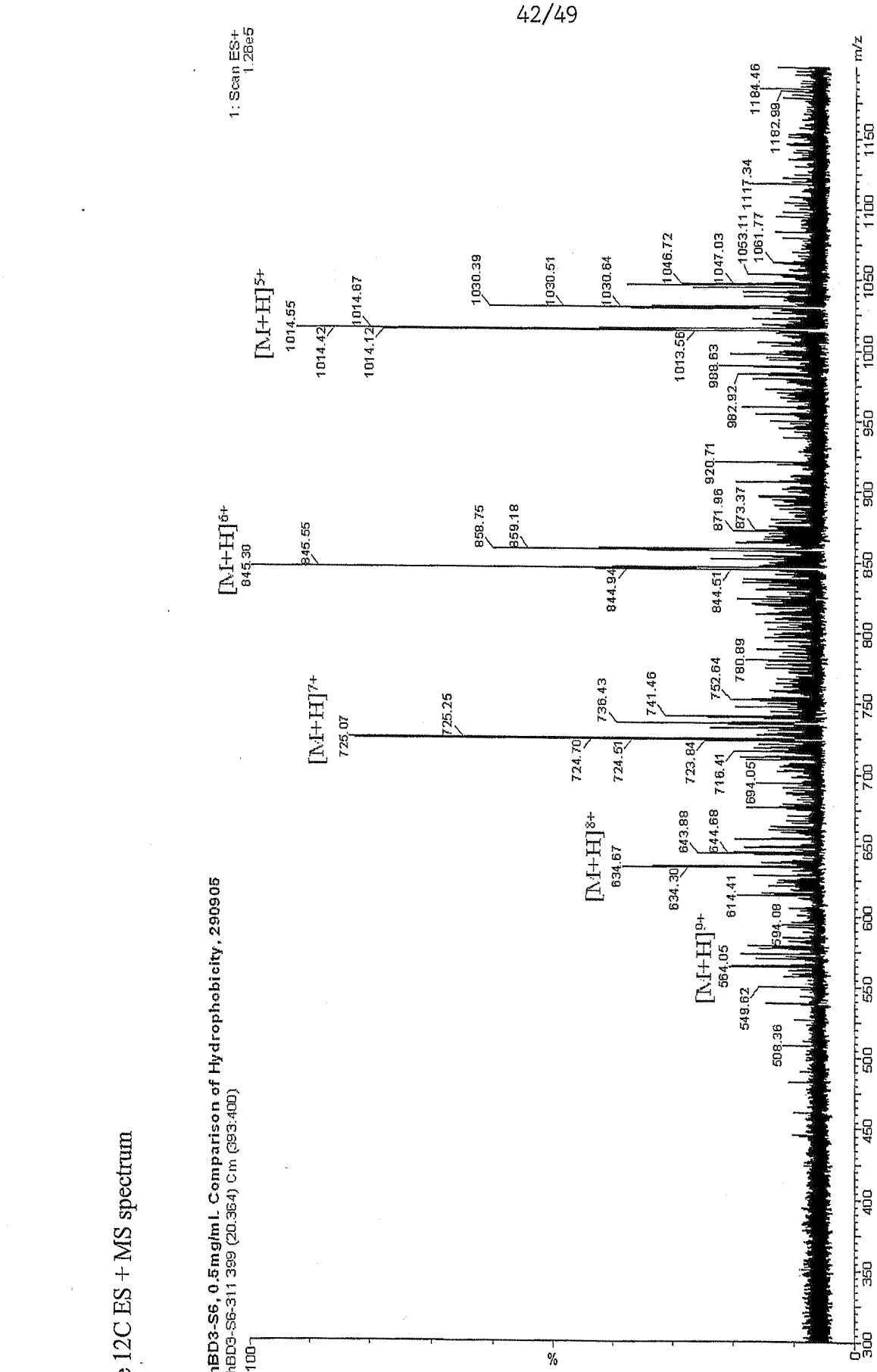


Figure 12B SIR chromatogram at m/z = 845.1 ($[M+6H]^{6+}$)

Figure 12C ES + MS spectrum



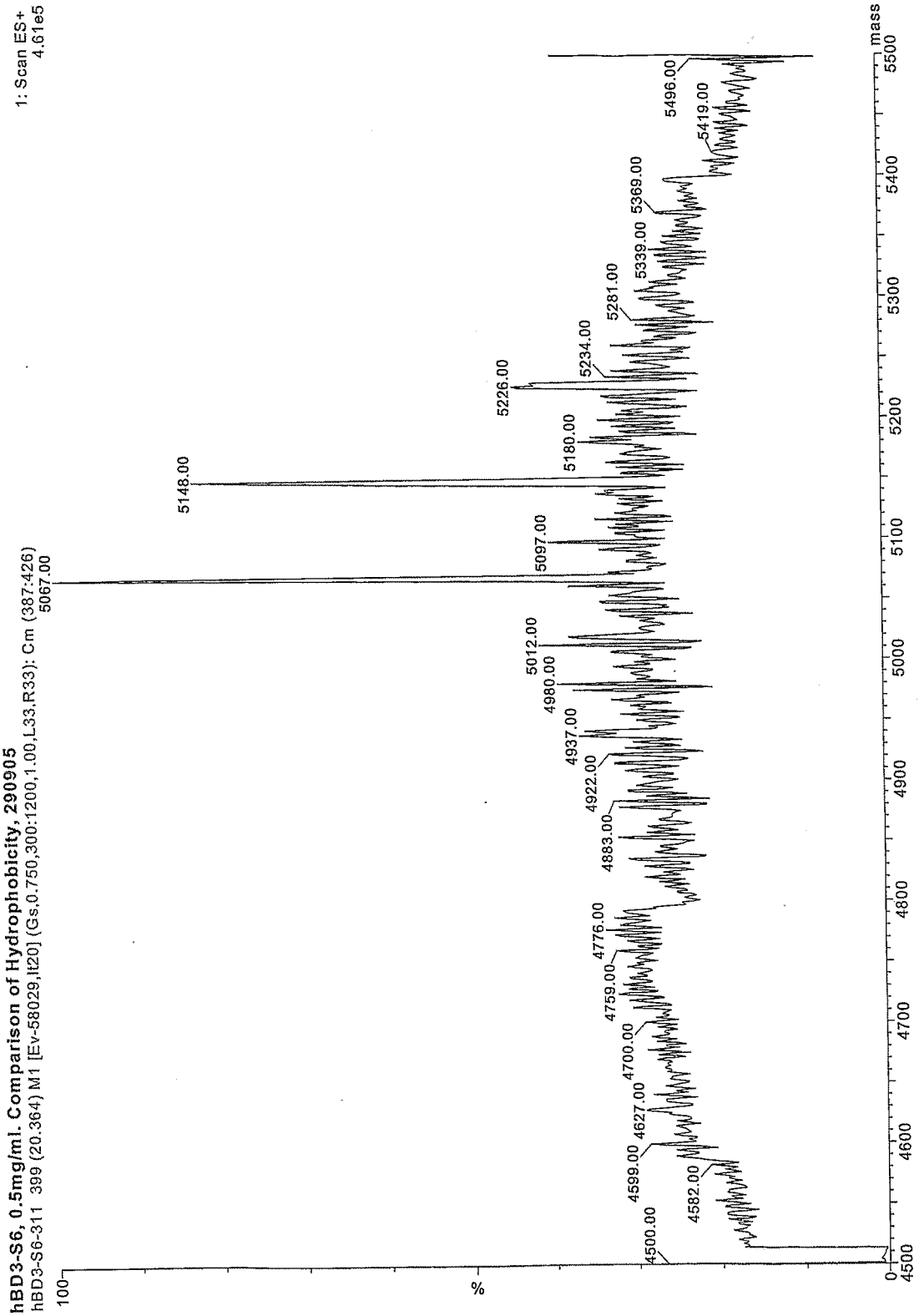
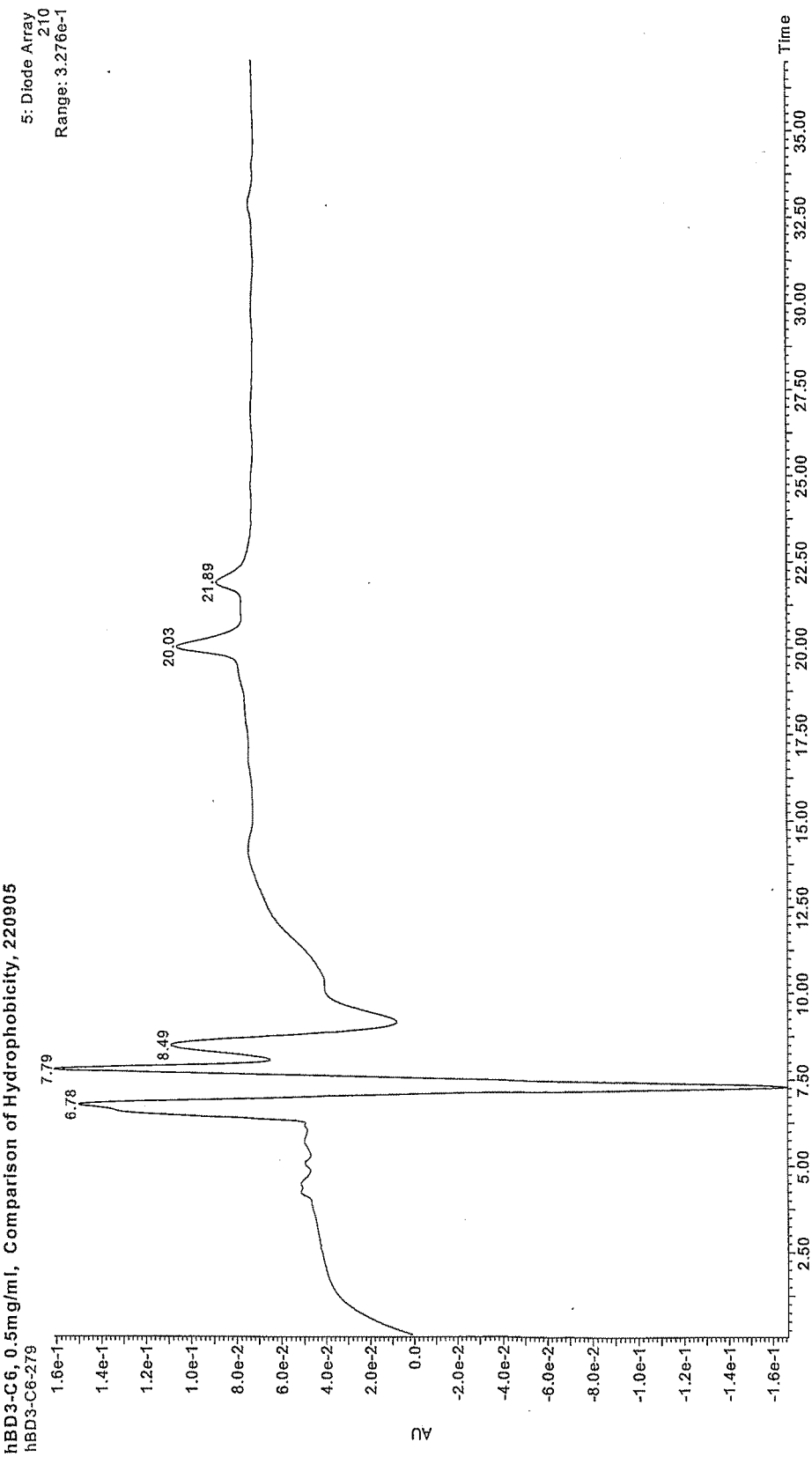


Figure 12D Deconvoluted MS spectrum

Figure 13A RP-HPLC-UV chromatogram and MS spectrum of C6 (sometimes coded C(Acm)6

UV at 210 nm



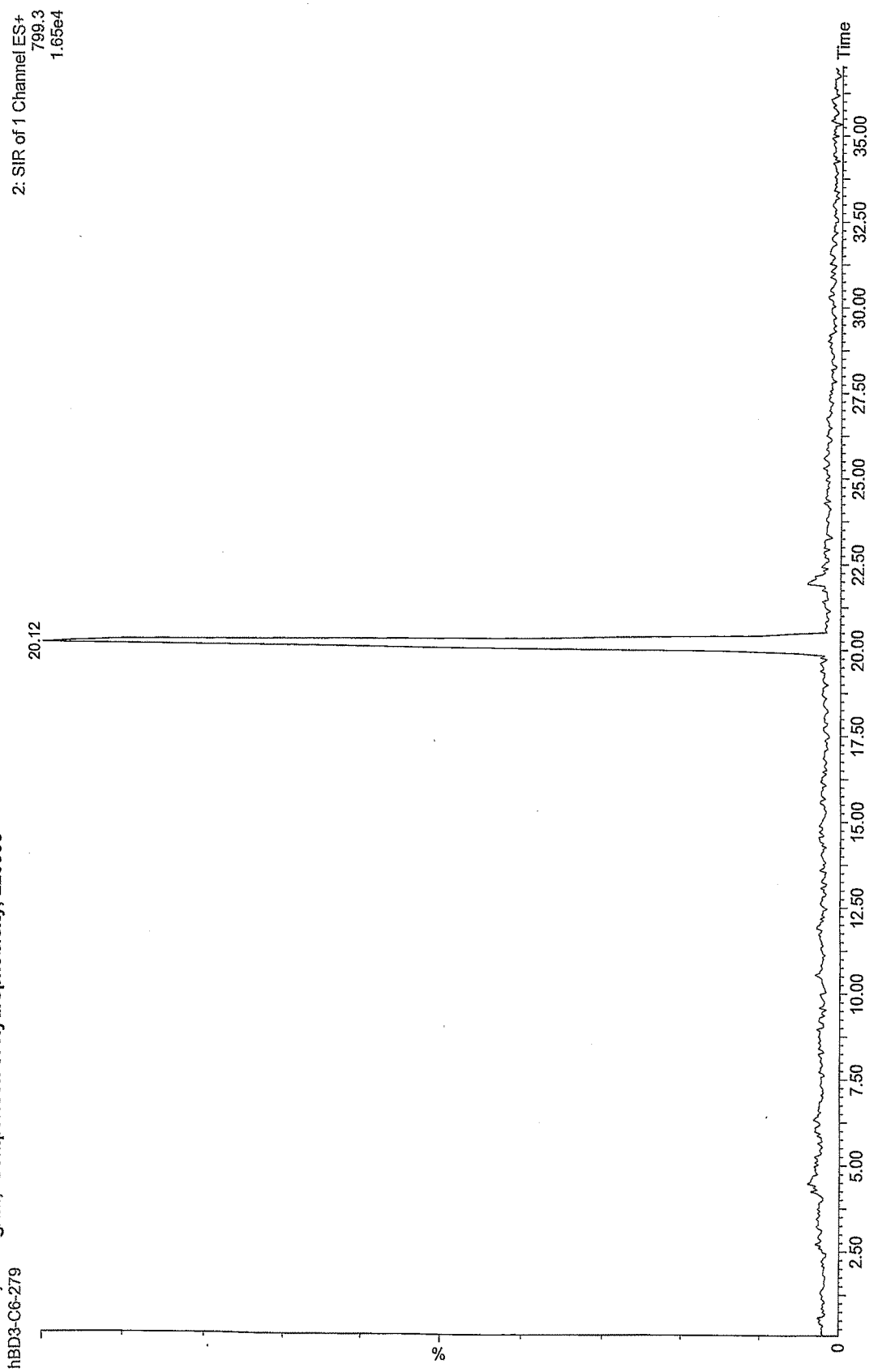


Figure 13B SIR chromatogram at m/z = 799.3 ($[M+7H]^{7+}$)

Figure 13C ES + MS spectrum

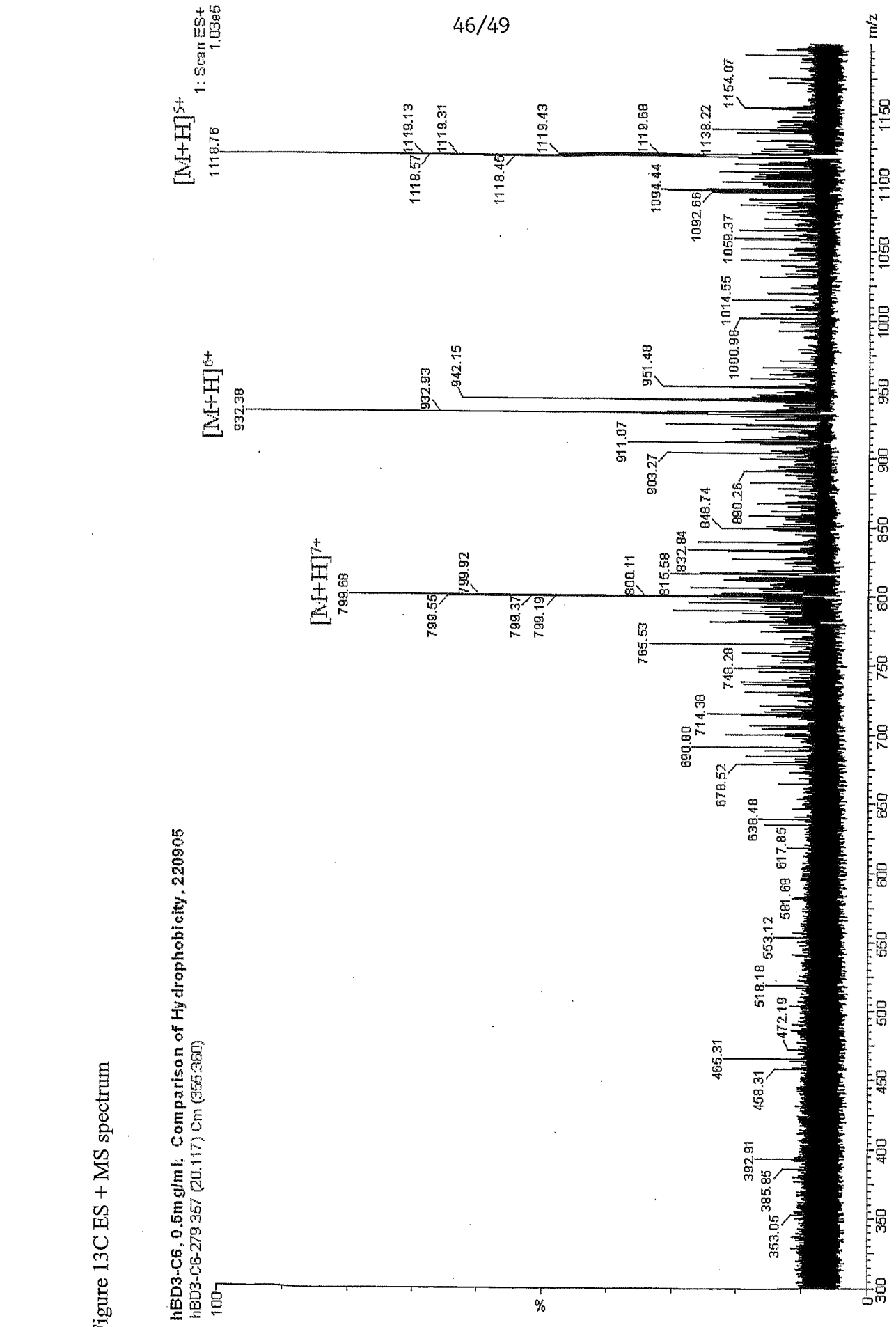


Figure 13D Deconvoluted MS spectrum

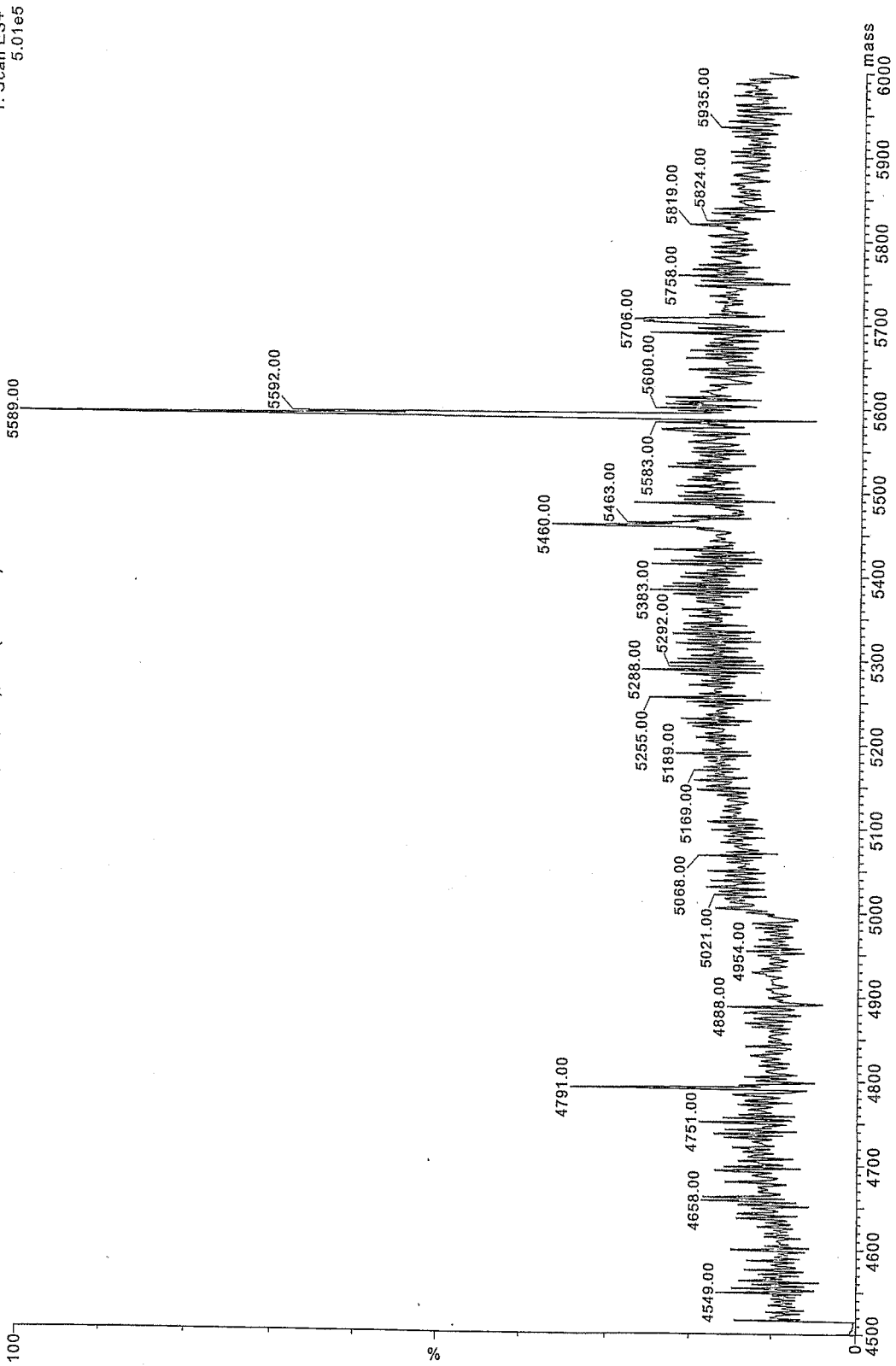


Figure 14 Comparison of overall molecular hydrophobicity of C-Terminus fragment peptides of hBD3 in terms of retention time of SIR chromatogram of charged fragments of peptides

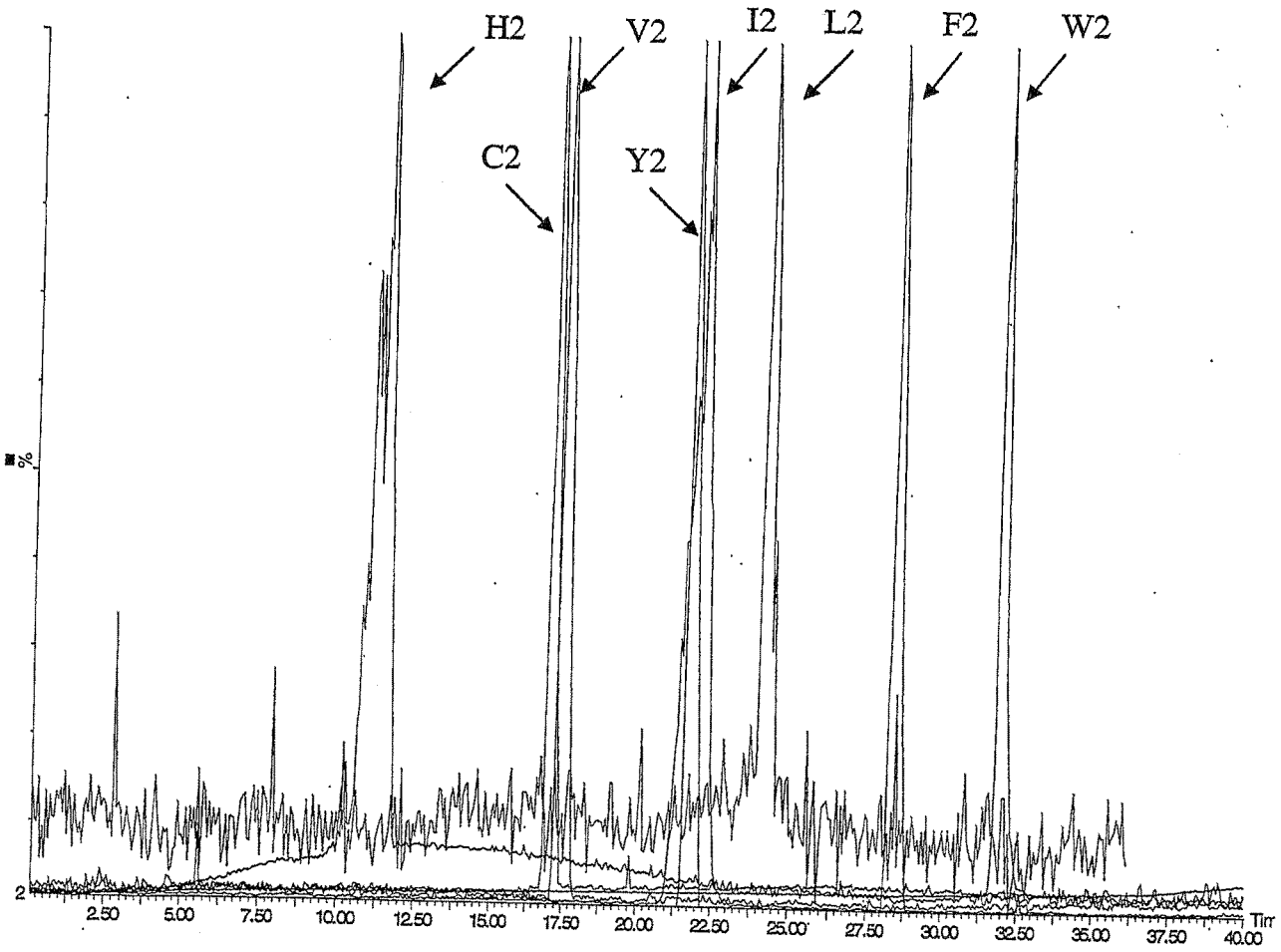
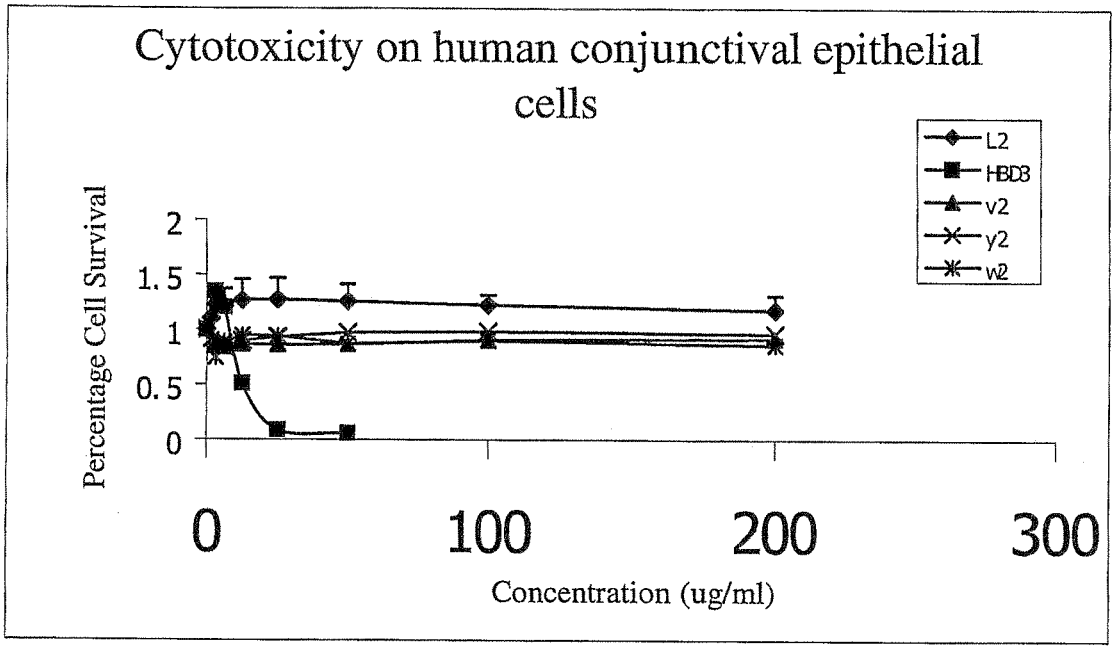


Figure 15 Cytotoxicity of peptides vs concentration



SEQUENCE LISTING

<110> Singapore Health Services Pte Ltd
Agency for Science, Technology and Research
Singapore Health Services Pte Ltd

<120> Antimicrobial peptides

<130> FP3651

<150> US 60/795,522
<151> 2006-04-27

<160> 45

<170> PatentIn version 3.4

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20 25 30

Cys Ser Thr Arg Gly Arg Lys Cys Cys Arg Arg Lys Lys
35 40 45

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<400> 2

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20 25 30

Xaa Ser Thr Arg Gly Arg Lys Xaa Xaa Arg Arg Lys Lys
35 40 45

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20 25 30

Arg Arg Lys Lys
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Arg Lys Xaa Xaa Arg Arg Lys Lys
35 40

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Xaa Ser Thr Arg Gly Arg Lys Xaa Xaa Arg Arg Lys Lys

20 25

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Arg Arg Lys Lys
20

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Leu Pro Lys

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Arg

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<220>
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<400> 13

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20 25 30

Trp Ser Thr Arg Gly Arg Lys Trp Trp Arg Arg Lys Lys
35 40 45

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<400> 14

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Arg Phe Ala Val Leu Ser Phe Leu Pro Lys Glu Glu Gln Ile Gly Lys
20 25 30

Phe Ser Thr Arg Gly Arg Lys Phe Phe Arg Arg Lys Lys
35 40 45

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20 25 30

Tyr Ser Thr Arg Gly Arg Lys Tyr Tyr Arg Arg Lys Lys
35 40 45

<210> 16
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20 25 30

Ser Ser Thr Arg Gly Arg Lys Ser Ser Arg Arg Lys Lys
35 40 45

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Arg Ala Ala Val Leu Ser Ala Leu Pro Lys Glu Glu Gln Ile Gly Lys
20 25 30

Ala Ser Thr Arg Gly Arg Lys Ala Ala Arg Arg Lys Lys
35 40 45

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20 25 30

Xaa Ser Thr Arg Gly Arg Lys Xaa Xaa Arg Arg Lys Lys
35 40 45

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20 25 30

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35 40 45

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20 25 30

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20 25 30

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35 40 45

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            20                  25                  30

Xaa Ser Thr Arg Gly Arg Lys Xaa Xaa Arg Arg Lys Lys
        35                  40                  45
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Xaa Ser Thr Arg Gly Arg Lys Xaa Xaa Arg Arg Lys Lys
        35                  40                  45
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20 25 30

Xaa Ser Thr Arg Gly Arg Lys Xaa Xaa Arg Arg Lys Lys
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20 25 30

Cys Ser Thr Arg Gly Arg Lys Xaa Cys Arg Arg Lys Lys
35 40 45

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20 25 30

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        35                  40                  45


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<400>  28

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Arg Xaa Ala Val Leu Ser Xaa Leu Pro Lys Glu Glu Gln Ile Gly Lys
            20                  25                  30


Xaa Ser Thr Arg Gly Arg Lys Xaa Xaa Arg Arg Lys Lys
        35                  40                  45


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<400>  29

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1               5                   10                  15


Leu Pro Lys Glu Glu Gln Ile Gly Lys Cys Ser Thr Arg Gly Arg Lys
            20                  25                  30


Cys Cys Arg Arg Lys Lys
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20 25 30

Arg Arg Lys Lys
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20

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Leu Pro Lys

<210> 36
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