[20220114] SK-B2RDEM - Röntgendiffractie en EM - 1H - UITHOF

Cursus: BETA-SK-B2RDEM Röntgendiffractie en electronenmicroscoop (SK-B2RDEM)

Tijdsduur:	2 uur en 30 minuten
Aantal vragen:	18
Gegenereerd op:	24 jan. 2022

Inhoud: Pagina's:

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Cursus: Röntgendiffractie en electronenmicroscoop (SK-B2RDEM)

Deeltentamen Protein Crystallography and Electronmicroscopy – cursus SK-B2RDEM
vrijdag 14 januari 2022

BELANGRIJK
- **Leg uw identiteitsbewijs (met foto) op tafel**
- **Antwoorden mogen in Engels of Nederlands gegeven worden.**
- **Tijd voor PX+EM samen is 2½ uur.**
- **Het maximum aantal punten is 45 voor zowel PX als EM, dus in totaal 90 punten.**
- **Beide onderdelen wegen even zwaar in het deeltentamen (50:50).**
- **Vragen 1 t/m 12 gaan over het PX deel, vragen 13 t/m 18 over het EM deel**

For the cryo-EM questions you may use the below formulas if needed

Formulas Cryo-EM

1. Nyquist frequency: $Ny = \frac{1}{2d}$ (*d*: pixel size on specimen level)
2. Signal-to-noise ratio: $SNR = \frac{\sum_x s(x)^2}{\sum_x n(x)^2}$
   s: (mean-free) signal; *n*: (mean-free) noise; *x*: pixel index.
3. Contrast transfer function:
   $$CTF(q) = K_S(q;\ \Delta z) \cdot K_t(q) \cdot \sin\left(\frac{\pi}{2}(C_S\lambda^3 q^4 - 2\lambda\Delta z \cdot q^2) + \Delta\varphi\right)$$
   q: spatial frequency; Δz: defocus; λ: wavelength; C_S: spherical aberration coefficient; $\Delta\varphi$: phase shift (if present); K_s and K_t: (Gaussian) envelopes caused by spatial and temporal incoherence of the electrons
4. Wiener Filter: $I'(q) = \frac{CTF(q)I_i(q)}{|CTF(q)|^2 + f(q)}$
   $I'(q)$: Fourier transformed of filtered image; $I_i(q)$: Fourier transformed of input image; $f(q)$: Wiener constant.
5. CTF reconstruction from different images: $I'(q) = \sum_{i=1}^{N} \frac{CTF_i(q)I_i(q)}{\sum_j^N |CTF_j(q)|^2 + f(q)}$
   $I_i(q)$: Fourier transformed of i^{th} input image; $CTF_i(q)$: CTF of i^{th} input image.
6. Crowther frequency: $r_{max} = (\Delta\alpha \cdot D)^{-1}$
   $\Delta\alpha$: angular increment between adjacent orientations; *D*: diameter of object.
7. Squared deviation of two images: $d = \sum_x (f_1(x) - f_2(x))^2$
   f_1(x): image 1; f_2(x): image 2; *x*: pixel index.
8. Cross-correlation coefficient: $CC = \frac{\sum_x f_1(x)f_2(x)}{\sqrt{\sum f_1(x)^2 \sum f_2(x)^2}}$
   The mean value of f_1(x) and f_2(x) is assumed to be zero.
9. Cross-correlation function: $CC(\Delta x, \Delta y, \Delta\varphi) = \frac{\sum_x f_1(x,y)T(\Delta x,\Delta y,\Delta\varphi)(f_2(x,y))}{\sqrt{\sum f_1(x,y)^2 \sum f_2(x,y)^2}}$
   $T(\Delta x, \Delta y, \Delta\varphi)$: Transformation of image (translation by $(\Delta x, \Delta y)$ rotation by $\Delta\varphi$).
10. Fourier Shell Correlation: $FSC(i) = \frac{\sum_{q \in i} f_1(q)f_2(q)}{\sqrt{\sum_{q \in i} f_1(q)^2 \sum_{\in i} f_1(q)^2}}$
    i: i^{th} shell in Fourier space; $q \in i$*:* frequencies located in shell *i*; f_1(q): Fourier transform of image 1; f_2(q): Fourier transform of image 2.
11. Relationship between imaged spatial frequency and scattering angle: $q \approx \frac{2\pi}{\lambda}\vartheta$
    q: (amount) of spatial frequency; λ: wavelength; ϑ: scattering angle / aperture

Aantal vragen: 18

In totaal zijn 90 punten voor deze toets te behalen, 49,58 punten zijn nodig om voor de toets te slagen.

1
2 pt.

In the methods section of a publication the following is mentioned: “Crystals of E. coli OmpF protein were obtained at 20 °C in a hanging drop with the reservoir solution, 100 mM sodium cacodylate, 200 mM MgCl2, 50% (w/v) PEG 200, pH 6.5 mixed in a 1:1 ratio with 8 mg/ml protein solution.”

Which compound in the reservoir solution is the "precipitant"? (2 points)

a. PEG 200

b. MgCl2

c. protein solution

d. sodium cacodylate

e. all of the above

f. none of the above

2
3 pt.

Describe the hanging drop method used for crystallization. Include in your answer how the droplet containing the protein changes over time to make it crystallize. (3 points)

3
2 pt.

What is the purpose of cryo-cooling crystals? (2 points)

4
4 pt.

Describe two different ways to modify a protein to improve the chance to obtain crystals of this modified protein. (4 points)

5
2 pt.

Consider an X-ray diffraction experiment performed on a protein cystal.

How many times does a reflection at maximum pass through the Ewald sphere during a 180° rotation? Ignore the possible presence of equivalent reflections for your answer. (2 points)

a. 0 times

b. 1 time

c. 2 times

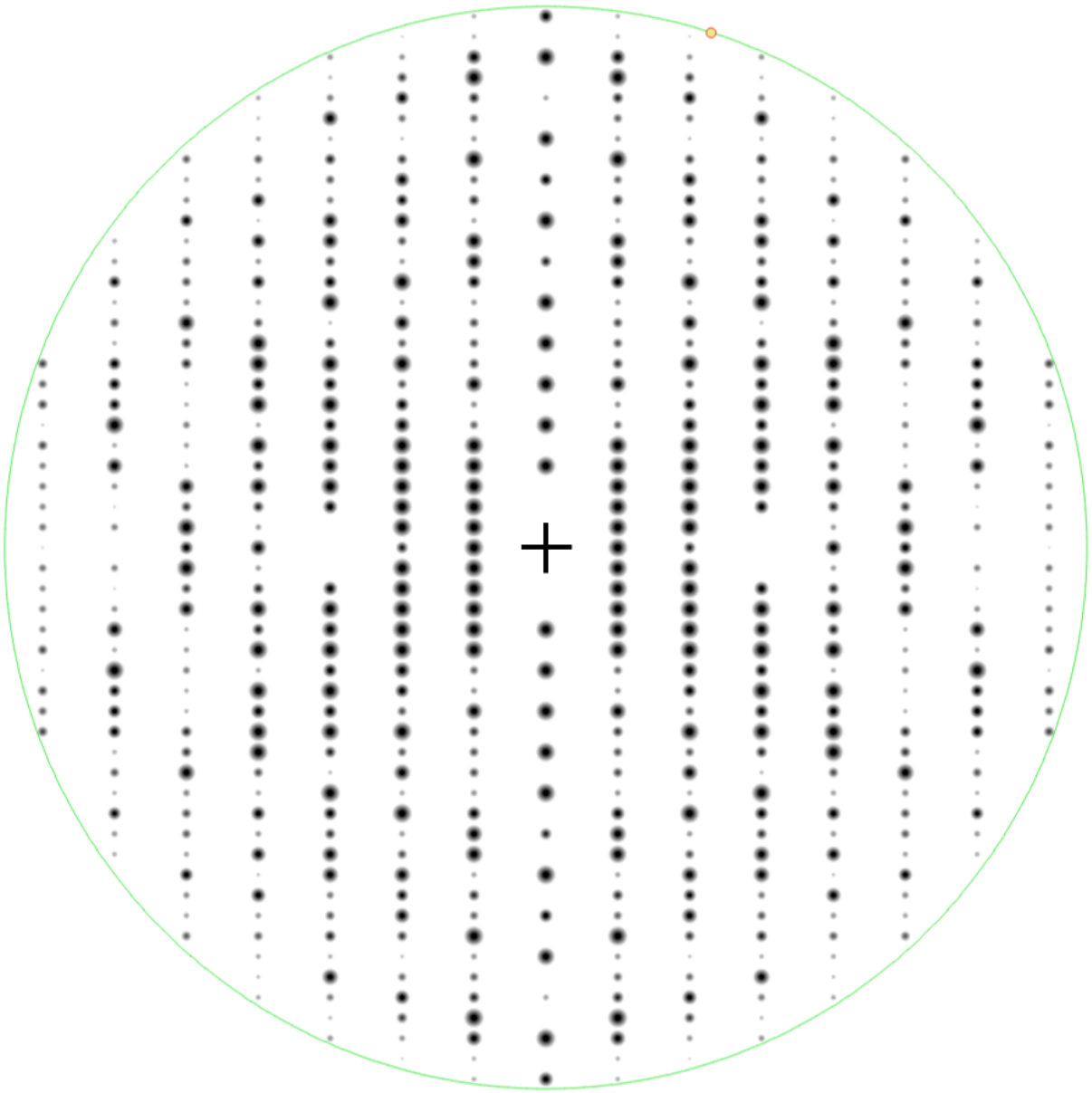
d. 3 times

e. 4 times

f. more than 4 times

6

3 pt.



In the diffraction data processing of a protein crystal, you observe the above pattern for the *(hk0)* data. The central cross indicates the 0 0 0. Which spacegroup is the most likely for this pattern? (3 points)

a. *P*4

b. *P*422

c. $P4_1$

d. $P2_1$

e. $P4_12_12_1$

f. *P*1

g. *P*222

h. *P*2

i. *P*6

j. *P*3

k. $P6_1$

l. $P2_12_12_1$

7 A diffraction dataset was collected with the following statistics.
2 pt.

	$Nrp1_{1-4}$
Data collection	
Space group	*P*321
Cell dimensions	
a, *b*, *c* (Å)	245.4, 245.4, 47.9
α, β, γ (°)	90, 90, 120
Resolution (Å)	81-2.7 (2.79-2.70)
R_{merge} (%)	10.8 (66.0)
$I/\sigma I$	8.4 (1.6)
Completeness (%)	99.4 (99.6)
Redundancy	5.0 (4.5)

What is het most likely explanation for the low-resolution cutoff? (2 points)

a. Detector too close to the crystal.

b. Detector too far away from the crystal.

c. Quality of the crystal insufficient.

d. The presence of a beamstop.

e. None of the options given.

8 Assume you have a native protein data set (FP) and two different derivative data sets (FPH1 and FPH2). Describe in a logical order how you can derive phase information for the structure factors of the native protein data set FP. (5 points)
5 pt.

9
3 pt.

A heavy atom in a crystal form with a two-fold rotation axis along y gives rise to a peak in a Patterson map at position $u = 0.8$, $v = 0$, $w = 0.28$.

What is a possible position of the heavy atom for z? (3 points)

a. 0.8

b. 0.14

c. 0.4

d. -0.4

e. 0.28

f. 0

g. 1.6

h. 0.56

i. None of the above.

10
4 pt.

How would you approach modelling of a protein-crystal structure with diffraction data up to 2.5 Å resolution *versus* a case with diffraction data up to 1 Å resolution. (4 points)

11 Two datasets have been collected on two crystals of the same protein called PirB. The structures have been refined using these datasets. The statistics of the datasets and the structures are given in the table below.

	PirB (A)	PirB (B)
Data collection		
Space group	$P2_1$	$P2_1$
Cell dimensions		
a, *b*, *c* (Å)	54.7 185.3 99.1	67.3 127.1 144.1
α, β, γ (°)	90 105.6 90	90 103.4 90
Resolution (Å)	52.7-3.4 (3.6-3.4)	70.1-4.5 (4.7-4.5)
No. reflections	25,778 (4,683)	13,858 (1,273)
R_{merge}	0.108 (0.824)	0.226 (0.907)
$I / \sigma I$	6.9 (1.8)	4.0 (1.6)
Completeness (%)	98.8 (99.2)	98.9 (98.0)
Redundancy	3.2 (3.2)	3.5 (3.5)
$CC_{1/2}$	0.949 (0.586)	0.960 (0.594)
Refinement		
# PirB in asymmetric unit	2	2
R_{work} / R_{free}	0.256 / 0.307	0.313 / 0.339
No. non-H atoms	9167	9264
B-factors (Å^2)	139	244
R.m.s. deviations		
Bond lengths (Å)	0.010	0.003
Bond angles (°)	0.82	0.65
Ramachandran most favored (%)	95.75	94.45
Ramachandran outliers (%)	0.52	0.62

3 pt. **a.** Explain why the R_{free} is always higher than the R_{work}. (3 points)

3 pt. **b.** Explain why the r.m.s. deviation (rmsd) of the bond lengts is higher for the PirB (A) structure compared to the PirB (B) structure. (3 points)

3 pt. **c.** Which of the two datasets, PirB (A) or PirB (B), has provided a more accurate structure? Explain your choice. (3 points)

12 Consider the following equation:

$$R_{\text{work}} = \frac{\sum_{hkl} \left| \left| F_{\text{obs}}(hkl) \right| - k \left| F_{\text{calc}}(hkl) \right| \right|}{\sum_{hkl} \left| F_{\text{obs}}(hkl) \right|}$$

A structure was refined against a diffraction dataset. Structure factor amplitudes derived from the diffraction data are:

Structure factor amplitude	(*h k l*)
9	(1 6 3)
40	(5 2 4)
83	(6 4 2)
29	(3 9 4)

Structure factor amplitudes derived from the refined model are:

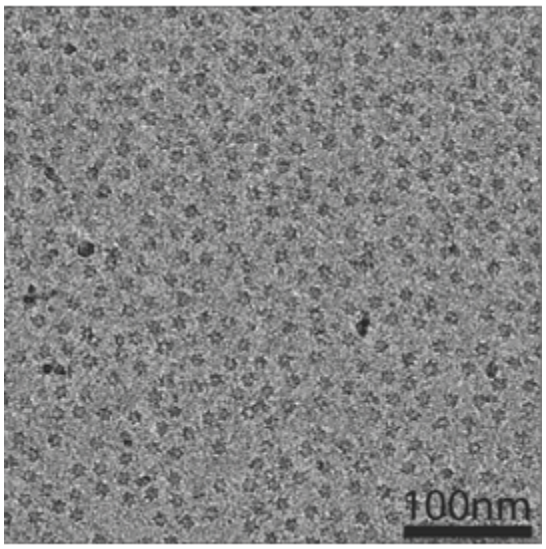
Structure factor amplitude	(*h k l*)
11	(3 9 4)
9	(2 5 7)
27	(6 4 2)
14	(5 2 4)
4	(1 6 3)

2 pt. **a.** What is the purpose of k (indicated by the arrow) in this equation? (2 points)

4 pt. **b.** Calculate the R_{work} for this refined structure. Important: assume that **k equals 3**. (4 points)

13 Does the below images depict frozen-hydrated (‘**cryo’**) samples or uranyl-acetate stained material? (6 P total)

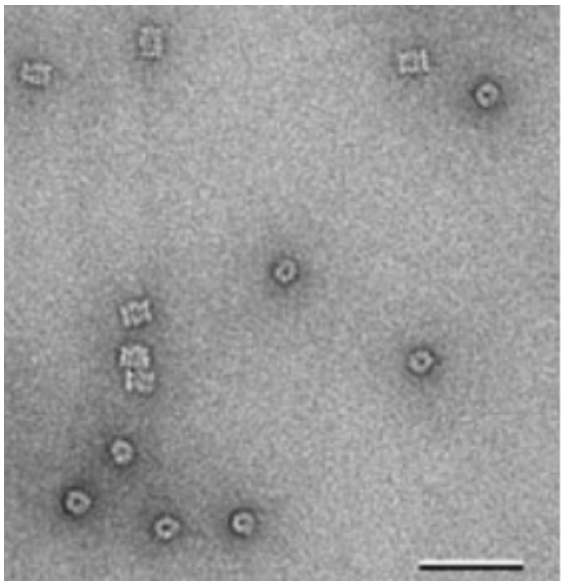
1 pt. **a.** Does this image depict frozen-hydrated (‘**cryo’**) sample or uranyl-acetate stained material? (1 P)



a. Cryo

b. Negative stain

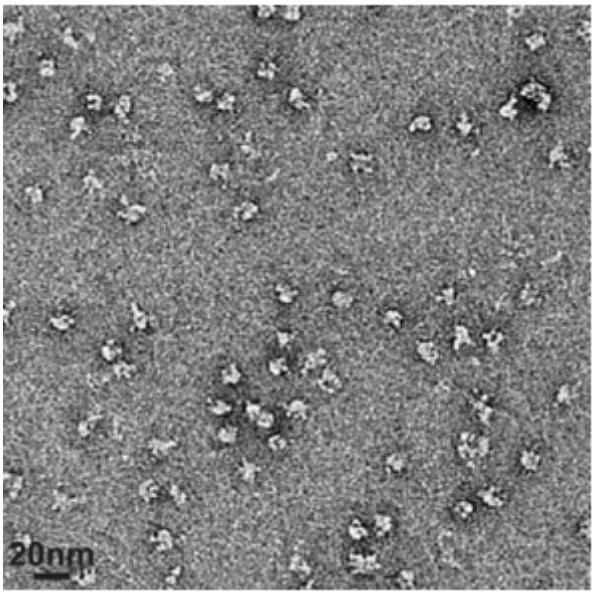
1 pt. **b.** Does this image depict frozen-hydrated (‘**cryo’**) sample or uranyl-acetate stained material? (1 P)



a. Cryo

b. Negative stain

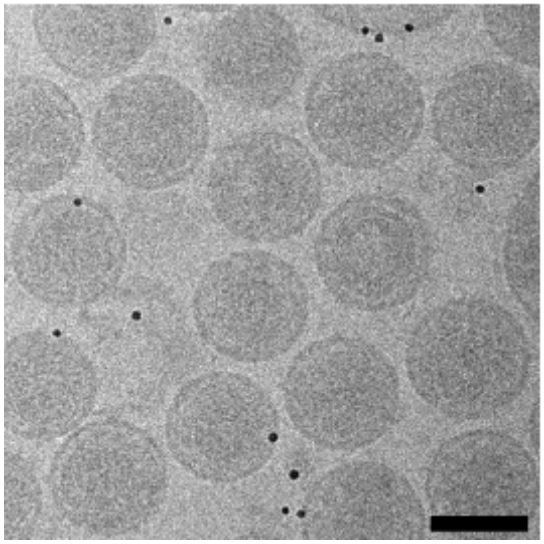
1 pt. **c.** Does this image depict frozen-hydrated (‘**cryo’**) sample or uranyl-acetate stained material? (1 P)



a. Cryo

b. Negative stain

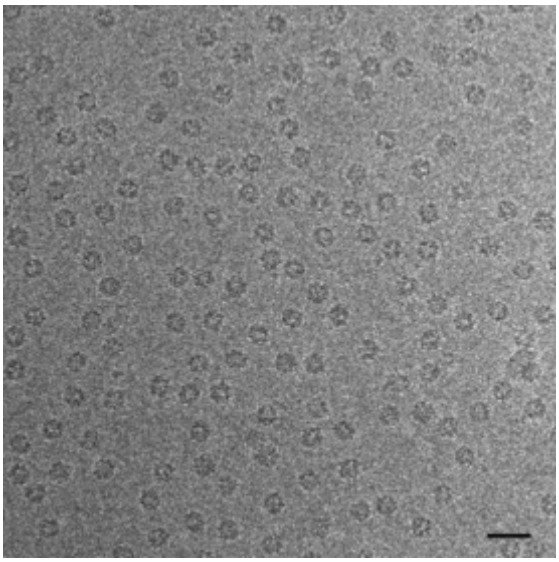
1 pt. **d.** Does this image depict frozen-hydrated (‘**cryo’**) sample or uranyl-acetate stained material? (1 P)



a. Cryo

b. Negative stain

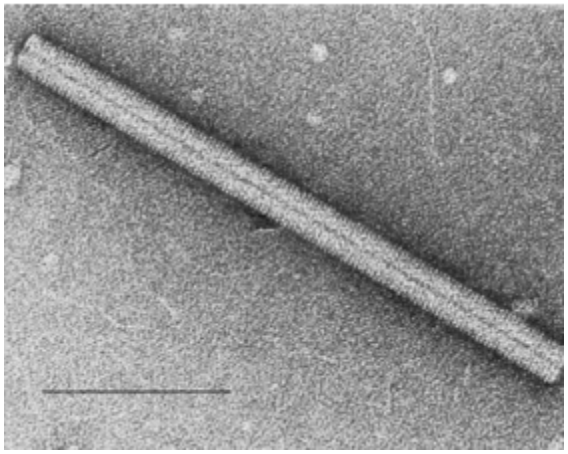
1 pt. **e.** Does this image depict frozen-hydrated (‘**cryo’**) sample or uranyl-acetate stained material? (1 P)



a. Cryo

b. Negative stain

1 pt. **f.** Does this image depict frozen-hydrated (‘**cryo’**) sample or uranyl-acetate stained material? (1 P)



a. Cryo

b. Negative stain

14 A researcher acquires images of frozen hydrated samples with a transmission electron microscope (TEM) equipped with a direct detector. The detector has 4,000 x 4,000 pixels and the physical size of a pixel is 10 µm x 10 µm The TEM is operated at a magnification of 50,000 x (1 m in the specimen plane corresponds to 50,000 m in the image plane). To acquire an image the sample is exposed to a dose of 30 electrons per $Å^2$ distributed over 10 s. (total: 8 P)

2 pt. **a.** Which area does each pixel correspond to on the specimen level? (2 P)

a. 5 m^2

b. 4 $Å^2$

c. 2 $Å^2$

d. 25 m^2

1 pt. **b.** Which minimal distance can possibly be resolved? (1 P)

a. 4 Å

b. 20 µm

c. 10 m

d. 1 Å

e. 2 Å

f. 0.5 Å

1 pt. **c.** What is the corresponding Nyquist frequency of the imaging conditions? (1 P)

a. 2 $Å^{-1}$

b. 0.1 m^{-1}

c. 0.5 $Å^{-1}$

d. 0.25 $Å^{-1}$

e. 0.1 µm

f. 1 $Å^{-1}$

2 pt. **d.** Which total area of the specimen is imaged (e.g., in μm^2)? (2 P)

a. 64 μm^2

b. 16 μm^2

c. 4 μm^2

d. 0.64 μm^2

e. 0.04 μm^2

f. 0.16 μm^2

2 pt. **e.** What is the electron dose rate a pixel is exposed to (measured in electrons per pixel per second; e/p/s) (2 P)

a. *12 e/p/s*

b. 0.75 e/p/s

c. 0.1 e/p/s

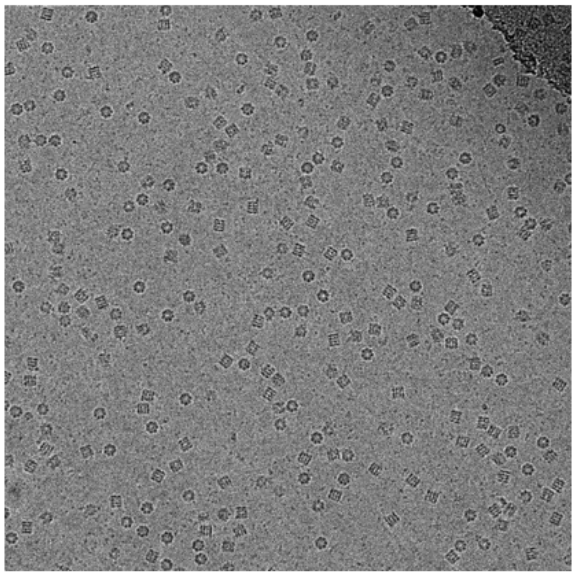
d. 3 e/p/s

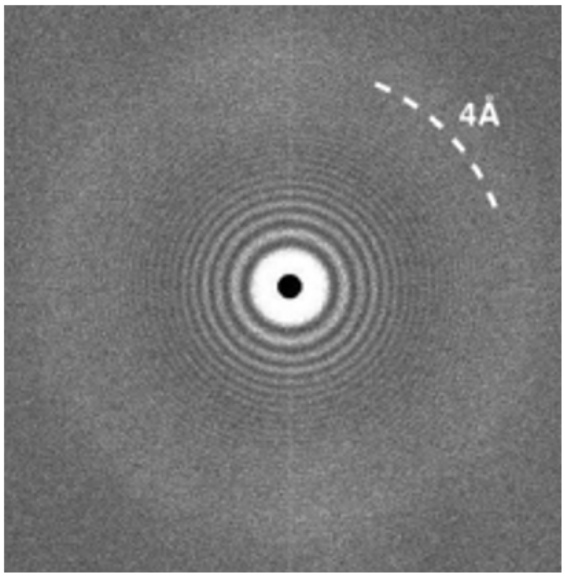
e. 30 e/p/s

f. 300 e/p/s

15 A researcher wants to determine the structure of a cylinder-shaped Hsp60 protein complex by *cryo-EM single particle analysis*. (20 points total)

3 pt. **a.** He/she collects 1,000 electron micrographs. One of those micrographs is shown below on the left. The amplitudes of its Fourier transform are shown on the right.





From the total dataset, 200 images are discarded because in the amplitudes of their Fourier transforms *rings* are not discernable beyond 1/4 $Å^{-1}$ resolution. Please explain in 1-2 sentences how you can determine the defocus of the image from these *'Thon' rings*. (3 P)

2 pt. **b.** In the remaining images 20,000 putative Hsp60 particles are localized. To remove false positives, these candidates are subjected to two-dimensional classification, reducing the number of trustworthy particles to 10,000. Which one of the below sentences describes the concept of this 2D classification? (2 P)

a. 2-D images are grouped according to 2-D projections of a 3-D density along different orientations

b. 2-D images are grouped according to their pairwise similarity

c. 2-D images are grouped according to their similarity to 2-D projections of different 3D densities

d. 2-D images are grouped according to their similarity to 2-D projections of a cylinder

e. 3-D densities are grouped according to the similarity of their 2-D projections

2 pt. **c.** The imaged Hsp60 complex has 7-fold rotational symmetry axis along the cylinder axis and 2-fold rotational symmetry perpendicular (see below).

How many symmetry-equivalent orientations does each particle have? (2 P)

a. 4

b. 1

c. 14

d. 2

e. 7

f. 49

1 pt. **d.** How many non-symmetrical particles would be required approximately to achieve a reconstruction with comparable resolution (compared to above 10,000 symmetrical particles)? (1 P)

a. 20000

b. 715

c. 1430

d. 10000

e. 140000

f. 70000

3 pt. **e.** Using extensive computation, the particle images are assigned to different views of an underlying three-dimensional (3D) density. How does the resolution r_{max} that can possibly be obtained from these views depend on the diameter of the molecule D and angular sampling of the orientations Δa (i.e., angular difference in rad between two adjacent views)? (3 P)

2 pt. **f.** The Hsp60 complex is approximately 12 nm in diameter and Δa is 2°(=1/30 rad). Which resolution r_{max} can be obtained in this case? (2 P)

a. 0.04 nm^{-1}

b. 2.5 $Å^{-1}$

c. 0.4 nm^{-1}

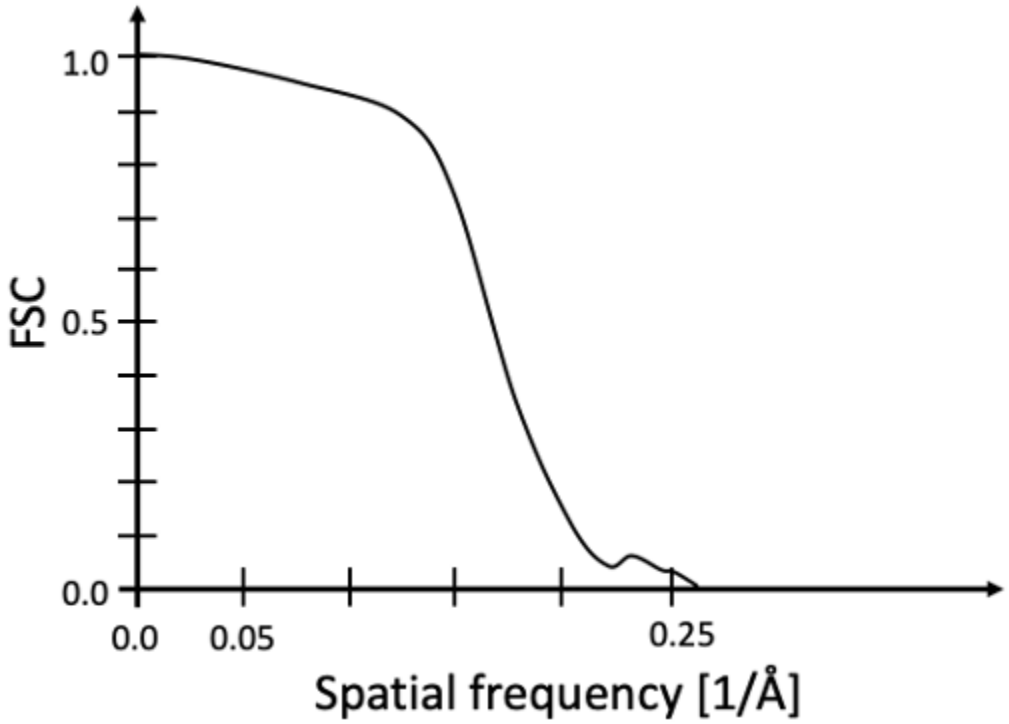
d. 2 $Å^{-1}$

e. 2.5 nm^{-1}

f. 0.4 $Å^{-1}$

2 pt. **g.** The resolution of the single particle reconstruction is measured by Fourier Shell Correlation (FSC). Please describe briefly, which densities are compared for this resolution measurement. (2 P)

2 pt. **h.** Which resolution do you determine from below FSC plot? (2 P)



a. 0.05 $Å^{-1}$

b. 0.1 $Å^{-1}$

c. 0.15 $Å^{-1}$

d. 0.2 $Å^{-1}$

e. 0.25 $Å^{-1}$

f. 0.3 $Å^{-1}$

2 pt. **i.** The resolution determined by FSC may be smaller than the r_{max} determined by Δα and diameter. Please provide two or more possible reasons. (2 P)

1 pt. **j.** Prior to computation of the FSC the densities have been masked. Please explain the effect of masking on the resolution. (1 P)

16 A spherical protein has a molecular mass of 300 kDa. The average density of a protein is =1.37 kg/l = 1.37 kg / (10^{-3} m^3) = 1.37 10^6 g/m^3.

4 pt. **a.** Please estimate the volume of the above protein. (1 g is approximately 6×10^{23} Da). (4 P)

- **a.** 220 μm^3
- **b.** 220 nm^3
- **c.** 822 nm^3
- **d.** 365 nm^3
- **e.** 0.365 $Å^3$
- **f.** 8.22 nm^3

2 pt. **b.** What is the approximate radius of this spherical protein? (2 P)

- **a.** 5.2 μm
- **b.** 4.4 nm
- **c.** 10.3 nm
- **d.** 3.3 Å
- **e.** 6.5 nm

17 A three-dimensional density can be reconstructed from its projection according to the Projection-Slice
2 pt. theorem. (2 P total)

The theorem says:

- **a.** The projection equals the slice of the Fourier transform of the 3D object perpendicular to the direction of the projection direction.
- **b.** The given projection equals the slice through the 3D object perpendicular to the direction of the projection direction.
- **c.** The Fourier transform of given projection equals the Fourier transform of the slice of the 3D object perpendicular to the direction of the projection direction.
- **d.** The Fourier transform of given projection equals the slice of the Fourier transform of the 3D object perpendicular to the direction of the projection direction.
- **e.** The given projection equals the Fourier transform of the slice through the 3D object perpendicular to the projection direction.
- **f.** The slice of given projection perpendicular to the projection direction equals the 3D object .

18 The Ewald sphere is a geometric construction in reciprocal space, which is helpful in electron
3 pt. microscopy and X-ray diffraction. The Ewald sphere has a radius r_E, which is determined by energy and wavelength of the incoming electron (or X-ray). (3 P total)

Which of the following statements are true: (3 P)

a. r_E increases with increasing energy of the electron.

b. r_E decreases with increasing energy of the electron.

c. r_E increases with increasing wavelength of the electron.

d. r_E decreases with increasing wavelength of the electron.

e. r_E increases with increasing velocity of the electron.

f. r_E decreases with increasing speed of the electron.

Lever kladpapier, als je dat gebruikt hebt, in bij de examinator.

Naam:				Handtekening:
Datum:	- -	Geboortedatum:	- -	

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Deeltentamen Protein Crystallography and Electronmicroscopy – cursus SK-B2RDEM
vrijdag 14 januari 2022

BELANGRIJK

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2. Signal-to-noise ratio: $SNR = \frac{\sum_x s(x)^2}{\sum_x n(x)^2}$
   s: (mean-free) signal; n: (mean-free) noise; x: pixel index.
3. Contrast transfer function:
   $$CTF(q) = K_S(q;\ \Delta z) \cdot K_t(q) \cdot \sin\left(\frac{\pi}{2}(C_S\lambda^3 q^4 - 2\lambda\Delta z \cdot q^2) + \Delta\varphi\right)$$
   q: spatial frequency; Δz: defocus; λ: wavelength; C_S: spherical aberration coefficient; $\Delta\varphi$: phase shift (if present); K_s and K_t: (Gaussian) envelopes caused by spatial and temporal incoherence of the electrons
4. Wiener Filter: $I'(q) = \frac{CTF(q)I_i(q)}{|CTF(q)|^2 + f(q)}$
   $I'(q)$: Fourier transformed of filtered image; $I_i(q)$: Fourier transformed of input image; $f(q)$: Wiener constant.
5. CTF reconstruction from different images: $I'(q) = \sum_{i=1}^{N} \frac{CTF_i(q)I_i(q)}{\sum_j^N |CTF_j(q)|^2 + f(q)}$
   $I_i(q)$: Fourier transformed of i^{th} input image; $CTF_i(q)$: CTF of i^{th} input image.
6. Crowther frequency: $r_{max} = (\Delta\alpha \cdot D)^{-1}$
   $\Delta\alpha$: angular increment between adjacent orientations; D: diameter of object.
7. Squared deviation of two images: $d = \sum_x (f_1(x) - f_2(x))^2$
   f_1(x): image 1; f_2(x): image 2; x: pixel index.
8. Cross-correlation coefficient: $CC = \frac{\sum_x f_1(x)f_2(x)}{\sqrt{\sum f_1(x)^2 \sum f_2(x)^2}}$
   The mean value of f_1(x) and f_2(x) is assumed to be zero.
9. Cross-correlation function: $CC(\Delta x, \Delta y, \Delta\varphi) = \frac{\sum_x f_1(x,y)T(\Delta x,\Delta y,\Delta\varphi)(f_2(x,y))}{\sqrt{\sum f_1(x,y)^2 \sum f_2(x,y)^2}}$
   $T(\Delta x, \Delta y, \Delta\varphi)$: Transformation of image (translation by $(\Delta x, \Delta y)$ rotation by $\Delta\varphi$).
10. Fourier Shell Correlation: $FSC(i) = \frac{\sum_{q \in i} f_1(q)f_2(q)}{\sqrt{\sum_{q \in i} f_1(q)^2 \sum_{\in i} f_1(q)^2}}$
    i: i^{th} shell in Fourier space; $q \in i$: frequencies located in shell i; f_1(q): Fourier transform of image 1; f_2(q): Fourier transform of image 2.
11. Relationship between imaged spatial frequency and scattering angle: $q \approx \frac{2\pi}{\lambda}\vartheta$
    q: (amount) of spatial frequency; λ: wavelength; ϑ: scattering angle / aperture

1 2 pt.

A	B	C	D	E	F
○	○	○	○	○	○

2
3 pt.

Antwoord:

3
2 pt.

Antwoord:

4
4 pt.

Antwoord:

5
2 pt.

A B C D E F

6
3 pt.

A B C D E F G H I J K L

7
2 pt.

A B C D E

8
5 pt.

Antwoord:

2

4

6

8

9
3 pt.

A B C D E F G H I

10
4 pt.

Antwoord:

2

4

6

8

11
9 pt.

a.

Antwoord:

b.

Antwoord:

c.

Antwoord:

12
6 pt.

a.

Antwoord:

b.

Antwoord:

13
6 pt.

a. A ○ B ○

b. A ○ B ○

c. A ○ B ○

d. A ○ B ○

e. A ○ B ○

f. A ○ B ○

14
8 pt.

	A	B	C	D	E	F
a.	○	○	○	○		
b.	○	○	○	○	○	○
c.	○	○	○	○	○	○
d.	○	○	○	○	○	○
e.	○	○	○	○	○	○

15
20 pt.

a.

Antwoord:

2

4

6

8

b. A B C D E

c. A B C D E F

d. A B C D E F

e.

Antwoord:

2

f. A B C D E F

g.

Antwoord:

2

4

h. A B C D E F

i. Antwoord:

2

4

j. Antwoord:

2

16 6 pt.

a. A ○ B ○ C ○ D ○ E ○ F ○

b. A ○ B ○ C ○ D ○ E ○

17 2 pt. A ○ B ○ C ○ D ○ E ○ F ○

18 3 pt. A ☐ B ☐ C ☐ D ☐ E ☐ F ☐

Er zijn meerdere antwoorden mogelijk

Lever kladpapier, als je dat gebruikt hebt, in bij de examinator.

Correctiemodel

1. A
2 pt.

2.
3 pt.

Beoordelingscriterium	Punten
During equilibration the drop containing the protein will shrink. The protein concentration will double in time. (If crystals appear the concentration of protein in solution will go down.)	3 punten
Totaal aantal punten:	*3 punten*

3.
2 pt.

Beoordelingscriterium	Punten
Vermindering stralingsschade. Of: Betere data opmeten (Atomen hebben kleinere amplitude)	2 punten
Totaal aantal punten:	*2 punten*

4.
4 pt.

Beoordelingscriterium	Punten
Criterium 1	4 punten
Totaal aantal punten:	*4 punten*

2 points for each (max of 4 points)

- Mutagenesis to remove amino acids that are predicted to form floppy loop regions or protein termini.
- Limited proteolysis to remove floppy segments in loop regions or at the protein termini.
- Removal of glycans by mutagenesis
- Enzymatic removal of glycans
- lysine methylation

5. C
2 pt.

6. D
3 pt.

7. D
2 pt.

8.
5 pt.

Beoordelingscriterium	Punten
Positions of heavy atoms are determined from a patterson map. The peaks in the Patterson map indicate interatomic vectors coming from the heavy atoms. When we have obtained the positions of the heavy atoms we can calculate the complex structure factors (for each hkl) for the heavy atom contribution with the formula for the atomic structure factor. Using the harker construction (circles) with vectors for the heavy atom contribution and circles for the reflection with only amplitude information the vector for the native protein can be otained at the intersection of circles. Two heavy atom derivatives are required to solve the phase ambiguity that exsists with one heavy atom derivatie only. The phase for the native dataset and for each reflection is the angle that the vector makes.	5 punten
Totaal aantal punten:	*5 punten*

9. B
3 pt.

10.
4 pt.

Beoordelingscriterium	Punten
Less reflection (less elements h); thus less x-ray information. At 3 Ang less data available (less information, less data ‘restraints’); minimize degrees of freedom. For example, use rigid domain instead of atoms At 1 Ang, data is sufficient, less stereochemical restraints or constraints are needed.	4 punten
Totaal aantal punten:	*4 punten*

11.
9 pt.

a.

Beoordelingscriterium	**Punten**
Rwork is calculated using reflections that were used to refine the structure. Rree reflections have not been used in the refinement. Hence the structure will fit better to the Rwork set.	3 punten
Totaal aantal punten:	*3 punten*

b.

Beoordelingscriterium	**Punten**
the PirB (A) dataset is to a higher resolution (=more diffraction data). That means compared to the lower PirB (B) dataset less emphasis (weight) was required on the geometrical terms (or more weight on the diffraction terms) resulting in a slightly higher rmsd for the bond lengts.	3 punten
Totaal aantal punten:	*3 punten*

c.

Beoordelingscriterium	**Punten**
PirB (A). This dataset is to a higher resolution (more diffraction data). (Anything else is not relevant.)	3 punten
Totaal aantal punten:	*3 punten*

12.
6 pt.

a.

Beoordelingscriterium	**Punten**
To get the calculated structure factors on the same scale as the measured structure factors.	2 punten
Totaal aantal punten:	*2 punten*

b.

Beoordelingscriterium	**Punten**
Sommatie Fobs is 9+40+83+29 = 161 Sommatie verschil gemeten – berekend: (|9-3*4|)+(|40-3*14|)+(|83-3*27|)+(|29-3*11|)=3+2+2+4=11 Rwork is 11/161=0.068 If the scale factor is forgotten (3 points): Sommatie verschil gemeten – berekend: (|9-4|)+(|40-14|)+(|83-27|)+(|29-11|)=5+26+56+18=105 Rwork is 105/161=0.652 (3 points)	4 punten
Totaal aantal punten:	*4 punten*

13. 6 pt.

a. 1 pt. A

b. 1 pt. B

c. 1 pt. B

d. 1 pt. A

e. 1 pt. A

f. 1 pt. B

14. 8 pt.

a. 2 pt. B

b. 1 pt. A

c. 1 pt. D

d. 2 pt. D

e. 2 pt. A

15.
20 pt.

a.

Correction criterion	Points
Contrast transfer function	1 point
oscillating behaviour	1 point
Fit to CTF / nodes of CTF reveal defocus value	1 point
Total points:	*3 points*

b. 2 pt. B

c. 2 pt. C

d. 1 pt. E

e.

Correction criterion	Points
Crowther criterion	1 point
r_max = 1/(D x dalpha)	2 points
Total points:	*3 points*

f. 2 pt. E

g.

Correction criterion	Points
split data 50:50	1 point
reconstruct independently	1 point
Total points:	*2 points*

h. 2 pt. D

i.

Correction criterion	Points
Particle number	1 point
heterogeneity of classes	1 point
Total points:	*2 points*

j.

Correction criterion	Points
Resolution increases because surrounding noise is removed	1 point
Total points:	*1 point*

16. 6 pt.

a. 4 pt. D

b. 2 pt. B

17. D
2 pt.

18. A
3 pt. D
E

Cesuur

Toegepaste raadscore: 9,164 pt

Behaalde punten	Cijfer
90	10
89	9,9
88	9,8
87	9,7
86	9,6
85	9,4
84	9,3
83	9,2
82	9,1
81	9,0
80	8,9
79	8,8
78	8,7
77	8,6
76	8,4
75	8,3
74	8,2
73	8,1
72	8,0
71	7,9
70	7,8
69	7,7
68	7,6
67	7,4
66	7,3
65	7,2
64	7,1
63	7,0
62	6,9
61	6,8
60	6,7
59	6,5
58	6,4
57	6,3
56	6,2
55	6,1
54	6,0
53	5,9
52	5,8
51	5,7
50	5,5
49	5,4
48	5,3
47	5,2
46	5,1
45	5,0
44	4,9
43	4,8
42	4,7
41	4,5
40	4,4
39	4,3
38	4,2
37	4,1
36	4,0
35	3,9
34	3,8
33	3,7
32	3,5

31	3,4
30	3,3
29	3,2
28	3,1
27	3,0
26	2,9
25	2,8
24	2,7
23	2,5
22	2,4
21	2,3
20	2,2
19	2,1
18	2,0
17	1,9
16	1,8
15	1,6
14	1,5
13	1,4
12	1,3
11	1,2
10	1,1
9	1,0
8	1,0
7	1,0
6	1,0
5	1,0
4	1,0
3	1,0
2	1,0
1	1,0
0	1,0

Vraag-identificatiecodes

Deze identifiers kunnen worden gebruikt om de precieze vraag in de vragenbanken te identificeren. Gebruik deze code in combinatie met de documentcode wanneer u feedback doorgeeft, zodat precies duidelijk is op welke vraag en -versie uw feedback van toepassing is.

Documentidentificatiecode: 40857-69853

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3	310714	f8fdf3ee-aa0b-c9a4-10f2-a5b2c6104279
4	310719	60e4cd69-3b5d-ef10-4dc6-52456a385f64
5	338670	f413ae48-70f1-ffc1-6ae1-a549150aa967
6	313434	162e4de3-02ad-6f31-118c-c28ede40f94b
7	313429	72a52bdd-7d67-70a7-acaa-835623cd1fa9
8	338890	3bc8251b-102a-5ea6-14bd-f8019fcc8939
9	486833	5b4544f6-b423-8806-1b7f-cb3e431eb385
10	313509	abab2a5a-99a9-1a6d-2234-9dea76768661
11	487098	6e53c897-bed3-72f5-5833-99548be47472
12	338880	6dfe5ff1-6742-9322-8d7f-2db99ef3cb0d
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16	486718	b4694453-45e7-c53c-f1cb-88fb43abc9fd
17	312894	b11fe61d-b0b0-5bbf-fdf5-44bbcd2ec27f
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