Notes for talk on Brooklyn work

for Thursday Sept. 30th 1954.

Introduction

{ limited to ribonuclease – not colleges, plane rides or gossip.

" " my work on ribonuclease : brief mention of other work (no other x-ray work)

general picture 1 3D Patterson

2 Isomorphous replacement studies.

3D Patterson

Mag. Lab. & Crith.

data collected by others – moderately accurate. (errors increase with $\frac{1}{d}$)

computed by Hagdoff on I.B.M. machines. (origin removed)

Average intensity curve : shows low to Å peak.

Plotting of the Patterson – Vittorio's method.

Each contour (assuming Fourier value) ≈ 100 elec.^{2}/Å^3.

5 Å ~~not~~ shell nor complete isotropic.

Minor plane effect : show sections : give explanation.

not enhanced peakiness is due to coherence of the vectors not their number.

Harker Section suggests second molecule is not near $x=\frac{2}{3}=0$.

Molecular location : plotting languinous.
(can be done by plotting the intensities).

Rods no rods anywhere.

Sequence of lumps in the c direction.

but other rods ① not very long ($\frac{1}{3}$ / length (35Å) of α helix)
② others in other directions. (remember relative weighting)

A mean "rod direction" = Elliotts Infra-red direction.

(also shown in asymmetry of 10Å · 5Å reflections).

Ribonuclease VI (Magdoff + Crick)

Grown using dye iodo-phenol blue
(tertiary butyl).

Coloured deep blue : highly pleochroic when view along a^*
strong absorption when electric vector ∥ to be
∴ dye plane of dye roughly ⊥ to c.

Cell dimensions.

	Space group	a	b	c	β	Vol.
VI	$C2$	70.60	38.99	51.65	103.96°	138,000
II	$P2_1$	30.28	38.39	53.16	105.83°	59,400

Patterson of hol's : superposition. (show slide)
(omit discussion of origin region) & comparison of 0ko's
(copy from paper).

assume screw did remain the same.

layering of ribonuclease $\overline{VI}$: due to a v. strong 200

Also 001 ≠ 0 Thus we can roughly locate molecules in $\overline{VI}$

∴ — — — — $\overline{II}$

Accuracy and Shrinkage (Magdoff + Crick)

Background : variability of ribonuclease lattices.

how large a heavy atom? - depends on accuracy.

Adding dyes - not described here : Accuracy investigation.

found fluctuations in intensity.

z (Studies limited to the hol's)

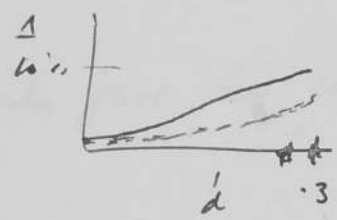
Spectrometer : - check on line profile.

- absorption correction.

General performance appears to be satisfactory.

Two similar crystals (50 : 50 tertiary butyl)

(checked temp. factor the same)



plotted $\Delta = \frac{\Sigma |I - I'|}{\Sigma \bar{I}}$ for groups.

calculated $\sqrt{\frac{4c}{a}}$, allowing for background.

Changes due to solvent : all dimensions.

Fig 3. tertiarybutyl alcohol v ethyl alcohol.

Fig 4. - - v monoacetin.

Fig 5. - - — v dye.

~~Shea~~ Shinkage effect

Study of two adjacent reflections $80\bar{9}$ and $80\bar{10}$

	$80\bar{9}$	$80\bar{10}$
initially	4300	3700
milk cold jet a top of capillary.	$\bar{5}300$	1600

5° drop

time-constant of return ~ 2 minutes.

Small 1°C jet near capillary : depended upon aspect.

in one case could be done by the sustained hear of the hand.

"natural" variation. 2300 3600 3500 2900 3600 2700 counts.

shown for all crystals of n.b.o. II studied.

for $80\bar{6}$ ... $80\bar{10}$: showed that 10°C steady gave less change than 1° local.

Quantitative work : wet v damp. .. Fig 6.

Large jets

cell dimensions		a	c	β
	Wet	30.49	53.20	105.94°
	Damp	30.17	52.96	106.03°
	Δ	·32	·24	·09°

Breadth of reflection

	peak width.	2θ for $30\bar{4}$	count for $80\bar{10}$
normal	·05°	9.680	12,500
slightly shrunk	·06°	9.75°	8,300
shrunk further.	·16°	9.82°	3,500

Patterson : very small near origin.

- changes appear somewhat greater than a "quantum" change.
- mols. may rotate slightly about axis not far from c.

3-1) data : comparison — fig. 7.

Photographic data : given as average.

Lesions (1) measure all dimensions very accurately.

(2) mount crystal wet and/or use large jets.

(3) concern for capillary absorption.

Implications : "dense" corresponds to ~ 30 H_2O or to alcohol } per protein mol.

β-lactoglobulin

Theory (Crick & Hughes)

Preliminary results only:

- 3D v. 2D.?
- How heavy an atom? (define N and n)

Results: P21

$$\frac{\overline{\Delta I}}{\overline{I_p}} \simeq 2\sqrt{\frac{n}{N}}\,\frac{f_H}{f_p}$$ centrosymmetric.

$$\simeq \sqrt{2}\sqrt{\frac{n}{N}}\,\frac{f_H}{f_p}$$ non-centrosymmetric.

$\simeq$ Heavy atom peak

General RMS background

- for near 3D Patt. $\simeq \frac{\sqrt{m_3}\, f_H^2}{(2Nf_p^2 + 2n f_H^2)} \simeq \sqrt{m_3}\,\frac{f_H^2}{2Nf_p^2}$

peak height

RMS background

- for difference 3D Patt. without errors $\simeq \frac{\sqrt{m_3}}{\sqrt{8}}\sqrt{\frac{n}{N}}\,\frac{f_H}{f_p}$

- for 2D $(\Delta F)^2$ Patt. $\simeq \frac{\sqrt{m_2}}{2}\,\frac{1}{\sqrt{Nn}}\,\frac{f_H}{f_p}\cdot\frac{1}{p}$

$\left(\overline{\frac{1}{I}}\, p = \overline{\text{error}}\right)$

Ratio of last two cases: $\frac{\text{3D centric}}{\text{2D centric}} = \sqrt{\frac{m_3}{2m_2}}\cdot p.$