Concentration (mg/ml) (1.55 x A280) 0.76 x A280)

Steps

(A) 3mL of phosphate buffer — blank

3mL of 3 unknown solutions total

1. add 20mL of goat liver + 2980 buffer
2. add 40mL of goat liver + 2960 buffer
3. add 60mL of goat liver + 2940 buffer

(B) Spectrophotometer to 280 nm, measure blank + 3 unknowns

1. 0.201 Ab
2. 0.439 Ab
3. 0.735 Ab

(C) Dilution factor

1. $\frac{3000}{20} = 150$ 2. $\frac{3000}{40} = 75$ 3. $\frac{3000}{60} = 50$

Scanned with CamScanner

D. Beer Lambert's Law [ratio: ↑A ∝ ↑C or ↓A ∝ ↓C]

$A = \varepsilon b C$ → Concentration looking for

- A → absorbance
- ε → extinct. Coefficient literature
- b → Path length = 1cm

Extraction of lactate dehydrogenase

- liver (goat)
- muscle
- heart

1. 0.135 M concentration
2. 0.295 M concentration
3. 0.493 M concentration

Scanned with CamScanner

① $A = \varepsilon b c$

$0.201 = (1.49)(1)c$

$0.135 = c$ concentration.

$\frac{1}{\text{meter}} \times \frac{\quad}{\quad}$ $\qquad M^{-1} = \frac{1}{M} \quad \frac{1}{cm}$

$0,201 = 1.49\,M^{-1}\,cm^{-1} \times 1\,cm \times c$

$0,201 = 1.49\,M^{-1} \times c$

② $0.439 = (1.49)(1)c$

$0.295 = c$ Concentration

$C \times 1,49\,M^{-1} = 0,201$

③ $0.735 = (1.49)(1)c$

$0.493 = c$ Concentration

$C = \frac{0,201}{1.49\,M^{-1}}$

$= 0,135\,M$

$C \times 1.49\,M^{-1} = .439$

$C = \frac{.439}{1.49\,M^{-1}} = .295\,M$

$C \times 1.49\,M^{-1} = .735$

$C = \frac{.735}{1.49\,M^{-1}}$

$= .493$

Scanned with CamScanner

d.f

$\frac{3000}{20} = 150$

① 20 ml + ~~3000~~ 2980 ml = ~~3020~~ 3000 ml

smol buffer ab = 0.201

② 40 ml + ~~3000~~ 2960 ml = ~~3040 ml~~ 3000 ml

$\frac{3000}{40} = 75$

ab = 0.439

③ 60 ml + 2940 ml = $\frac{3000}{60} = 50$

ab: 0.735

↓↑ A ∝ ↑ C l Beer Lambert law

① A = εbC → ext. coefficient ↳ Literature 41.49 $m^{-1}cm^{-1}$

Path length = 1 cm

0.201 =

C = ?

Extraction of Lactase dehydrogenase

↳ liver

↳ muscle

↳ heart

Scanned with CamScanner