| EXP. NUMBER | EXPERIMENT/SUBJECT | DATE |
|---|---|---|
| 1 | Acid/Base Properties of Amino Acids | 5/20/09 |

02

| NAME | LAB PARTNER | LOCKER/DESK NO. | COURSE & SECTION NO. |
|---|---|---|---|
| Jason Largen | Tim Van | | Bio Chem Lab |

Title: Acid/Base Properties of Amino Acids

Reference: handout on black board: Amino Acid titration
pg 29-32 Modern Exp. Bio Chemistry 3rd

Synopsis: Amino Acids can often be identified by performing a titration and plating the results. Amino Acids that contain ionizable R-groups will have 3 inflection points on the curve. The location of these show the pKa for each ionizable group and can be compared to known values. Given an unknown amino acid standardized sample a plot of the titration results using ~~1M~~ standardized HCl + ~~1M~~ NaOH will be produced. This plot will be used to find the pKa's, pI, and molecular weight to identify the sample.

Procedure:

NaOH Standardization
KHP sample dissolved into ≈100mL
DI water in 250 E-flask. 2drops Phenylphthalein added, Titrate w/NaOH ≈1M until pink
- do 2x's

HCl Standardization
Use exactly 10.00mL HCl ≈1M. into 250 mL E flask, add ≈50 mL DI water titrate w/ NaOH
- do 2x's

Observations
trial #1 2.1666g KHP
10.90mL NaOH
#2 2.2327g
11.00mL
first trial darker Pink.

#1 8.95mL NaOH
#2 8.95

Record all buret data including initial mL & final mL

-½

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| | | 5/20/09 | |

| NAME | LAB PARTNER | LOCKER/DESK NO. | COURSE & SECTION NO. |
|---|---|---|---|
| Jason Larson | | | |

Procedure Cont

Obtain ~.4 g of unknown x2
dissolve each into 20.00mL DI water

titrate #1 sample w/ NaOH
and #2 w/ HCl
titrate each until Reaching
a pH of 1.00 w/ HCl +
pH of 12.5 w/ NaOH
record mL titrant at intervals
of 0.2 to 0.3 pH changes.

Repeat titration using 20mL DI water
to use as a blank

pH measured using electronic pH meter
+ magnetic Stirrer

Unknown Sample #1

was pickel, but would
not dissolve, even after
heating to 75°C.

Switched to unknown #2.

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05

| NAME | LAB PARTNER | LOCKER/DESK NO. | COURSE & SECTION NO. |
|---|---|---|---|
| Jason Largo | | | |

Sample #1, 0.03938g

| pH | mL HCl titrated |
|---|---|
| 5.54 | 0 |
| 2.77 | 0.51 |
| 2.49 | 0,82 |
| 2.14 | 1,39 |
| 1,82 | 2,03 |
| 1,48 | 2,96 |
| 1,22 | 4,02 |
| 1,00 | 5,73 |

Water Blank, w/NaOH

| pH | mL NaOH |
|---|---|
| 6.44 | 0 |
| 11.52 | 0,05 |
| 11.81 | 0.10 |
| 11,94 | 0.15 |
| 12,22 | 0,20 |
| 12,31 | 0.28 |
| 12,40 | 0.40 |
| ~~11.45~~ | |
| 12,45 | 0,45 |
| 12,51 | 0,51 |

Sample #2, 0.3942g

| pH | mL NaOH titrated |
|---|---|
| 5,48 | 0 |
| 7.63 | 0,08 |
| 8.15 | 0,19 |
| 8,43 | 0,36 |
| 8,76 | 0.61 |
| 9,05 | 0.94 |
| 9,34 | 1,30 |
| 9.66 | 1,70 |
| 9,94 | 2,07 |
| 10,30 | 2,48 |
| 10,60 | 2,99 |
| 10,95 | 3,32 |
| 11,23 | 3,63 |
| 11,58 | 3,97 |
| 11,87 | 4,22 |
| 12,19 | 4,60 |
| 12,53 | 5,40 |

Water blank w/HCl

| pH | mL HCl | pH | mL HCl |
|---|---|---|---|
| 6.31 | 0 | 1,50 | 0,83 |
| 2,73 | 0,05 | 1,43 | 1,05 |
| 2,41 | 0,09 | 1,36 | 1,22 |
| 2,23 | 0,12 | 1,26 | 1,60 |
| 2,10 | 0,19 | 1,17 | 2,05 |
| 2,01 | 0,22 | 1,12 | 2,30 |
| 1,94 | 0,29 | 1,08 | 2,60 |
| 1,81 | 0,39 | 1,02 | 3,02 |
| 1,71 | 0,48 | | |
| 1,61 | 0,62 | | |

killed

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| | | 5/26/09 |

06

| NAME | LAB PARTNER | LOCKER/DESK NO. | COURSE & SECTION NO. |
|---|---|---|---|
| Jason Lurger | | | |

## Calculations

NaOH Molarity #1 2.1666g KHP / 204.2 g/mol / 0.01090L = 0.9734 M

#2 2.2327g / 204.2 g/mol / 0.0110L = 0.9940

Avg = 0.9837 M NaOH

Show work for averages

HCl molarity #1 ~~8.95 mL~~ ~~0.00895L NaOH added~~ / ~~NaOH M~~ (0.00895L)(0.9837M) / 0.010L = 0.8804

mL HCl titrated

MW Acid = 393.8 mg / 2.403 mmol = 163.9 g/mol

base 1 = 394.2 / 2.5 = 157.68

what about at 3rd equivalence?

Avg mw = 148.79 g/mol #2 Same volume

Avg = 0.8804 M HCl

Missing uncorrected titration curves and how you determined water corrected values (-1)

Include this table)

## Discussion/Conclusions

There seems 2 be 3 inflection points, suggesting that our amino acid has an ionizable R group. One ~~inflection~~ pka appears just below a pH of 2, ~~around~~ ~~1.90~~, at ~~1.5~~ 1.25 millimoles of Acid added. ~~Using 1.5 as the equivalents~~ 1/2

Using 1.25 as the equivalents, and looking at the graph, the second pka is ≈ 9.0 PH, and the third is more difficult to determine. If 1.25 is correct, the third should be at 3.75, but this is very close to a sharp rise, at pH11 the graph seems to have a flat point around ph 12.2. The average Molecular Weight was 148.79 g/mol

Identifying the unknown is tough, as the graph doesn't clearly show the 2 basic pKas as they are near each other. It seems to be ① arginine, tyrosine or lysine. The graph most closely resembles arganine to me, however this is unlikely due to only needing to go to 12.5 PH during titration. The mw suggest Lysine, which is next closest to the graph as well.

Lysine was unknown?

Lysine mw = 147, pka1 = 2.16, 2 = 9.18, R = 10.79

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| | | | 12/5/15 |

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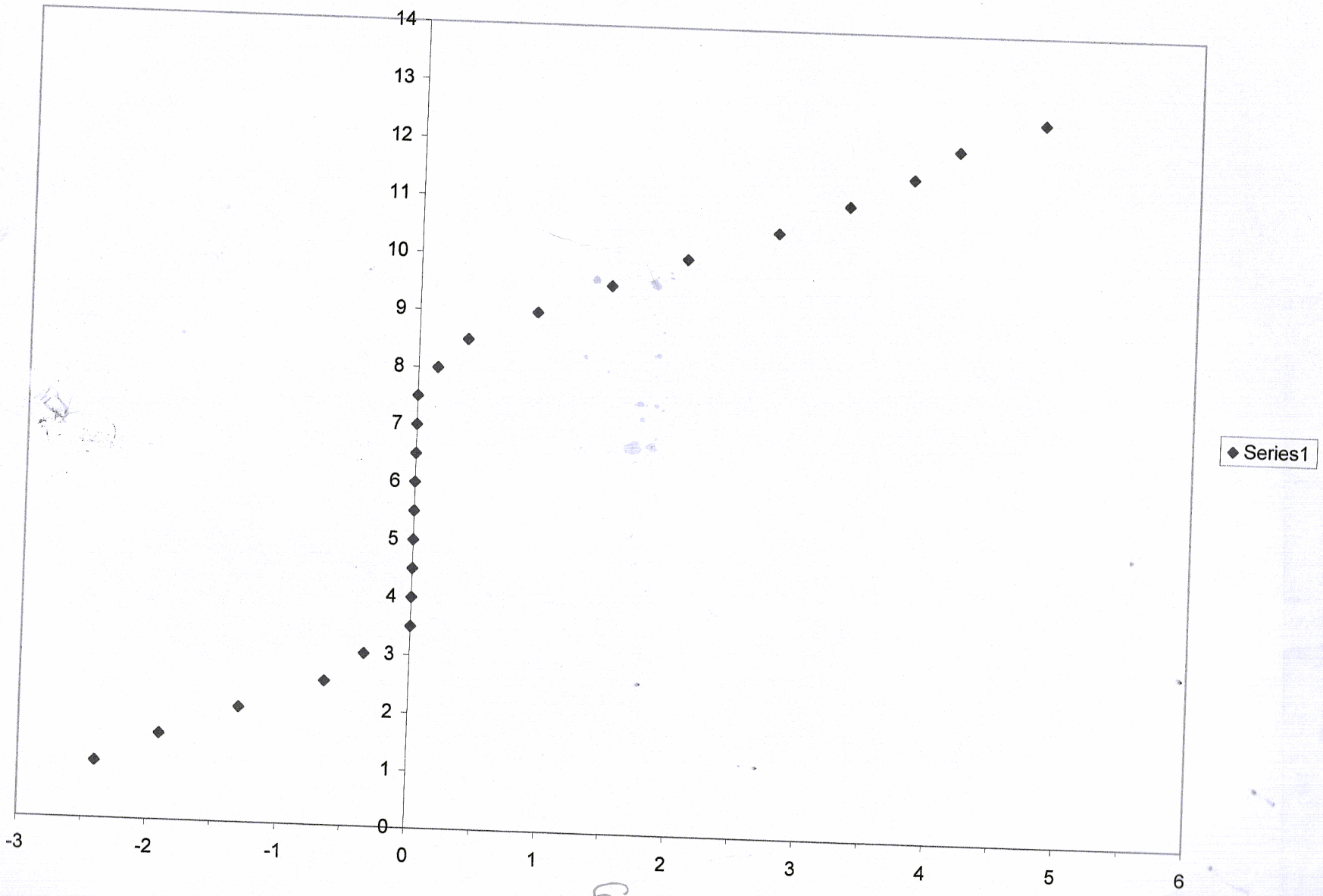


Graph not labelled with equivalent pts & pka values

(-1)