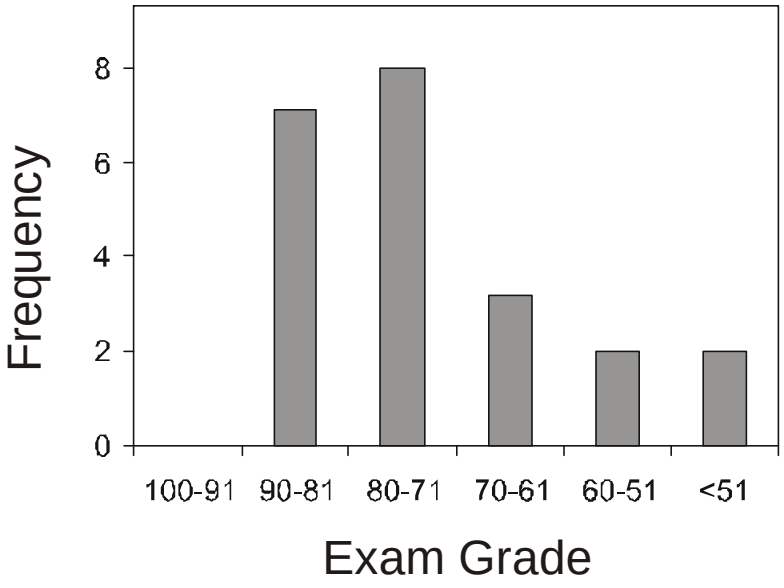
Chemistry 5361/8361 Thursday, December 5, 2002

Midterm Exam 3
Answer Key

Exam 2 Mean: 72
Exam 2 Median: 78
Exam 2 St. Dev.: 16

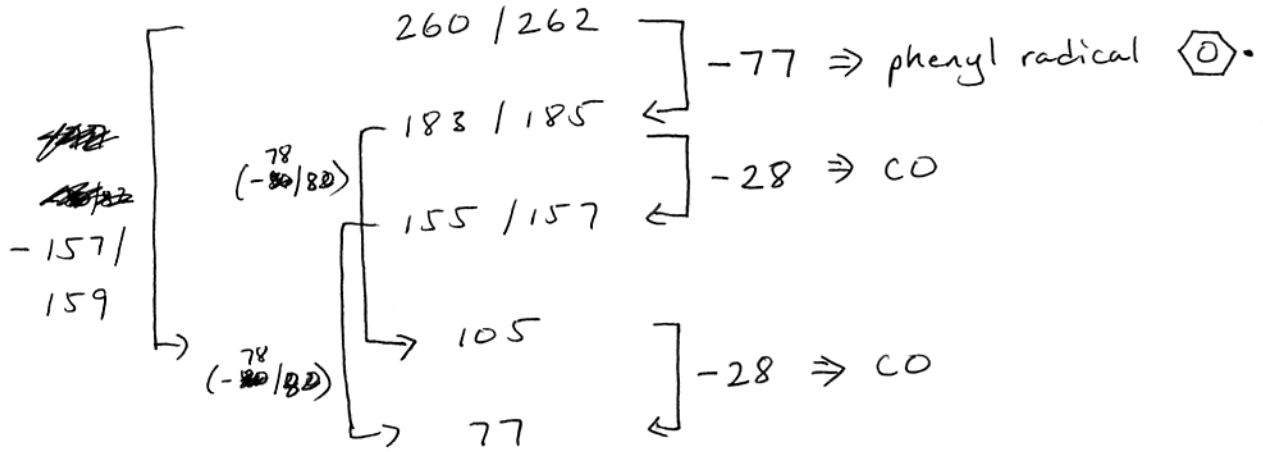


EXAM 3 SOLUTIONS

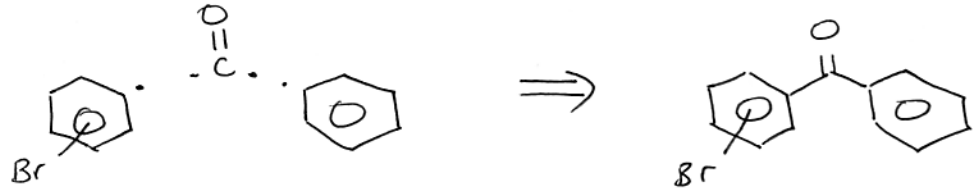
1. KEY PIECES OF INFORMATION HERE:

- THE ^{13}C NMR SPECTRUM SHOWS 8 PEAKS IN THE ARYL REGION, SO WE KNOW OUR UNKNOWN HAS AT LEAST 8 ARYL CARBONS. COULD BE MORE THOUGH - EQUIVALENT CARBONS WOULD GIVE JUST ONE CARBON PEAK.
- ^1H NMR IS CONSISTENT W/ ABOVE - LOTS OF ARYL PROTONS.
- ^{13}C NMR ALSO SHOWS A CARBONYL PEAK @ $\delta = 195.4$ ppm.
- THE EI-MS SHOWS A NUMBER OF M/M+2 PEAK PAIRS WITH 1:1 PEAK INTENSITY — 155-157, 183-185, AND 260-262. THESE STRONGLY SUGGEST THE PRESENCE OF BROMINE IN ALL OF THESE IONS; ABUNDANCE ($^{79}Br : {}^{81}Br$) = 1:1.

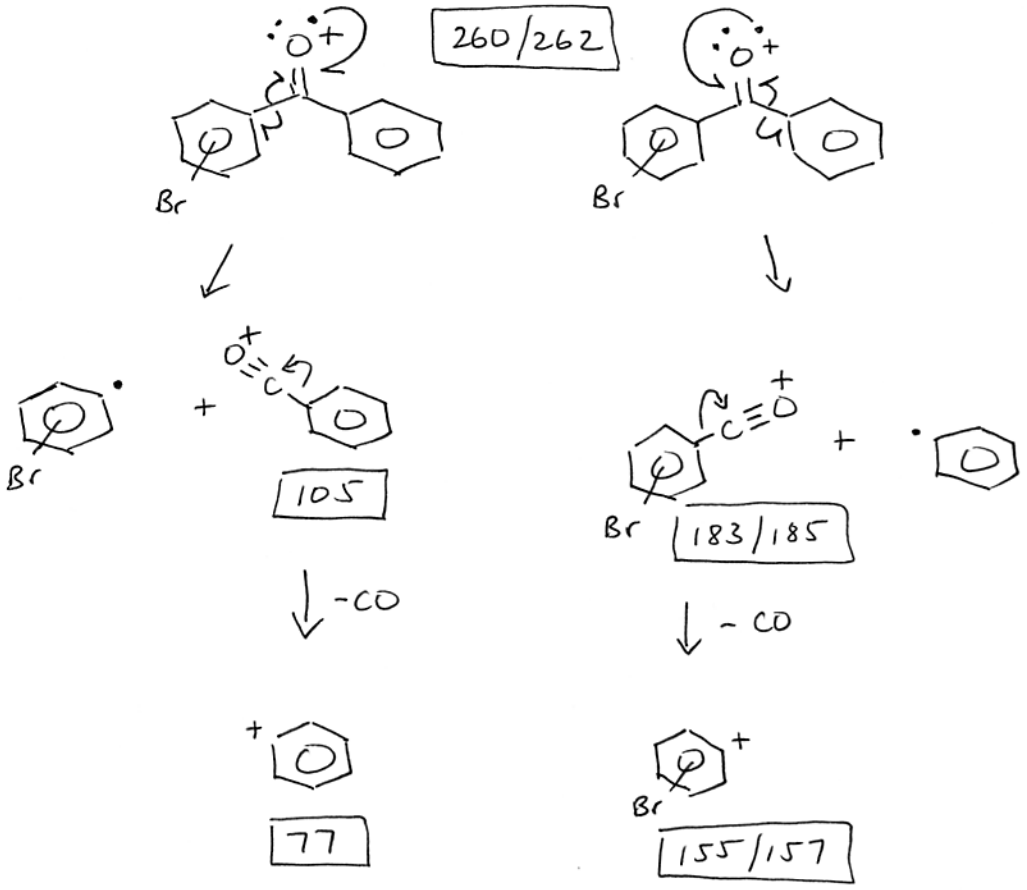
OBSERVED LOSSES FOLLOW A DEFINITE PATTERN:



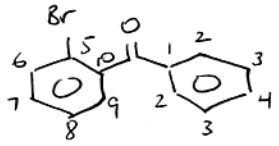
I'VE WRITTEN "-78/80" IN PARENTHESES BECAUSE THESE AREN'T POSSIBLE LOSSES; BROMINE WEIGHS 79/81. BUT -157/159 IS POSSIBLE: (79/81 + 78). IF 78 IS ANOTHER ARYL GROUP, THEN COULD BE



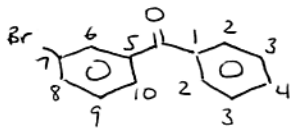
THIS MAKES EXCELLENT SENSE:



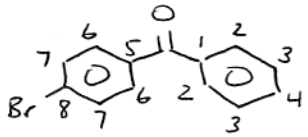
SO, WHERE IS THE BROMINE? ONLY ONE OF THREE POSSIBILITIES (ortho, meta, para) GIVES 8 ARYL ^{13}C PEAKS:



10 ^{13}C PEAKS EXPECTED.

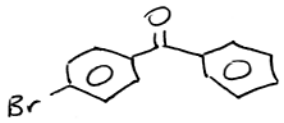


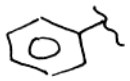
10 ^{13}C PEAKS EXPECTED



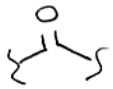
8 ^{13}C PEAKS EXPECTED

SO, ANSWER IS p-bromobenzophenone,



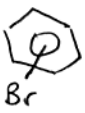


4 POINTS



4 POINTS (1 POINT FOR ALDEHYDE.)

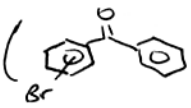
-Br 2 POINTS FOR BROMINE.



2 POINTS (w/ BROMINE ANYWHERE)

2 POINTS FOR BROMINE IN THE RIGHT PLACE, ON ARYL RING (para).

(x3) 2 POINTS EACH FOR ANY STRUCTURE THAT GIVES PLAUSIBLE m/z = 77, 105, AND 183/185 FRAGMENTS.

( GETS ALL OF THESE POINTS.)

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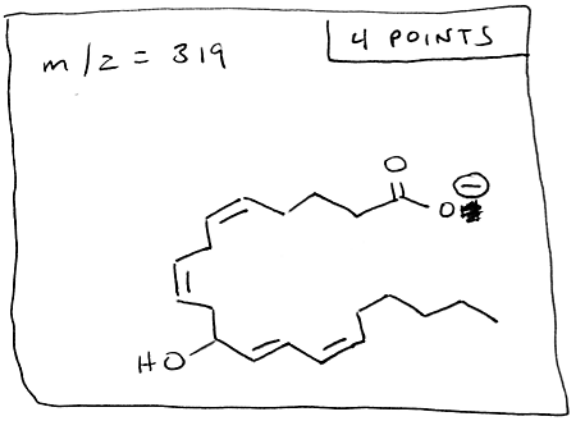
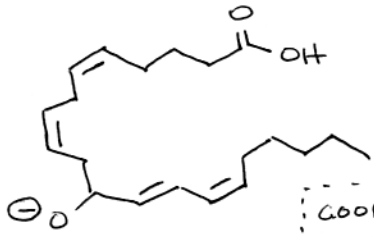
20 POINTS TOTAL.

2. a) 11-HETE IS BEING ANALYZED BY NEGATIVE ION ESI-MS. SO, m/z = 319 IS MOST LIKELY DEPROTONATED ($[M-H]^-$) VERSION OF m/z = 320.

m/z = 319 — 4 POINTS



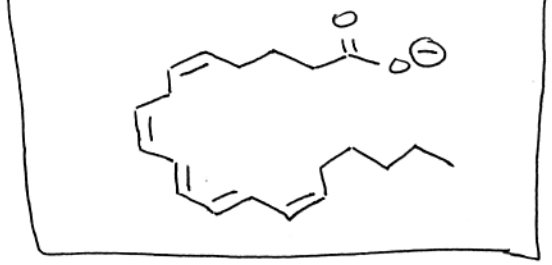
LESS LIKELY, BUT STILL PRESENT:



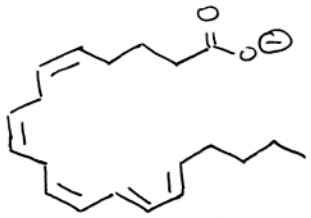
GOOD FOR 3 POINTS

319 → 301 IS LOSS OF 18 ($-H_2O$):

m/z = 301 — 4 POINTS



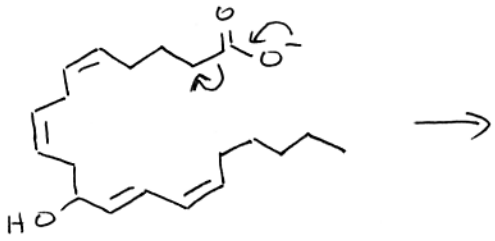
OR

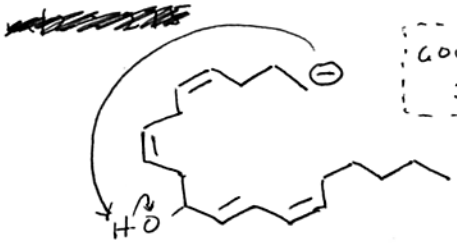


GOOD FOR 3 POINTS

(IF DEPROTONATES FROM OTHER END, THOUGH LESS PLAUSIBLE)

319 → 275 IS LOSS OF 44 ($-CO_2$):

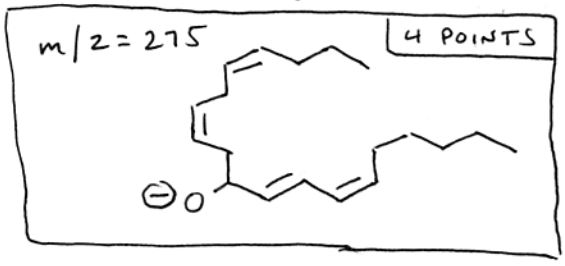




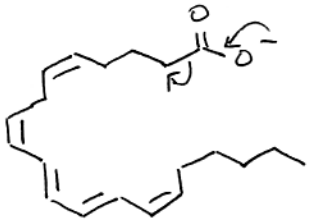
GOOD FOR 3 POINTS

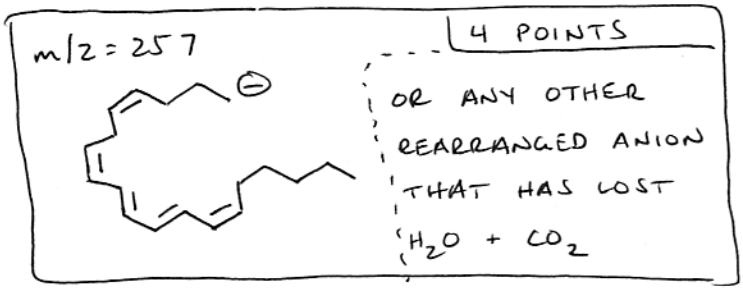
MOLECULE IS FLEXIBLE; H^+ TRANSFER WORKS WELL.

m/z = 275 — 4 POINTS

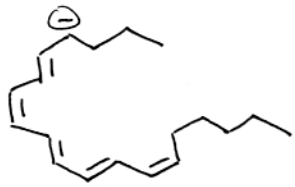


319 → 301 → 257 IS LOSS OF 18 + 44 ($-H_2O$, $-CO$)





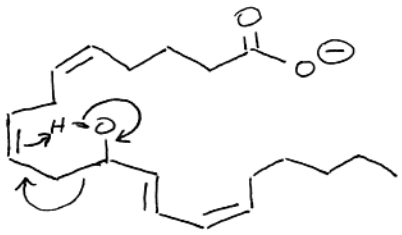
↓ i.e., H^+ transfer

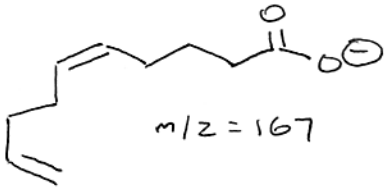


~~BOTH OF THESE REARRANGEMENTS OCCUR FROM THE~~

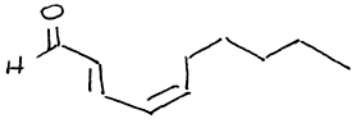
~~OXIDATION:~~

b)





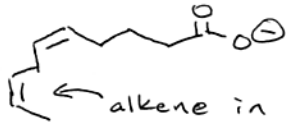
+



SCORING:

6 POINTS FOR DAUGHTER ION STRUCTURE.

2 POINTS FOR ANY DAUGHTER THAT CLEAVES IN RIGHT PLACE (i.e.,



2 POINTS FOR ANY m/z = 167 ION.

2 POINTS FOR NEUTRAL FRAGMENT. MUST BE CORRECT.

6 POINTS FOR MECHANISM

2 POINTS FOR ANY 6-MEMBERED CYCLIC MECHANISM (CANNOT BE RADICAL, BUT OTHERWISE ANYTHING GOES).

3. a. ALL THREE SPECTRA CONTAIN SERIES OF PEAKS THAT CORRESPOND TO MULTIPLY CHARGED IONS. SPECTRUM (i) CONTAINS ONLY ONE ION SERIES, THAT OF THE HIV-P MONOMER

 .

CAN SOLVE W/ A SET OF SIMULTANEOUS EQUATIONS, USING $m/z = 2157$ & 1798 PEAKS AS AN EXAMPLE:

$$\frac{(m+z)}{z} = 2157 \qquad \frac{(m+z+1)}{z+1} = 1798$$

$$m+z = 2157z \qquad m+z+1 = 1798(z+1)$$

$$m = 2156z \qquad m = 1797z + 1797$$

$$2156z = 1797z + 1797$$

$z = 5$ (for 2157 peak)

$m = 10780$

SO, IN SPECTRUM (i),

m/z		z	ion mass
1541	$(\boxed{E} \cdot 7H)^{7+}$	+7	10787
1798	$(\boxed{E} \cdot 6H)^{6+}$	+6	10786
2157	$(\boxed{E} \cdot 5H)^{5+}$	+5	10785
2696	$(\boxed{E} \cdot 4H)^{4+}$	+4	10784

SPECTRUM (ii) HAS SAME  PEAKS AS SPECTRUM (ii), PLUS SOME NEW ONES. SINCE THE SOLUTION IS STILL DILUTE, WE MIGHT IMAGINE THAT THESE PEAKS CORRESPOND TO  SIMULTANEOUS EQUATIONS.

ON $m/z = 1912$ AND 2295:

$$\frac{(m+z)}{z} = 2295 \qquad \frac{(m+z+1)}{z+1} = 1912$$

$$m + z = 2295z \qquad (m+z+1) = 1912(z+1)$$

$$m = 2294z \qquad m = 1911z + 1911$$

$$2294z = 1911z + 1911$$

$z = 5$ (for 2295 peak)

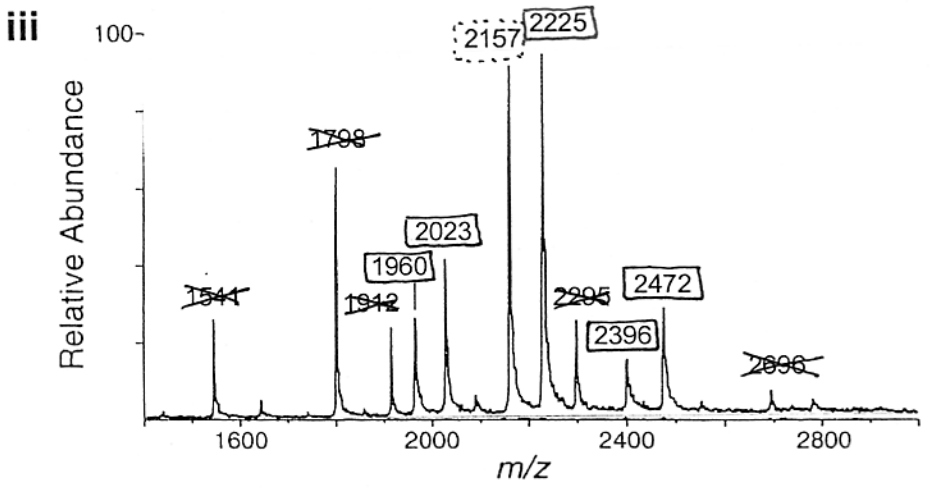
$m = 11470$

SO, IN SPECTRUM (ii),

m/z		z	ion mass
1912	$\left(\boxed{} \cdot 6H \right)^{6+}$	+6	11476
2295	$\left(\boxed{} \cdot 5H \right)^{5+}$	+5	11475
2867	$\left(\boxed{} \cdot 4H \right)^{4+}$	+4	11474

(PEPSTATIN A WEIGHS 690 DALTONS.)

IN SPECTRUM (iii), ONLY 5 PEAKS ARE UNACCOUNTED FOR:



PRESUMABLY, THESE CORRESPOND TO  AND  .

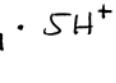
(empty) (filled)

AS IN SPECTRUM (ii), THE PEAKS ARE PARTNERED

$1960 \rightarrow 2023$, $? \rightarrow 2225$, $2396 \rightarrow 2472$,

BUT $m/z = 2225$ APPEARS TO BE MISSING ITS PARTNER.

IT ISN'T: 2157 IS ITS PARTNER, WHICH MAKES SENSE:

m/z ( $\cdot 5H^+$) = m/z ( $\cdot 10H^+$).

↑ twice the mass, twice the charge.

WE CAN MAKE ABSOLUTELY SURE OF THIS BY DOING SIMULTANEOUS EQUATIONS:

FOR THE 1960 - 2157 PAIR,

$$\frac{m+z}{z} = 2157 \qquad \frac{m+z+1}{z+1} = 1960$$

$$m+z = 2157z \qquad m+z+1 = 1960z + 1960$$

$$m = 2156z \qquad m = 1959z + 1959$$

$$2156z = 1959z + 1959$$

$z = 10$ (for 2157 peak)

$m = 21560$

m/z		z	ion mass
1960	$(\square \cdot 11H)^{11+}$	+11	21571
2157	$(\square \cdot 10H)^{10+}$	+10	21570
2396	$(\square \cdot 9H)^{9+}$	+9	21569

FOR THE 2023 - 2225 PAIR,

$$\frac{m+z}{z} = 2225 \qquad \frac{m+z+1}{z+1} = 2023$$

$$m+z = 2225z \qquad m+z+1 = 2023(z+1)$$

$$m = 2224z \qquad m = 2022z + 2022$$

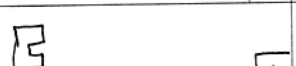
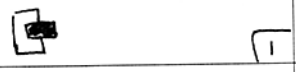
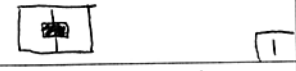
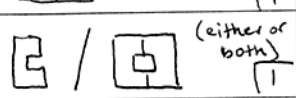
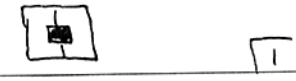
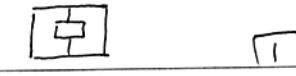
$$2224z = 2022z + 2022$$

$z = 10$ (for 2225 peak)

$m = 22240$

m/z		z	ion mass
2023		+11	22251
2225		+10	22250
2472		+9	22249

SCORING:

m/z ratio	cartoon	ion charge (z)	ion mass (m)
1541		+7	10787
1798		+6	10786
1912		+6	11476
1960		+11	21571
2023		+11	22251
2157	 (either or both)	+5/+10	10785/ 21570
2225		+10	22250
2295		+5	11475
2396		+9	21569
2472		+9	22249
2696		+4	10784

1 POINT EACH CARTOON;

1 POINT FOR Z AND M TOGETHER

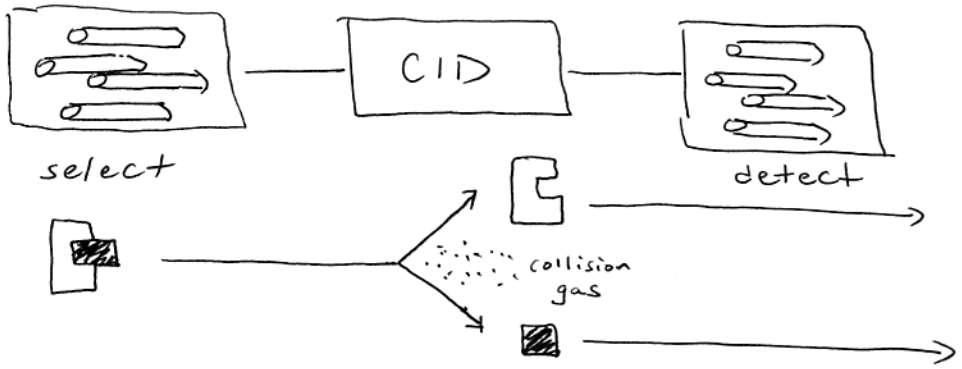
1 POINT FOR THIS PAIR CORRECT

2 POINTS FOR ALL THREE OF THESE CORRECT

11 POINTS | 11 POINTS | 3 "BONUS" POINTS

= 25 POINTS TOTAL

b) IF THE INHIBITOR WERE NONCOVALENTLY BOUND, IT SHOULD BE POSSIBLE TO DISPLACE IT IN A TANDEM MS/MS, COLLISION-INDUCED DISSOCIATION (CID) EXPERIMENT:

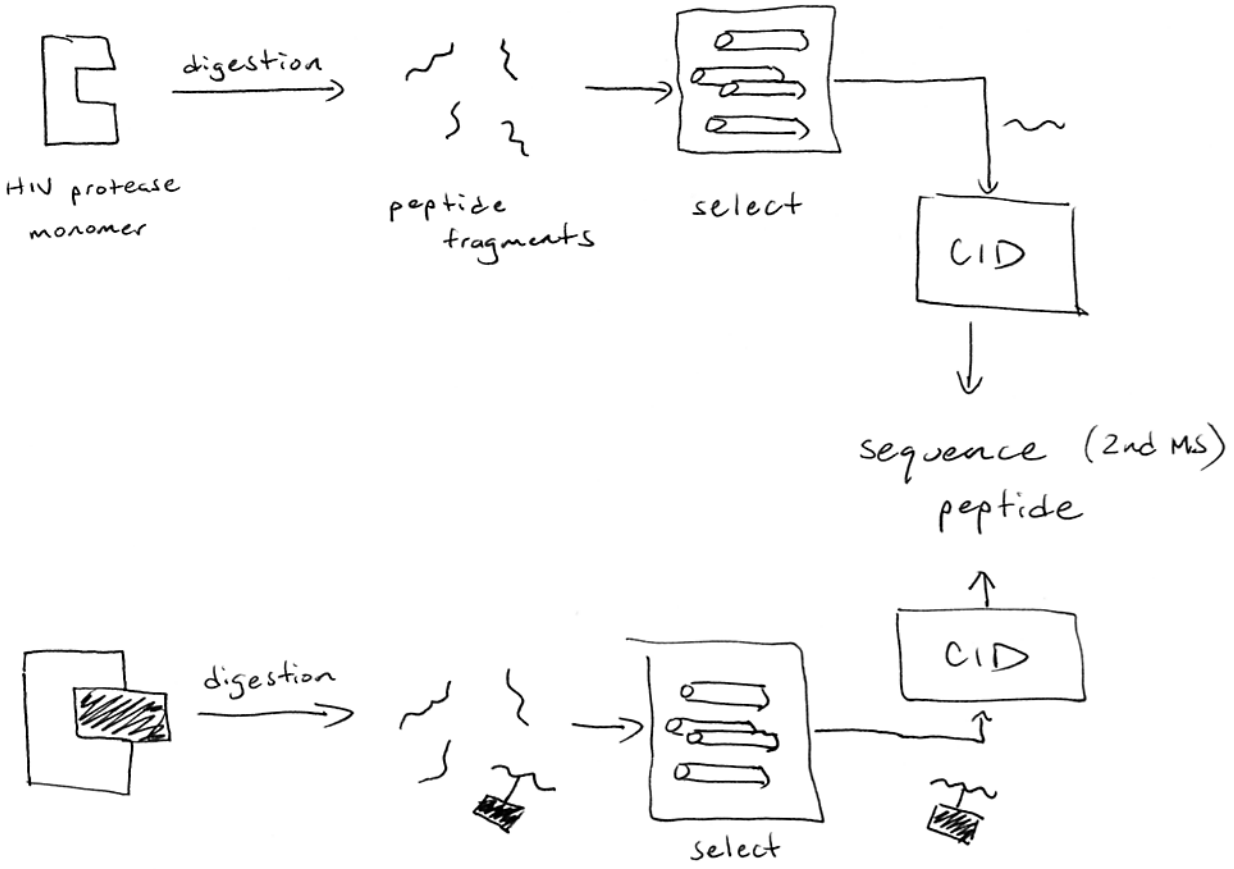


IF COMPLEX IS NONCOVALENT, THIS SHOULD BE VERY EASILY OBSERVED. IF COMPLEX IS COVALENT IT SHOULD BE VERY DIFFICULT OR IMPOSSIBLE; HARD CID WOULD DO IT, BUT ALSO BREAK OTHER BONDS IN THE PROTEIN AS WELL.

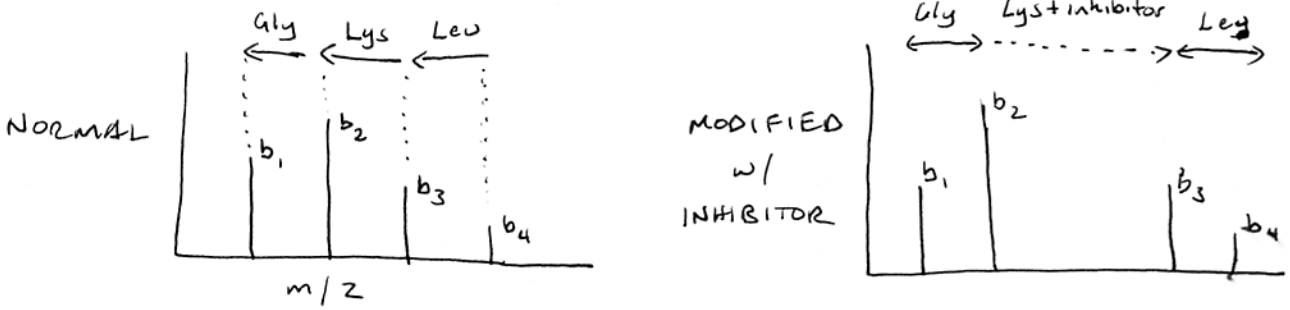
ALSO POSSIBLE: CHEW UP PROTEIN ENZYMATICALLY, AND SEE WHETHER ANY FRAGMENTS ARE HEAVIER THAN THEY SHOULD BE IN MS. THIS IS ACTUALLY THE ANSWER TO PART (c), BUT IT WOULD WORK HERE TOO.

SCORING: 15 POINTS FOR SOMETHING THAT WOULD WORK.
10 POINTS FOR SOMETHING THAT WOULD WORK PHYSICALLY, BUT HAS FATAL FLAW.
5 POINTS FOR ANY MASS SPEC EXPERIMENT.

c) THE BEST WAY TO DO THIS IS PROBABLY AN AN ANALYSIS OF FRAGMENTS OF HIV PROTEASE. ENZYMATIC CLEAVAGE OF HIV PROTEASE BREAKS IT UP INTO PEPTIDE PIECES. THESE PIECES CAN THEN BE ANALYZED BY MS-MS:



THE ONE PEPTIDE FRAGMENT THAT CHANGES MASS IS THE ONE THAT BINDS THE INHIBITOR. THIS FRAGMENT CAN BE ANALYZED BY CID:



WHEREVER CID CLEAVAGE PATTERN DIFFERS FROM PROTEIN WITHOUT INHIBITOR ATTACHED, THAT IS WHERE INHIBITOR IS BOUND.

SCORING: 10 POINTS FOR SOMETHING THAT WOULD WORK.
5 POINTS FOR ANY MASS SPEC EXPERIMENT.