

Supplementary Information

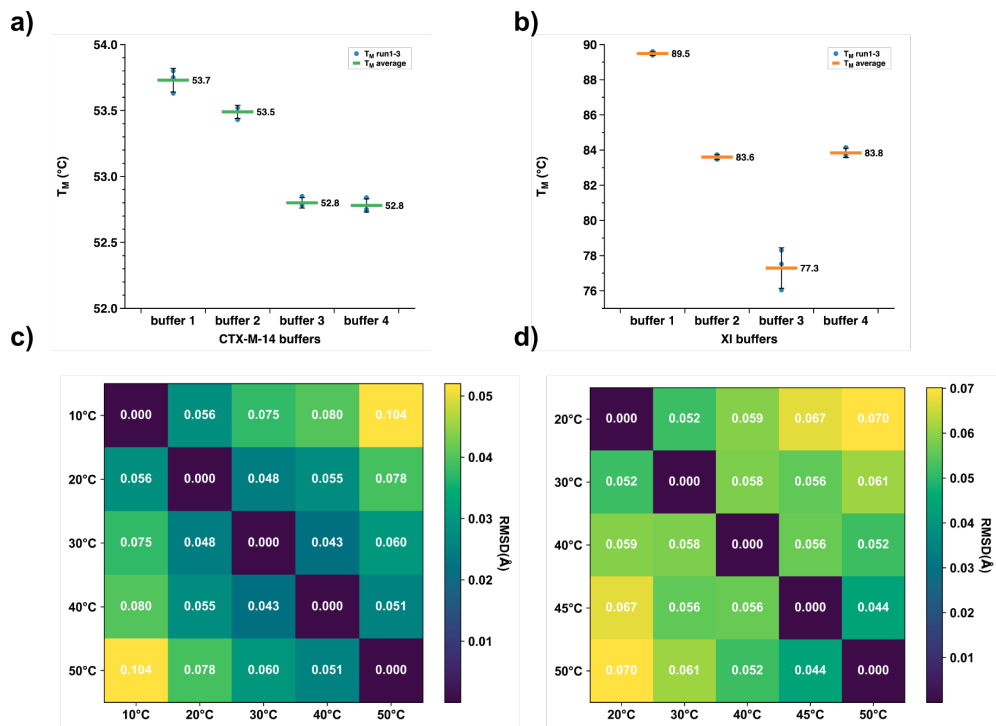
Probing the modulation of enzyme kinetics by multi-temperature, time-resolved serial crystallography

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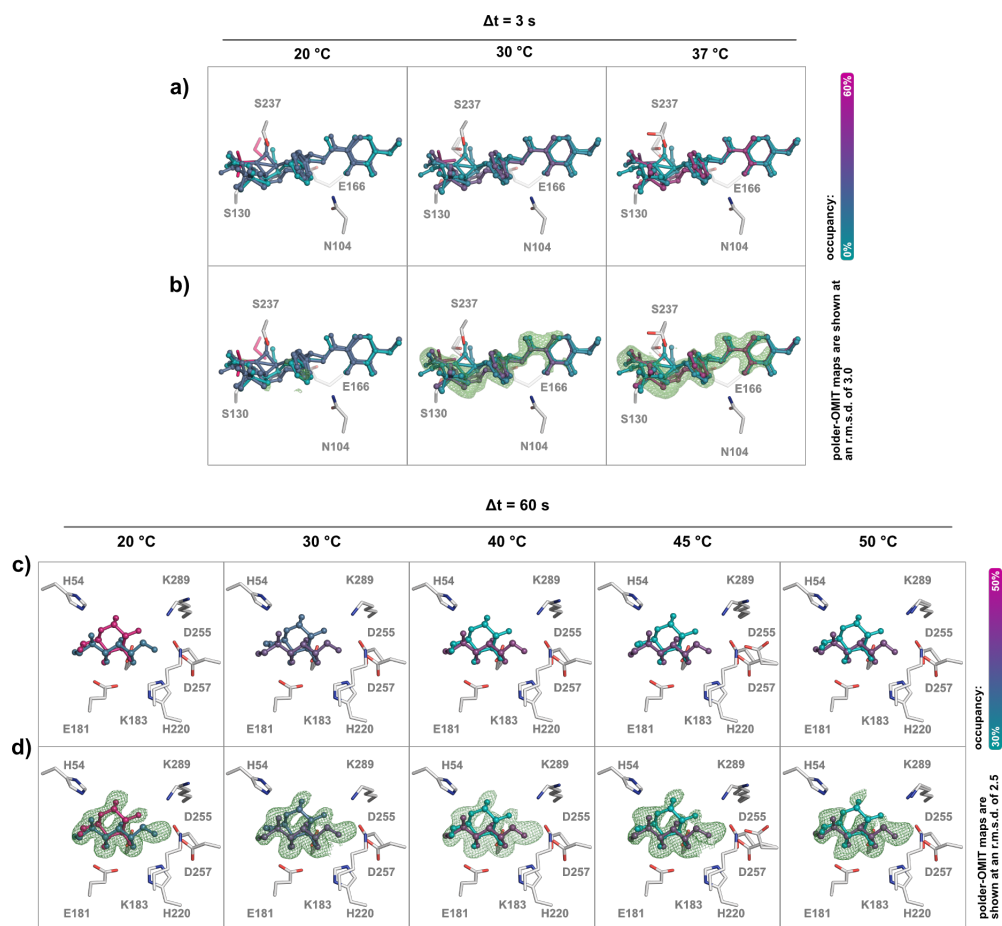
Contents

Supplementary Figures	2
Supplementary Tables	4
Crystallographic data statistics	4
Supplementary Notes	10
Environmental control box	10
Humidity controlled SSX	18
X-ray induced heating of protein μ -crystals	21
Supplementary Methods	24
Numerical calculations of the X-ray induced heating of μ -crystals	24
Characterisation of behaviour of RoPE space plots on number of diffraction patterns.	25
Group occupancy refinement settings	28
Electron density figure details	29
Supplementary Code	30
Script for the shifted inverse gamma distribution	30
Script to calculate the C_{α} RMSD	32
Mathematica 10.4 code for the numerical calculations	35

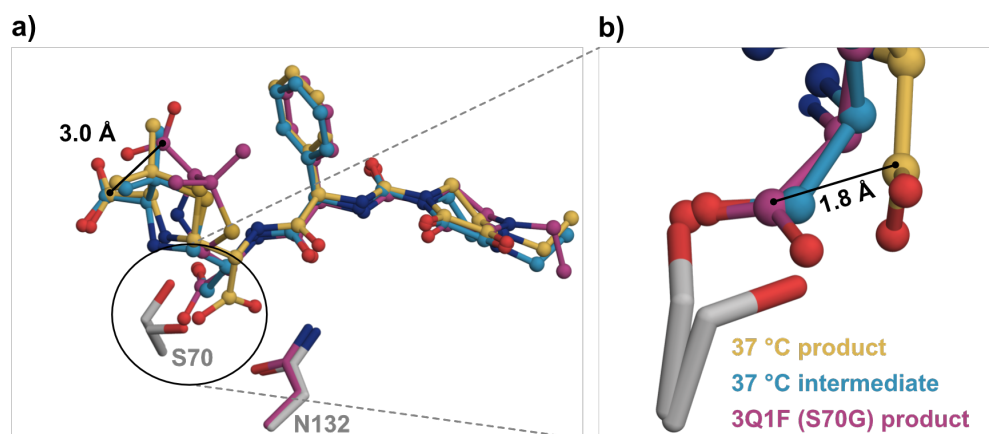
Supplementary Figures



Supplementary Figure 1 In-solution melting temperature reflects the activity optima. a,b) T_M determined by nanoDSF ($n=3$), with standard-deviation error bars) in 4 different buffer systems (Tab. 1) for each model system, (a) CTX-M-14 green, (b) XI orange. The in-solution melting temperature reflects the activity optima of the mesophilic and hyperthermophilic proteins, respectively. c,d) Pairwise C_α RMSD heatmap. Backbone RMSDs in Å between pairs of structures at different temperatures for both model systems (c) CTX-M-14, (d) XI. Source data are provided as a Source Data file.



Supplementary Figure 2 Altering enzyme kinetics by temperature modulation. a,b) CTX-M-14 active site, 3 s after reaction initiation at 20 °C, 30 °C, and 37 °C; c,d) XI active site, 60 s after reaction initiation at 20 °C, 30 °C, 40 °C, 45 °C, and 50 °C; a,c) refined fractional occupancy level; b,d) polder-OMIT map shown around the ligands as a green mesh at the indicated RMSD levels.



Supplementary Figure 3 5D-SSX reveal new product complex conformation. a,b) CTX-M-14 active site, 3 s after reaction initiation at 37 °C; Comparison to a previously published piperacillin product complex (S70G, PDB-ID: 3Q1F) shows a new orientation of the hydrolysis product in the wild-type protein. Notably, the carboxyl group close to the catalytic Ser70 is shifted by almost 2 Å.

Supplementary Tables

Crystallographic data statistics

Supplementary Table 1 Data collection and refinement statistics for CTX-M-14 apo data. Values in the highest resolution shell are shown in parentheses.

Temperature (°C) (PDB- ID)	10 °C (9G7V)	20 °C (9G7W)	30 °C (9G7X)	40 °C (9G7Y)	50 °C (9G7Z)
Data collection					
Space group	P322 ₁	P322 ₁	P322 ₁	P322 ₁	P322 ₁
Cell dimensions					
<i>a</i> , <i>b</i> , <i>c</i> (Å)	42.23, 42.26, 234.09	42.22, 42.24, 234.15	42.28, 42.31, 234.66	42.32, 42.34, 234.84	42.40, 42.44, 235.33
α , β , γ (°)	90.0, 90.0, 120.0	90.0, 90.0, 120.0	90.0, 90.0, 120.0	90.0, 90.0, 120.0	90.0, 90.0, 120.0
Resolution range (Å)	77.62 - 1.70 (1.76 -1.70)	77.62 - 1.70 (1.76 -1.70)	77.62 - 1.70 (1.76 -1.70)	77.62 - 1.70 (1.76 -1.70)	77.62 - 1.70 (1.76 -1.70)
Total reflections	12049024 (713700)	9717308 (575163)	7141365 (422198)	9167246 (544370)	6703777 (398067)
Unique reflections	27454 (2685)	27455 (2685)	27454 (2685)	27454 (2685)	27455 (2685)
Redundancy	428.9 (265.8)	353.9 (214.2)	260.1 (157.2)	333.9 (202.7)	244.2 (148.3)
Completeness (%)	100 (100)	100 (100)	100 (100)	100 (.00)	100 (100)
Mean I/ σ (I)	7.99 (1.99)	7.40 (2.17)	6.32 (1.79)	7.02 (1.71)	5.65 (1.04)
Wilson B-factor (Å ²)	22.12	21.19	21.97	23.14	25.98
<i>R</i> _{split}	0.101 (0.503)	0.112 (0.453)	0.127 (0.550)	0.112 (0.590)	0.133 (0.962)
CC1/2	0.985 (0.756)	0.979 (0.803)	0.972 (0.723)	0.978 (0.714)	0.967 (0.481)
CC*	0.996 (0.928)	0.995 (0.944)	0.993 (0.916)	0.994 (0.913)	0.992 (0.806)
Refinement					
Reflections used in refinement	27325 (2673)	27341 (2676)	27337 (2671)	27344 (2675)	27322 (2557)
<i>R</i> _{work}	0.1614 (0.2504)	0.1678 (0.2417)	0.1569 (0.2526)	0.1550 (0.2527)	0.1598 (0.3133)
<i>R</i> _{free}	0.1901 (0.2452)	0.1907 (0.2444)	0.1836 (0.2390)	0.1784 (0.2756)	0.1861 (0.2915)
Reflections used for <i>R</i> _{free}	1354 (112)	1354 (111)	1353 (112)	1354 (112)	1353 (111)
Number of non-hydrogen atoms	2242	2230	2209	2196	2177
macromolecules	2067	2060	2046	2044	2037
ligands	5	5	5	5	5
solvent	170	165	158	147	135
Average B-factor (Å ²)	28.15	27.03	27.87	28.20	31.46
macromolecules	27.24	26.22	26.97	27.36	30.69
ligands	36.95	34.88	38.77	42.07	46.95
solvent	38.89	36.81	39.20	39.32	42.47
<i>RMS deviations</i>					
Bond lengths (Å)	0.005	0.004	0.005	0.005	0.005
Bond angles (°)	0.772	0.675	0.812	0.825	0.795

Supplementary Table 2 Data collection and refinement statistics of the XI apo data.
Values in the highest resolution shell are shown in parentheses.

Temperature (°C) (PDB- ID)	20 °C (9G5N)	30 °C (9G5S)	40 °C (9G5W)	45 °C (9G5X)	50 °C (9G6I)
<i>Data collection</i>					
Space group	I 2 2 2	I 2 2 2	I 2 2 2	I 2 2 2	I 2 2 2
Cell dimensions					
<i>a</i> , <i>b</i> , <i>c</i> (Å)	94.2 103.05 99.25	94.2 103.05 99.25	94.2 103.05 99.25	94.2 103.05 99.25	94.2 103.05 99.25
α , β , γ (°)	90.0 90.0 90.0	90.0 90.0 90.0	90.0 90.0 90.0	90.0 90.0 90.0	90.0 90.0 90.0
Resolution	71.49 - 1.7	71.49 - 1.7	71.49 - 1.7	71.49 - 1.7	71.49 - 1.7
range (Å)	(1.761 - 1.7)	(1.761 - 1.7)	(1.761 - 1.7)	(1.761 - 1.7)	(1.761 - 1.7)
Total reflections	20298653	10229855	10613133	81189855	23289164
Unique reflections	53351 (5284)	53338 (5277)	53317 (5271)	53331 (5273)	53297 (5253)
Redundancy	380.3(224.3)	191.7(113.2)	198.8(117.7)	1521.2(904.5)	436.3(258.8)
Completeness (%)	99.96 (99.96)	99.93 (99.83)	99.89 (99.72)	99.92 (99.75)	99.85 (99.38)
Mean I/ σ (I)	5.55(3.31)	3.83(1.50)	3.59(0.98)	8.57(2.88)	4.87(1.14)
Wilson B-factor	15.26	17.54	20.14	19.82	21.96
R_{split}	20.92(33.98)	26.71(66.18)	24.74(102.55)	10.09(31.25)	17.46(90.39)
CC ^{1/2}	92.76(76.23)	89.39(56.02)	92.27(38.03)	98.64(86.29)	96.25(45.20)
CC*	98.10(93.01)	97.16(84.74)	97.97(74.23)	99.66(96.25)	99.04(78.90)
<i>Refinement</i>					
Reflections used in refinement	53351 (5284)	53338 (5277)	53317 (5271)	53331 (5273)	53297 (5253)
R_{work}	0.1624 (0.2901)	0.1680 (0.3272)	0.1727 (0.3735)	0.1453 (0.3158)	0.1590 (0.3597)
R_{free}	0.1906 (0.3290)	0.2096 (0.3910)	0.2072 (0.4166)	0.1824 (0.3611)	0.1903 (0.3987)
Reflections used for R_{free}	2607 (291)	2607 (291)	2606 (291)	2605 (290)	2603 (288)
Number of non-hydrogen atoms	3637	3566	3638	3552	3543
macromolecules	3275	3234	3325	3261	3284
ligands	2	3	2	3	2
solvent	360	329	311	288	257
Average B-factor (Å ²)	17.69	20.2	23.47	22.92	25.36
macromolecules	16.22	19.02	22.33	21.86	24.56
ligands	16.02	11.48	17.73	12.44	18.2
solvent	31.14	31.91	35.64	35.04	35.67
<i>RMS deviations</i>					
Bond lengths (Å)	0.006	0.016	0.004	0.013	0.004
Bond angles (°)	0.8	1.34	0.65	1.23	0.67

Supplementary Table 3 Data collection and refinement statistics of the CTX-M-14 Piperacillin data at a time-delay of 3 s. *Values in the highest resolution shell are shown in parentheses.*

Temperature (°C) (PDB-ID)	20 °C (9G80)	30 °C (9G81)	37 °C (9G82)
Data collection			
Space group	P322 ₁	P322 ₁	P322 ₁
Cell dimensions			
<i>a</i> , <i>b</i> , <i>c</i> (Å)	42.15, 42.15, 234.14	42.20, 42.20, 234.53	42.25, 42.25, 234.85
α, β, γ (°)	89.99, 89.99, 119.98	89.99, 89.99, 119.93	89.99, 89.98, 119.93
Resolution range (Å)	78.07 - 1.70 (1.76 -1.70)	78.07 - 1.70 (1.76 -1.70)	78.07 - 1.70 (1.76 -1.70)
Total reflections	1266008 (75053)	3599661 (216133)	4883576 (290896)
Unique reflections	27981 (2739)	27439 (2662)	27982 (2734)
Redundancy	45.2 (27.4)	131.2 (81.2)	174.5 (106.4)
Completeness (%)	1.00 (1.00)	1.00 (1.00)	1.00 (1.00)
Mean I/ σ (I)	1.91 (0.25)	4.46 (1.04)	4.36 (0.66)
Wilson B-factor (Å ²)	26.44	22.25	24.78
R_{split}	0.364 (4.577)	0.182 (0.995)	0.163 (1.595)
CC1/2	0.862 (0.034)	0.952 (0.446)	0.959 (0.252)
CC*	0.962 (0.257)	0.988 (0.786)	0.990 (0.634)
Refinement			
Reflections used in refinement	26826 (1948)	27336 (2363)	27851 (2569)
R_{work}	0.2281 (0.4130)	0.1645 (0.3038)	0.1687 (0.3592)
R_{free}	0.2556 (0.4706)	0.1948 (0.3061)	0.1995 (0.3746)
Reflections used for R_{free}	1315 (103)	1354 (141)	1377 (154)
Number of non- hydrogen atoms	2362	2371	2408
macromolecules	2156	2148	2193
ligands	41	41	41
solvent	165	182	174
Average B-factor (Å ²)	36.58	29.28	33.14
macromolecules	35.32	28.21	31.89
ligands	49.55	30.60	39.74
solvent	43.45	40.71	44.02
RMS deviations			
Bond lengths (Å)	0.002	0.005	0.003
Bond angles (°)	0.861	1.046	0.949

Supplementary Table 4 Data collection and refinement statistics of the XI 60 s data.
Values in the highest resolution shell are shown in parentheses.

Temperature (°C) (PDB- ID)	20 °C (9G6L)	30 °C (9G6M)	40 °C (9G6N)	45 °C (9G6O)	50 °C (9G6P)
<i>Data collec- tion</i>					
Space group	I 2 2 2	I 2 2 2	I 2 2 2	I 2 2 2	I 2 2 2
Cell dimen- sions					
<i>a, b, c</i> (Å)	94.2 103.05 99.25	94.2 103.05 99.25	94.2 103.05 99.25	94.2 103.05 99.25	94.2 103.05 99.25
α, β, γ (°)	90.0 90.0 90.0	90.0 90.0 90.0	90.0 90.0 90.0	90.0 90.0 90.0	90.0 90.0 90.0
Resolution range (Å)	71.49 - 1.7 (1.761 - 1.7)	71.49 - 1.7 (1.761 - 1.7)	71.49 - 1.7 (1.761 - 1.7)	71.49 - 1.7 (1.761 - 1.7)	71.49 - 1.7 (1.761 - 1.7)
Total reflec- tions	9415749	12055922	11970298	26069844	8506453
Unique reflections	53357 (5281)	53350 (5281)	53330 (5270)	53342 (5278)	53317 (5273)
Redundancy	176.4(104.5)	225.9(134.2)	224.3(133.2)	488.4(290.4)	159.4(94.7)
Completeness (%)	99.97 (99.91)	99.95 (99.91)	99.92 (99.70)	99.94 (99.85)	99.89 (99.75)
Mean $I/\sigma(I)$	4.37(2.68)	4.99(2.46)	4.42(1.65)	6.39(2.67)	3.24(0.93)
Wilson B- factor	15.89	17.92	19.59	21.24	22.4
R_{split}	25.28 (39.40)	20.92 (40.72)	21.34 (60.88)	15.16 (38.29)	27.93 (105.74)
$CC^{1/2}$	89.59(73.39)	92.56(76.39)	93.27(62.3)	96.44(79.42)	89.5(36.2)
CC^*	97.22(92.01)	98.05(93.07)	98.24(87.62)	99.09(94.09)	97.19(72.91)
<i>Refinement</i>					
Reflections used in refinement	53357 (5281)	53350 (5281)	53330 (5270)	53342 (5278)	53317 (5273)
Reflections used for R_{free}	2608 (291)	2607 (290)	2605 (289)	2608 (291)	2606 (290)
R_{work}	0.1657 (0.2765)	0.1549 (0.2899)	0.1604 (0.3401)	0.1479 (0.2998)	0.1744 (0.3566)
R_{free}	0.2051 (0.3178)	0.1922 (0.3259)	0.1997 (0.4004)	0.1859 (0.3468)	0.2153 (0.3938)
<i>Number of non-hydrogen atoms</i>	3617	3553	3590	3568	3507
macromolecules	3259	3224	3282	3270	3221
ligands	27	27	27	27	27
solvent	331	302	281	271	259
<i>Average B- factor</i> (Å ²)	19.03	21	23.39	24.18	26.14
macromolecules	17.77	19.93	22.44	23.3	25.37
ligands	14.24	16.09	18.23	19.95	21.63
solvent	31.89	32.89	35.02	35.25	36.23
<i>RMS devia- tions</i>					
Bond lengths (Å)	0.011	0.014	0.006	0.017	0.006
Bond angles (°)	1.09	1.29	0.8	1.49	0.78

Supplementary Table 5 Data collection and refinement statistics of the XI 180 s data.
Values in the highest resolution shell are shown in parentheses.

Temperature (°C) (PDB-ID)	20 °C (9I79)	50 °C (9I7L)
Data collection		
Space group	I 2 2 2	I 2 2 2
Resolution range	71.49 - 1.70 (1.76 - 1.70)	71.49 - 1.70 (1.76 - 1.70)
Space group	I 2 2 2	I 2 2 2
Cell dimensions		
<i>a</i> , <i>b</i> , <i>c</i> (Å)	94.20 103.05 99.25	94.20 103.05 99.25
α, β, γ (°)	90.0 90.0 90.0	90.0 90.0 90.0
Total reflections	6213031	5924612
Unique reflections	53348 (5279)	53334 (5270)
Multiplicity	116.4(68.7)	111.0(65.8)
Completeness (%)	99.95 (99.87)	99.92 (99.66)
Mean I/ σ (I)	3.99(2.22)	3.38(0.87)
Wilson B-factor	16.45	22.39
R_{split}	29.43(49.09)	25.62(115.22)
$CC^{1/2}$	86.30(62.09)	91.40(33.03)
CC^*	96.26(87.53)	97.73(70.37)
Refinement		
Reflections used in refinement	53348 (5279)	53334 (5268)
Reflections used for R_{free}	2607 (291)	2606 (290)
R_{work}	0.1719 (0.2873)	0.1690 (0.3618)
R_{free}	0.2116 (0.3444)	0.2066 (0.4436)
Number of non-hydrogen atoms	3617	3503
macromolecules	3253	3223
ligands	27	27
solvent	337	253
Protein residues	387	386
RMS(bonds)	0.016	0.013
RMS(angles)	1.30	1.22
Average B-factor (Å ²)	18.94	25.78
macromolecules	17.73	25.09
ligands	16.09	22.01
solvent	30.85	34.99

Supplementary Notes

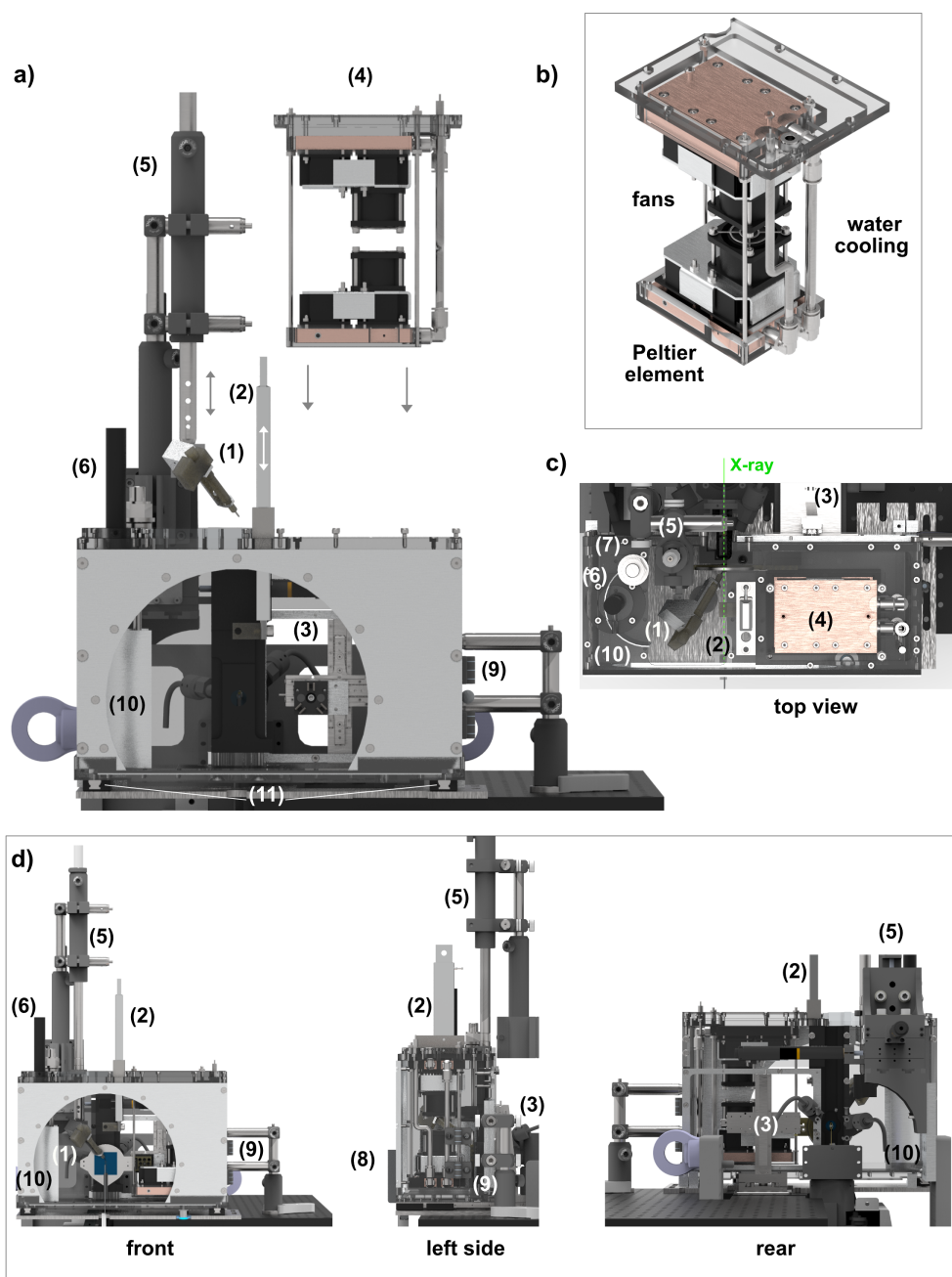
Environmental control box

Design

To maintain a controlled humidity environment for our hit-and-return (HARE) chip setup including the liquid-application-method for time-resolved crystallography (LAMA) [40, 42], required the development of a solution that could accommodate this experimental setup. To this end we have constructed a modular, compact environmental control box that encloses our previously described chip setup including the LAMA droplet injector nozzle on a footprint of 118 mm x 283 mm (Sup. Fig. 4, 5) [43]. Humidity control is achieved by flowing dry air either directly into the box, or first passing through a water bath (20-95 °C), with the proportion of gas through each channel controlled by a toggling ball valve in a proportional-integral-derivative (PID) feedback loop. The water bath is connected to the environmental control box via a silicone hose, to which a heating belt is attached preventing condensation. The set point can be achieved with high accuracy, enabling humidity control within 1 percent point of relative humidity, which allows to perform controlled crystal dehydration if that is required. Many beamlines are equipped with a temperature control solution via a gas stream that is directed at the sample, sometimes encompassing wide temperature windows but typically limited to room temperature. Interestingly, however, a combined temperature-humidity control is rarely found. Such a situation mandates the use of e.g., glass capillaries to maintain the crystals in a humid environment, which complicates time-resolved applications that are based on in-situ mixing. Historically, flow-cells were developed for time-resolved applications that allowed for in-situ mixing experiments with single crystals and for trapping reaction intermediates on comparably slow time scales [85–87], which were recently extended to SSX experiments [88]. However, to the best of our knowledge these were not applied to multi-temperature experiments. Serial crystallography experiments can also be conducted in controlled,

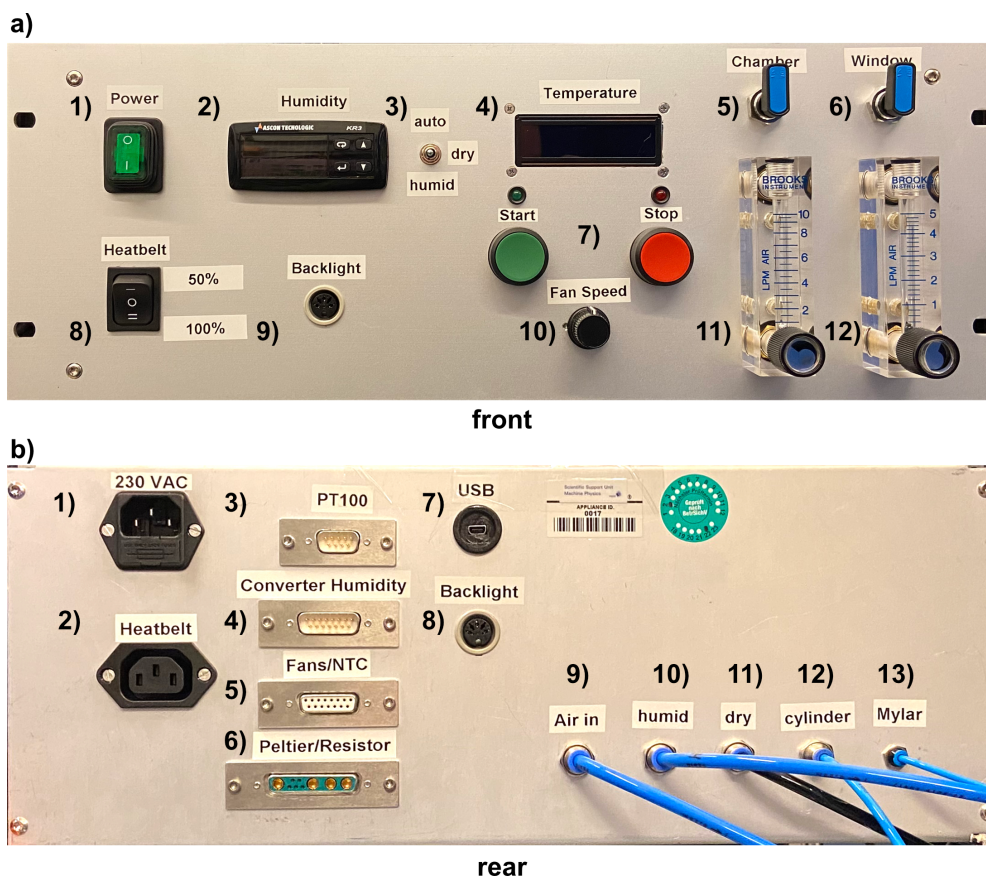
closed-boundary environments as demonstrated by the drop-on-demand device, which permits humidity control and fully anaerobic experiments via the exchange of the surrounding atmosphere [89]. In addition to humidity control, the temperature within our enclosure can be adjusted anywhere within the range of approximately 7 °C to above 70 °C. An air-stream reflector directs the stream of humid air around the chip. In order to enable effective control over this wide temperature range, the box uses two interchangeable modular temperature control units (Sup. Fig. 5). Module-1 covers a temperature range from approximately +7 °C to approximately +50 °C, while module-2 covers a temperature range from approximately +50 °C to over +70 °C. Rapid exchange of the modules is possible without tools, enabling switching between different temperature regimes within a few minutes. Module-1 contains water-cooled Peltier-elements that enable active cooling or heating of the interior of the box. To ensure that the temperature is equilibrated across the box, the module is equipped with fans. Cooling water and electric power are fed in through the top side of the module. The heating element in module-2 is a power resistor network that disseminates sufficient heat to increase the interior temperature of the box to over 70 °C, while the relative humidity can be sustained at over 95%. Temperature control is achieved by a PID controller that sets the current through the Peltier elements and resistor network, respectively, to maintain the target temperature. The base plate of the box is made from durable polyether ether ketone (PEEK). To drain condensation water, several cotton wicks are fixed around the bottom corners and connected to an active pumping system that quickly drains excess liquid from the box. The sides and the lid of the box are made of 6 mm thick acrylic glass, while the rear panel (X-ray side) is made of acrylic glass and polyoxymethylene (POM). As an X-ray entrance window, an 8 mm opening in the POM is covered with two spaced layers of COC foil, about 1 mm apart. To enable easy replacement of the X-ray entrance window, the COC foil is placed on a magnetically mounted ring that tightly seals the inside of the box. While one side

panel is solid, the other side panel has a feedthrough for the humidity tube, and an access hatch through which HARE chips can be loaded onto the sample translation stage. The front panel (detector side) is an aluminum frame with a 190 mm opening. This is sealed with two X-ray transparent Mylar foils (6 μm), as an exit window for the diffracted beam. To reduce condensation on this window, the space between the Mylar foils is continuously flushed with warm air. The lid of the box contains feedthroughs for the humidity and temperature sensors, as well as for the electropneumatic retractable infrared (IR) backlight, the LAMA-nozzle lever, and an access port for heating module exchange. The translation stage system implemented at the T-REXX endstation is not humidity resistant and therefore has to be kept outside of the box. Hence, the translation stages are connected to the box via a flexible bellow, custom cut from two layers of commercial plastic wrap. On the inside, the bellow is sealed between the translation stages and the kinematic mount for the chip holder [63, 90]. To reduce the heat capacity of the chip holder and thus achieve faster temperature equilibration, the previously described aluminum chip holder was redesigned from PEEK, providing the same functionality at a lighter weight [63]. The LAMA nozzle is attached *via* a kinematic mount to a retractable lever that enables retraction of the nozzle from its injection position during chip exchange. To avoid the unnecessary opening of the box, which might lead to temperature and humidity fluctuations, the nozzle retracts into a parking position under the lid of the box. Fine alignment of the LAMA nozzle in the injector position is achieved *via* motorized translation stages (Thorlabs). The whole box system is mounted on rails, residing on a stainless-steel baseplate that can be moved between “data collection” or “beam location” position, where the latter allows to use the X-ray scintillator built into the beam-shaping device (BSD; Arinax, Moirans, France). Operation of the serial crystallography environmental control enclosure is achieved *via* an external control unit where temperature and



(1): LAMA nozzle (4): Peltier module (7): temperature sensor (10): air stream reflector
 (2): IR backlight (5): LAMA guillotine (8): beamstop (11): retraction rails
 (3): translation stages (6): humidity sensor (9): access hatch

Supplementary Figure 5 The Serial Crystallography Environmental Control Box. a) overview of the environmental control box, with retracted LAMA nozzle and Peltier module-1. b) closeup of the Peltier module-1, c) top-view providing an overview of the arrangement inside the box, d) front-, side- and rear-view of the box. Note: for clarity module-2 and some technical elements of the box (e.g. tubing, electric connections etc.) or the beamline are not shown or described in detail. Elements mentioned in the text are numbered and shown in the figure legend.



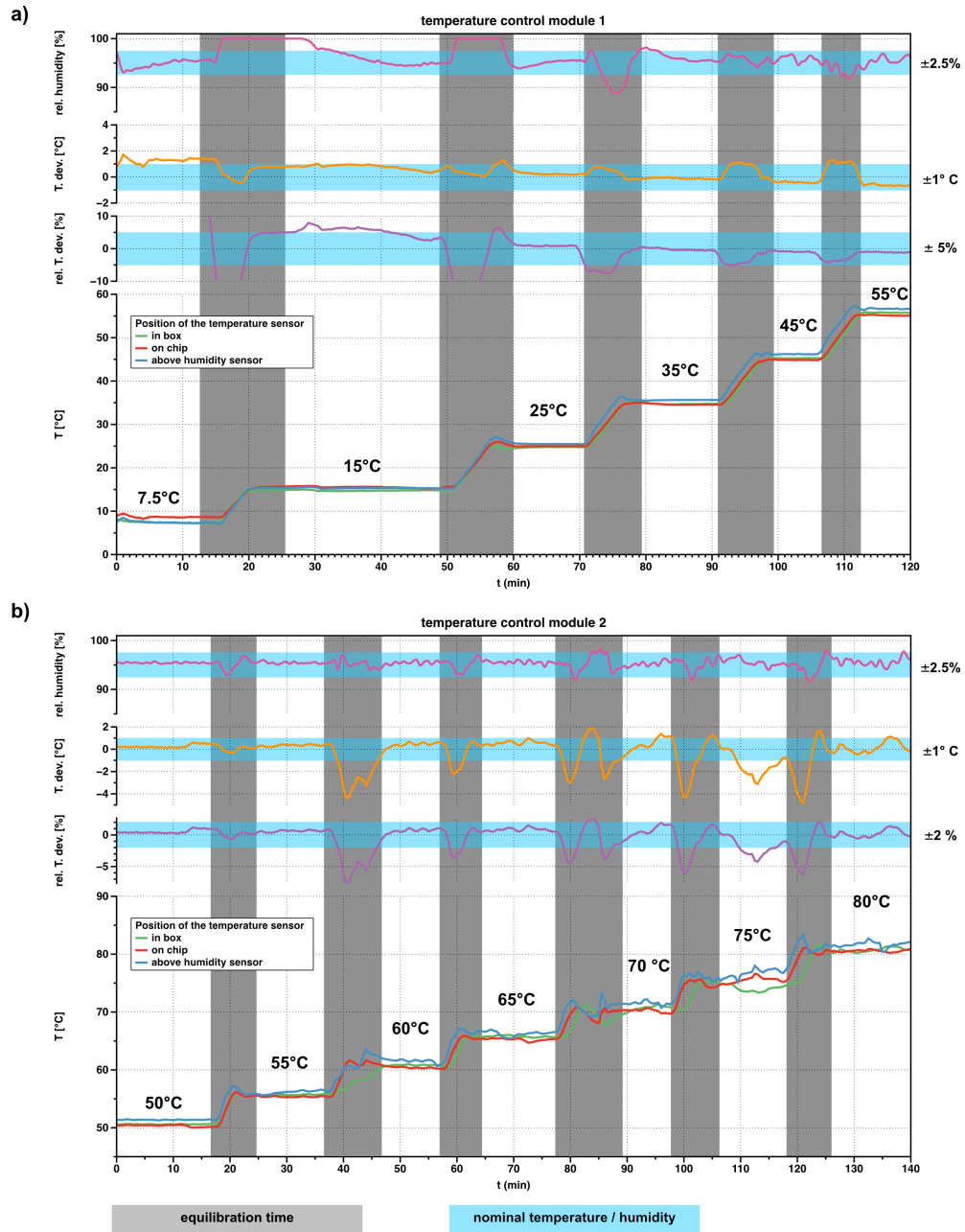
Supplementary Figure 6 Control unit of the serial crystallography environmental control box. a) front panel: 1) main power switch; 2) humidity control/display; 3) air flow switch; 4) temperature display; 5) airflow on/off switch; 6) inflatable front-window flow on/off switch; 7) Temperature control start/stop button; 8) heat-belt switch; 9) socket for IR backlight switch; 10) fan-speed regulator; 11) air-flow valve; 12) front-window flow valve. b) rear panel: 1) main power inlet; 2) heat belt power outlet; 3) PT100 temperature sensor; 4) humidity sensor; 5) fan connection; 6) Peltier/resistor connection; 7) USB connection to PC; 8) backlight connection; 9) pressurized air in; 10) air flow to water bath; 11) dry air outlet; 12) backlight cylinder air outlet; 13) inflatable front-window air outlet.

box can be achieved and maintained, we characterised the temperature increase from 7.5 °C to 80 °C. The data show that for both temperature control modules the humidity values quickly reach the target values. Over a temperature window of ca. 70 °C the humidity remains stable within 2.5% of the set point. Analysis of the deviation of the chip temperature from the box temperature shows that the chip temperature follows the box temperature with a median difference of 0.7 °C, over a temperature window of

Supplementary Table 6 Environmental control parameter settings

	desired temp.	flow rate	desired humid- ity	actual humid- ity	water bath temp	heat belt power	module current
	$^{\circ}\text{C}$	L min^{-1}	(%)	(%)	$^{\circ}\text{C}$	(%)	(A)
module-1	7.5	5	95	95	75	0	-12.3
	15	5	95	95	75	50	-2.1
	25	5	95	95	75	100	1.7
	35	4	95	95	85	100	3.5
	45	4	95	95	95	100	4.8
	55	4	95	95	95	100	6
module-2	50	4	95	95	95	100	2.4
	55	4	95	95	95	100	2.9
	60	4	95	95	95	100	2.6
	65	4	95	95	95	100	2.9
	70	4	95	95	95	100	3.1
	75	4	95	95	95	100	4.3
	80	4	95	95	95	100	3.4

ca. 70 $^{\circ}\text{C}$. Temperature and humidity typically equilibrate across the box and the chip within 10-15 minutes. We also examined the reliability of maintaining environmental set points during X-ray data collection. To this end we collected X-ray diffraction data at 20 $^{\circ}\text{C}$, 40 $^{\circ}\text{C}$, 55 $^{\circ}\text{C}$ and 80 $^{\circ}\text{C}$, for ca. 120 minutes and recorded temperature and humidity values in 30 s intervals during this period (Sup. Tab. 7). Remarkably, during the data collection the target humidity could be maintained within approximately 1%, while the temperature remained stable within approximately 0.5 $^{\circ}\text{C}$. Clear deviations from this behavior are only observed during chip exchange, when the hatch of the box is opened and during the subsequent re-equilibration time while the environment stabilises. Re-equilibration of the environmental parameters could be achieved within 10-15 minutes, depending on the duration of the manual chip exchange. In conclusion, these data show that after an equilibration time of approximately 10-15 minutes the environment in the control box has reached its target value, and can be maintained throughout extended periods of time, well beyond the typical data collection time of a chip (ca. 30 minutes). This enables collection of serial X-ray diffraction data at a variety of different temperature and humidity levels with high accuracy and precision.



Supplementary Figure 7 Characterization of the environmental control. a) module-1: 7.5 °C – 55 °C; b) module-2: 50-80 °C; The temperature was successively increased from 7.5 °C to 55 °C and from 50 °C to 80 °C respectively. The temperature was measured at three positions inside of the chamber: green inside the box near the panel opposite to the access hatch, blue directly above the humidity sensor, red directly on the chip. The target humidity was set to 95%. The grey bars indicate the equilibration time, blue bars indicate the target window.

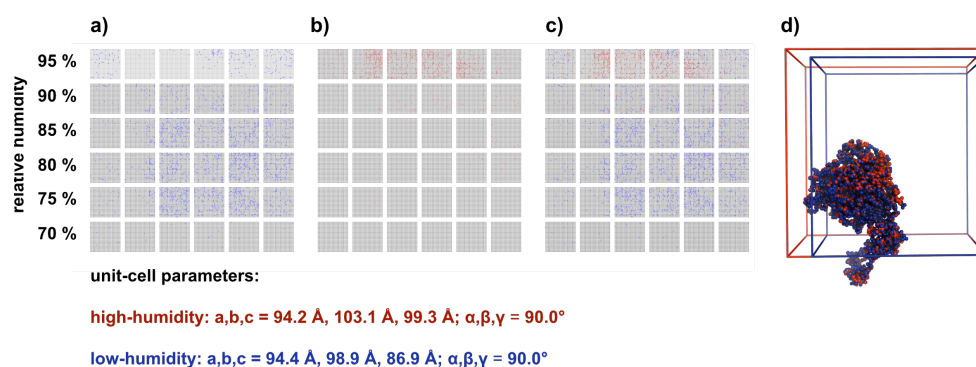
Supplementary Table 7 Environmental parameters during X-ray diffraction data collection. *values are derived from a long-term (10 h) experiment

nominal temp. (°C)	nominal rel. humidity (%)	measured rel. humidity (%)	RMSD (%)	measured temp. (°C)	RMSD (°C)
20	95	95.4	0.5	19.8	0.2
40	95	95.5	0.9	40.3	0.3
55	95	95.5	0.7	55.3	0.7
80	95	95.2	0.7	80.1	0.3
20*	95*	95.2*	0.7*	19.8*	0.2*

Humidity controlled SSX

A hallmark of protein crystals is their large solvent content, which is typically in the range between 40% and 70% of the crystal volume. An advantage of this property is that proteins generally retain their function even in the crystalline state [91, 92]. A commonly known disadvantage is, however, their sensitivity to changes in environmental humidity, which in addition to the higher rates of radiation damage associated with higher temperatures makes routine data collection at even ambient temperatures a difficult task. Accordingly, starting with traditional wax-enclosed glass capillaries, environmental control has consistently been a key aspect of macromolecular crystallography, and several solutions have been developed to maintain crystal hydration for single crystals [4, 93]. Open- and closed boundary environmental control solutions have been developed since the advent of structural biology. In the simplest instances, closed boundary devices include glass capillaries, which contain protein crystals and typically a drop of mother liquor to sustain a humid atmosphere during data collection [93]. More modern variations of this classic solution are the many fixed-target serial crystallography environments, which protect protein microcrystals against evaporation by some form of X-ray transparent window material [94]. For single crystal experiments a variety of solutions, sometimes for advanced parameter control, such as humidity, temperature, and electric fields have also been developed over the years [95–97]. With the onset of cryo-crystallography, larger boxes were developed, which sometimes enclosed

the stream of cryogenic gas in a dry atmosphere to prevent ice-formation during data-collection [98]. However, for single-crystal experiments the majority of environmental control solutions fall into the open-boundary category, such as placing the crystal in an vapor stream with controlled humidity [36, 96, 99–102]. Historically, the water content of protein crystals was controlled by post-crystallization treatments via chemical dehydration prior to crystal freezing [34, 103]. However, controlling the humidity around the mounted crystals enables the convenient identification of the optimal conditions for a particular sample during an X-ray diffraction experiment [36, 96, 99–102]. Adjusting the relative humidity either prior to, or during data collection can improve several aspects of data quality (resolution, mosaicity and anisotropy) [36, 37, 39, 99, 101, 104–107]. To estimate the effect of the environmental humidity on diffraction data quality we monitored the unit-cell size of XI as a function of decreasing humidity. We started data collection at a relative humidity of 95% and reduced the humidity in steps of 5% per compartment row on the chip (**Fig. 8**). With decreasing humidity the crystals do not simply cease to diffract but undergo a change in unit-cell size. While at or above a relative humidity of 95% most diffraction patterns could be indexed with a larger unit-cell ($a = 94.2 \text{ \AA}$, $b = 99.3 \text{ \AA}$, $c = 103.1 \text{ \AA}$; $\alpha, \beta, \gamma = 90.0^\circ$), but the proportion rapidly changed to a smaller unit cell ($a = 94.6 \text{ \AA}$, $b = 99.4 \text{ \AA}$, $c = 87.5 \text{ \AA}$; $\alpha, \beta, \gamma = 90.0^\circ$) as the relative humidity dropped from 90% to 75%. If the humidity is reduced even further the micro-crystals cease to diffract, presumably due to complete dehydration. This emphasizes the sensitivity of protein micro-crystals to environmental humidity, which must be precisely controlled to maintain their diffraction properties. On the other hand, this also opens the opportunity for crystals with large unit cells to be specifically dehydrated to modulate their diffraction properties. The response of protein crystals hydration to their environment has long been known [34] and chemical dehydration devices [103, 108–110] as well as dedicated de-humidification devices have successfully been used for this purpose on single, loop-mounted crystals [39, 99, 101].



Supplementary Figure 8 Humidity dependent unit-cell modulation displayed in a hit map. The HARE chips consist of 6x6 compartments, each divided into 24x24 features. Each feature on the HARE-chip that can contain a crystal is represented by a small grey square; if a diffraction pattern is recorded the particular square is highlighted in colour. A blue square indicates a diffraction pattern corresponding to a low humidity unit cell, while a red square corresponds to a diffraction pattern in the larger high-humidity unit cell. Humidity has been reduced by 5% for each row of compartments. a) recorded diffraction patterns corresponding to low-humidity unit-cells b) recorded diffraction patterns corresponding to high-humidity unit-cells; c) overlay of low- and high-humidity unit-cell diffraction pattern hits; d) comparison of high- and low-humidity unit-cells.

With our environmental control box these post-crystallization optimization protocols are now open to serial crystallography.

X-ray induced heating of protein μ -crystals

While the environmental control box provides a stable temperature bath for the protein crystals, their temperature is also altered by the incident X-ray beams during data collection. As laid out by Warren et al. [111] the simplest estimate of beam-induced temperature changes is provided by an adiabatic model that ignores any heat-exchange with the environment. Thus the maximum possible increase in temperature is given by the energy absorbed by the mass and the specific heat capacity of the sample. Since the energy absorbed by the mass can also be expressed by the absorbed dose, this can be expressed as:

$$\Delta T = \frac{Q}{m \cdot c_p} = \frac{Q_D}{c_p} \quad (1)$$

,wherein ΔT is the temperature change, $\frac{Q_D}{m}$ is the energy absorbed per unit mass, c_p is the specific heat capacity, and Q_D is the absorbed dose. The specific heat capacity of protein crystals is approximated via those described for tetragonal lysozyme (1.8×10^3 J/ kg K) [112]. The absorbed dose was calculated using *RADDOSE-3D* [113], based on the parameters given in Table 8, below.

However, as an adiabatic model clearly ignores heat-exchange with the mother-liquor, to the crystalline silicon-chips, to the air-interface inside the wells, as well as convectional cooling, photoelectron escape or any energy conversion by radiolytic processes, we consider the adiabatic temperature increase an upper boundary, while realistic temperature changes are certainly well below these values. In order to further investigate this, we have modelled the thermal diffusion process into the surrounding regions using numerical simulations (Suppl. Methods). To predict the spread of heat, if thermal diffusion is taken into consideration we employed a conservative model. To

Supplementary Table 8 Raddose-3D parameters to calculate the upper boundary of X-ray induced heating.

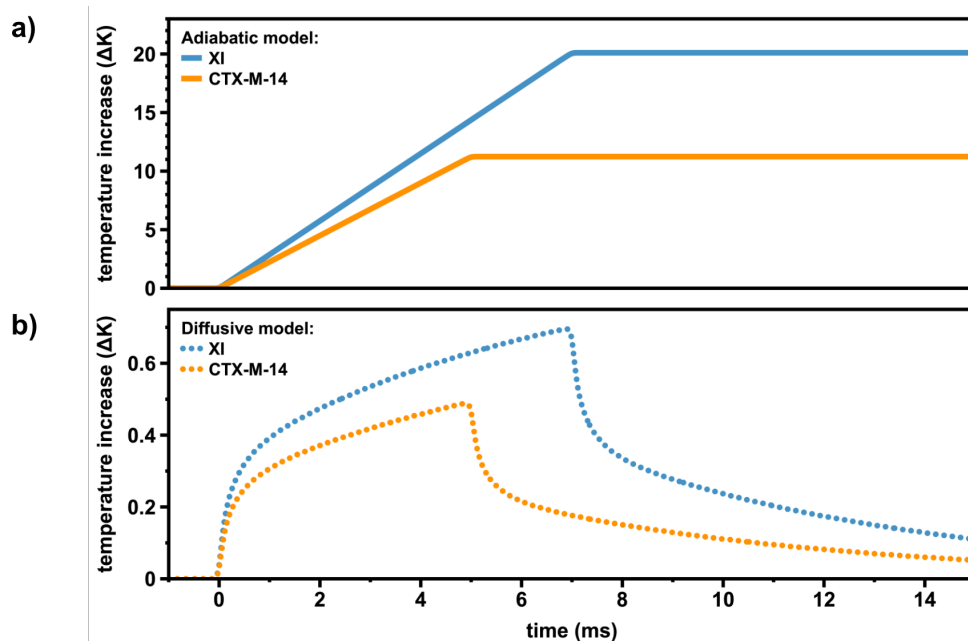
Protein	XI	CTX-M-14
Crystal type	Cuboid	
Dimensions [μm]	20, 20, 10	15, 15, 10
Beam type	Gaussian	
Flux [ph/s]	1.00×10^{12}	1.10×10^{12}
FWHM [μm]	30, 10	30, 10
Energy [keV]	12.699	12.699
Collimation Rectangular [μm]	10, 9	10, 9
Wedge	0, 0.0001	0, 0.0001
Exposure time [s]	0.007	0.005
Average Dose (exposed region) [J/kg]	36222	20234
specific heat capacity [J/kg K]	$1.8 \times 1.8 \times 10^3$	1.8×10^3
adiabatic model ΔT [K]	20.12	11.24
diffusive model ΔT [K]	0.7	0.5

this end we utilised the thermal diffusivity of water ($\alpha = 1.4 \times 10^{-6} \text{mm}^2/\text{s}$) as a lower-bound estimate, compared to crystalline environments (Sup. Tab. 9).

Supplementary Table 9 Parameters used to calculate the X-ray induced heating in protein mirco-crystals.

Abbreviation	Quantity	Units	Reference
c_p	Heat capacity for tetragonal lysozyme	$1.8 \times 10^3 \text{ J / kg} \cdot \text{K}$	Kriminski et al 2003 [112]
α	Thermal diffusivity of water	$1.4 \times 10^{-6} \text{ mm}^2 / \text{s}$	Blumm et al 2003 [114]

The simulations, which include the diffusion of heat to the surrounding region, therefore exhibit an over-estimation of the possible temperature increase. Considering this more realistic scenario with heat dissipation into the surrounding region, yields a temperature increase of 0.7 K for XI and 0.5 K for CTX-M-14, respectively (Sup. Fig. 9; Sup. Tab. 9). However, with respect to turnover kinetics the delay time after reaction initiation has to be taken into consideration. While the X-ray exposure time was 5 and 7 ms, respectively, the delay times were orders of magnitude larger for both CTX-M-14 (3s), and XI (60s, 180s). This long time delay enables progression of ligand binding and turnover kinetics, irrespective of any potential X-ray induced heating. While we note that this temperature increase of 0.5°- 1°C may further influence the



Supplementary Figure 9 X-ray induced heating in protein μ -crystals. (upper panel - adiabatic model): A temperature increase of up to 20K can be observed if no heat dissipation into the surrounding medium is considered. (lower panel - diffusive model): However, if the surrounding region is thermally conductive the temperature increase is limited to less than 1K, for each case. Source data are provided as a Source Data file.

turnover kinetics, it is important to emphasize that this upper boundary can be considered a constant offset that applies to all time-points. That is the deviation from the nominal temperature change would affect all time- and temperature-points uniformly, and therefore not lead to deviations from the relative temperature changes set by the environmental control box.

Supplementary Methods

Numerical calculations of the X-ray induced heating of μ -crystals

Calculations were made in order to approximately determine the temperature increase caused by the absorption of the X-ray beam. The absorbed dose Q_D was first determined using RADDOS-3D [113], based on a static model without thermal diffusion (see below). The temperature rise ΔT is then determined by $\Delta T = \frac{Q_D}{c}$, where c is the specific heat capacity.

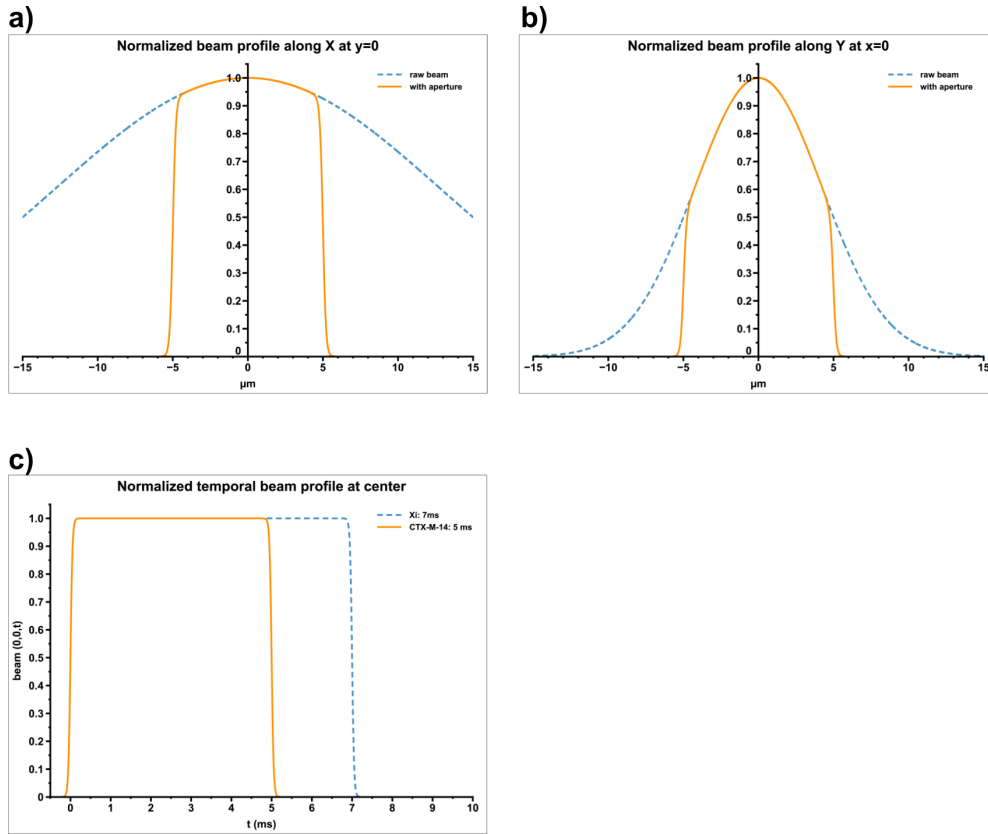
If thermal diffusion is taken into account, the resulting temperature increase is lower, as the absorbed energy spreads into the surrounding regions. To quantify this effect, numerical simulations were performed using *Mathematica* [115]. For the details of the Mathematica-code, please refer to the code-section below.

The heat equation $\dot{u} = \Delta u$ was applied, restricted to the two spatial dimensions perpendicular to the beam axis. Additionally, a time- and position-dependent source term, $R \cdot Q_S(\mathbf{r}, t)$, was included to represent the heat input from the X-ray beam. This beam was modelled as a collimated Gaussian profile $Q_S(\mathbf{r})$ active only during the exposure time t_{exp} (Sup. Fig. 10).

$$\partial_t u(\mathbf{r}, t) = \alpha \Delta u(\mathbf{r}, t) + R \cdot Q_S(\mathbf{r}, t), \quad \text{with} \quad (2)$$

$$Q_S(\mathbf{r}, t) = \begin{cases} Q_S(\mathbf{r}) & \text{if } 0 \leq t \leq t_{\text{exp}} \\ 0 & \text{otherwise} \end{cases} \quad (3)$$

The parameter R was chosen so that the temperature increase when heat diffusion is neglected and thermal diffusivity α is zero, matches the results obtained from RADDOS-3D, thus $R = \frac{\Delta T}{t_{\text{exp}}}$. Enabling thermal diffusion by assigning α a non-vanishing value results in a temperature increase of less than 1 K, compared to roughly 20 K from calculations without diffusion.

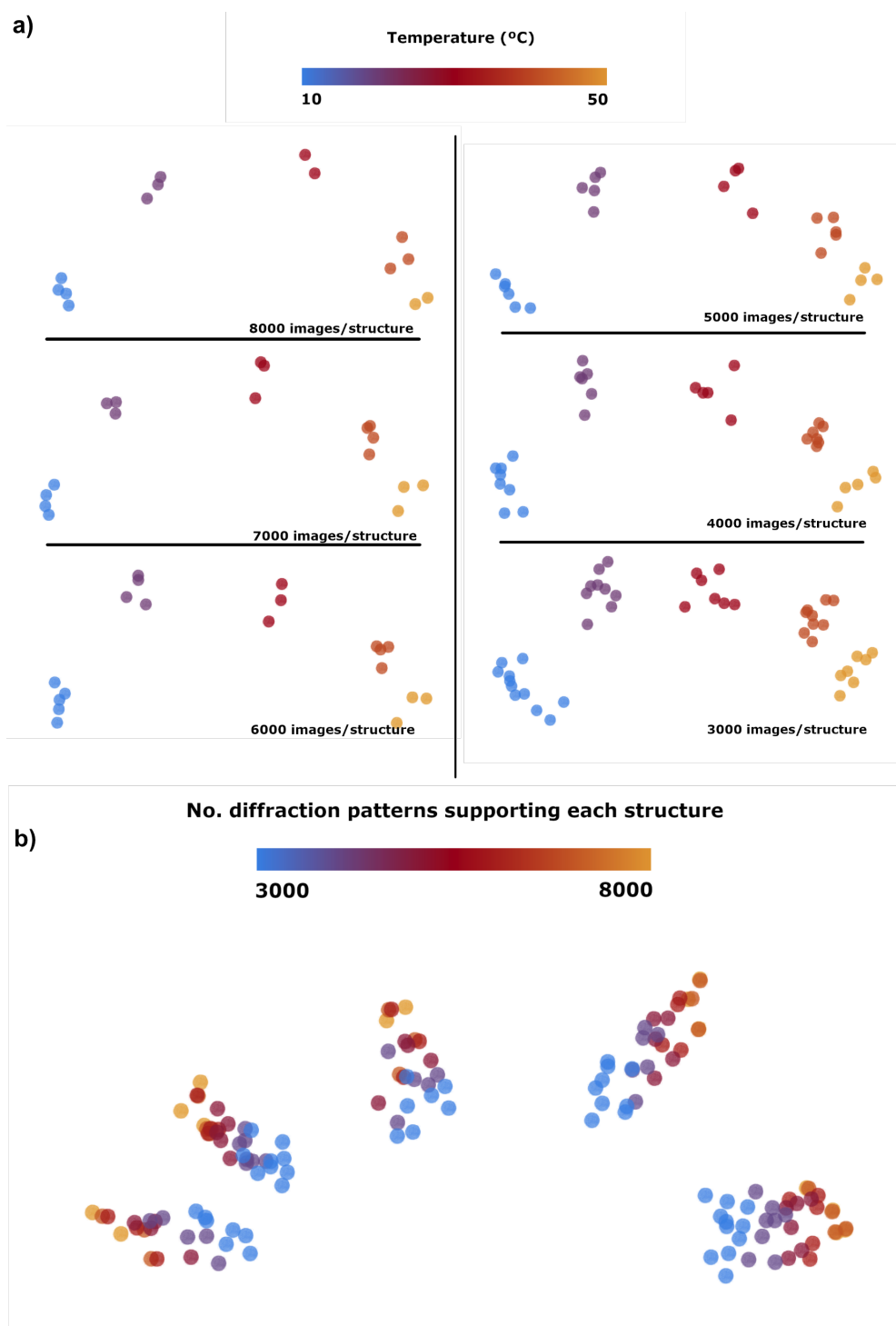


Supplementary Figure 10 Beam profile used in numerical calculations for the X-ray induces heating. a) normalized beam profile along X at y=0; b) normalized beam profile along Y at x=0; c) normalized temporal beam profile at the centre. Source data are provided as a Source Data file.

Characterisation of behaviour of RoPE space plots on number of diffraction patterns.

RoPE space plots take advantage of the oversampling of serial diffraction data in order to establish a statistical support for interpretation of structural differences, by splitting the data for independent structure solutions. This needs to strike a balance, for a given number of diffraction patterns, between the total number of structures and the number of diffraction patterns assigned to each structure. The number of diffraction patterns will influence the quality of the data supporting the analysis. Here

we vary the approximate number of diffraction patterns in each structure from 3000 to 8000 images, in steps of 1000 images. Naturally, with more images per structure, fewer structures are generated. We see that the separation is present for all image numbers per structure (Sup. Fig. 11 a), but to some extent, the "tightness" of each cluster increases with more supporting images per structure. Showing all structures at the same time, this time coloured by number of images (Sup. Fig. 11 b), the clusters corresponding to respective temperatures are still recognisable. However, this shows that structures supported by fewer images are more similar to each other despite being different temperatures. This is likely because of poorer quality diffraction intensity estimates and greater reliance on the geometry term rather than the X-ray data during refinement. However, structures supported by more images can reveal more features associated with the given temperature, due to higher quality intensity estimates, and therefore are spaced further apart from one another.



Supplementary Figure 11 Titration of the²⁷ number of diffraction patterns for ROPE analysis. a) Titration of successively lower number of diffraction patterns per structure used to generate the RoPE space. b) RoPE space summarising the behaviour of varying the number of diffraction patterns supporting each structure.

Group occupancy refinement settings

Following group occupancy refinement setting have been used for CTX-M-14:

```
1 occupancies {
2     individual = None
3     remove_selection = None
4
5     constrained_group {
6         selection = resseq 301 and chain A and altloc A
7         selection = resseq 302 and chain A and altloc B
8         selection = resseq 302 and chain A and altloc C or resseq 70 and chain A
          and altloc C
9         selection = resseq 162 and chain S or resseq 163 and chain S or resseq 164
          and chain S or resseq 165 and chain S or resseq 167 and chain S or resseq 168
          and chain S or resseq 171 and chain S or resseq 173 and chain S or resseq 174
          and chain S or resseq 176 and chain S or resseq 177 and chain S or resseq 180
          and chain S or resseq 181 and chain S or resseq 1 and chain B
10    }
11    constrained_group {
12        selection = resseq 70 and chain A and altloc C
13        selection = resseq 70 and chain A and altloc A
14    }
15 }
```

Following group occupancy refinement setting have been used for XI:

```
1 occupancies {
2     individual = None
3     remove_selection = None
4     constrained_group {
5         selection = chain C and element Mg
6     }
7     constrained_group {
8         selection = chain D and element Mg
9     }
10    constrained_group {
11        selection = chain A and resseq 402 and resname HOH or chain A and \
12            resseq 403 and resname HOH or chain A and resseq 404 and \
13            resname HOH or chain A and resseq 405 and resname HOH or \
14            chain A and resseq 406 and resname HOH or chain A and \
15            resseq 407 and resname HOH
16        selection = chain A and resseq 401 and resname GLO
17    }
```

```

17         selection = chain A and resseq 401 and resname GLC
18     }
19 }

```

Electron density figure details

Supplementary Table 10 PyMol color settings to display volume elements.

Color	map RMSD.	R	G	B
Blue	1.0	19	43	157
Cyan	1.5	153	219	223
Green	2.0	101	198	66
Yellow	2.5	255	240	0
Orange	3.0	252	131	0
Red	3.5	245	38	0

Supplementary Code

Script for the shifted inverse gamma distribution

```
1  #!/usr/bin/env python3
2  # -*- coding: utf-8 -*-
3  """
4  Created on Wed Jul 17 10:08:46 2024
5
6  @author: gargi
7  """
8
9  import numpy as np
10 import matplotlib.pyplot as plt
11 from scipy.special import gamma
12 from scipy.optimize import curve_fit
13
14 # Define a Shifted Inverse Gamma Function
15 def sigd(x, a, b, d):
16     return (b**a / gamma(a)) * ((x - d)**(-a - 1)) * (1 / np.exp(b / (x - d)))
17
18 # Common settings for plots
19 plt.rcParams['figure.dpi'] = 300
20 plt.rcParams['savefig.dpi'] = 300
21
22 # File paths and colors
23 file_paths = ['', '', '', '', '', '']
24
25
26 colors = ['royalblue', 'darkviolet', 'limegreen', 'gold', 'orangered']
27 labels = ['10 C ', '20 C ', '30 C ', '40 C ', '50 C ']
28
29 all_fits = []
30
31 for idx, (file_path, color, label) in enumerate(zip(file_paths, colors, labels)):
32     b_values = np.loadtxt(file_path, dtype=float)
33     freq, bins, _ = plt.hist(b_values, bins=80, density=True, color='skyblue')
34     plt.xlim(0, 150)
35
36     bincenters = 0.5 * (bins[:-1] + bins[1:])
37     s = min(bincenters)
```

```

38     d = 0.9 * s
39
40     popt, _ = curve_fit(lambda x, a, b: sigd(x, a, b, d), bincenters, freq, bounds
41                         =([2, 0], [4, 200]))
42     fit = sigd(bincenters, popt[0], popt[1], d)
43
44     all_fits.append((bincenters, fit, color, label))
45
46     plt.xlim(0, 100)
47     plt.ylim(0, 0.10)
48     plt.xlabel('B Values [ $\sim 2$ ']')
49     plt.ylabel('Frequency')
50     plt.title(f'SIGD Fit {label} Data')
51     plt.plot(bincenters, fit, color)
52     plt.show()
53
54 # Combined plot
55 for bincenters, fit, color, label in all_fits:
56     plt.plot(bincenters, fit, color, label=label)
57
58 plt.legend(loc="upper right")
59 plt.xlabel('B Values [ $\sim 2$ ']')
60 plt.ylabel('Frequency')
61 plt.title('SIGD Fit : CTXM')
62 plt.xlim(0, 100)
63 plt.ylim(0, 0.10)
64 plt.show()

```

Script to calculate the C_α RMSD

```
1  #!/usr/bin/env python3
2  # -*- coding: utf-8 -*-
3  """
4  Created on Wed Jul 17 10:51:55 2024
5
6  @author: gargi
7  """
8
9  #!/usr/bin/env python3
10 # -*- coding: utf-8 -*-
11 """
12 Created on Thu Jul 11 12:05:00 2024
13
14 @author: user
15 """
16
17 from Bio.PDB.PDBParser import PDBParser
18 from Bio.SVDSuperimposer import SVDSuperimposer
19 import numpy as np
20 import matplotlib as mpl
21 import matplotlib.pyplot as plt
22
23 # Define amino acids
24 AA = ["ALA", "CYS", "ASP", "GLU", "PHE", "GLY", "HIS", "ILE", "LYS", "LEU", "MET",
25       "ASN", "PRO", "GLN", "ARG", "SER", "THR", "VAL", "TRP", "TYR"]
26
27 # Define PDBs
28 p = PDBParser(QUIET=True)
29 structure_files = ["", "", "", "", ""]
30 structures = [p.get_structure(f"ctxm_{i*10}c", file) for i, file in enumerate(
31     structure_files, start=1)]
32
33 # Superposition with SVD
34 def align(model1, model2, atom_types=["CA"]):
35     model1_coords = [a.coord for a in model1[0].get_atoms() if a.parent.resname in
36                      AA and a.name in atom_types]
37     model2_coords = [a.coord for a in model2[0].get_atoms() if a.parent.resname in
38                      AA and a.name in atom_types]
39     si = SVDSuperimposer()
```

```

36     si.set(np.array(model1_coords), np.array(model2_coords))
37     si.run()
38     return si
39
40 # Calculate RMSDs and populate the matrix
41 num_structures = len(structures)
42 C_aRMSD = np.zeros((num_structures, num_structures))
43
44 rmsds = []
45 for i in range(num_structures):
46     for j in range(i, num_structures):
47         si = align(structures[i], structures[j])
48         rmsd_value = si.get_rms()
49         rmsds.append(rmsd_value)
50         C_aRMSD[i, j] = rmsd_value
51         C_aRMSD[j, i] = rmsd_value # Symmetric matrix
52
53 formatted_rmsds = ['%.3f' % elem for elem in rmsds]
54 print(formatted_rmsds)
55
56 structures_labels = ["10 C ", "20 C ", "30 C ", "40 C ", "50 C "]
57
58 # Plotting
59 fig, ax = plt.subplots()
60 im = ax.imshow(C_aRMSD)
61
62 plt.rcParams["font.family"] = "arial"
63 mpl.rc('text.latex', preamble=r'\usepackage{amsmath}')
64
65 # Show all ticks and label them with the respective list entries
66 ax.set_xticks(np.arange(len(structures_labels)), labels=structures_labels, weight="
    bold")
67 ax.set_yticks(np.arange(len(structures_labels)), labels=structures_labels, weight="
    bold")
68
69 # Loop over data dimensions and create text annotations
70 for i in range(len(structures_labels)):
71     for j in range(len(structures_labels)):
72         ax.text(j, i, f'{C_aRMSD[i, j]:.3f}', ha="center", va="center", color="w",
            weight="bold")
73

```

```

74 ax.set_title(r'C$_{\mathbf{\alpha}}$ Pairwise RMSD : CTX-M-14', weight="bold")
75
76 cbar = plt.colorbar(im, ax=ax)
77 cbar.set_label(label='RMSD($\mathbf{AA}$)', rotation=-90.0, va="bottom", weight='
    bold')
78 cbar.ax.set_yticklabels(np.arange(0.00, 0.09, 0.01), fontweight='bold')
79
80 fig.tight_layout()
81 plt.rcParams['figure.dpi'] = 800
82 plt.rcParams['savefig.dpi'] = 800
83 plt.show()

```

Mathematica 10.4 code for the numerical calculations

```

1 Notebook[{
2 Cell[BoxData[
3   RowBox[{
4     RowBox[{{"(*",
5       RowBox[{"Spatial", " ", "domain", " ", "in", " ", "micrometers"}], "*)"}],
6     "\[IndentingNewLine]",
7     RowBox[{
8       RowBox[{
9         RowBox[{"size", "=", "50"}], ";"}], "\[IndentingNewLine]",
10      RowBox[{
11        RowBox[{
12          RowBox[{"{",
13            RowBox[{"xmin", " ", "xmax"}], "}"}, "=",
14          RowBox[{"{",
15            RowBox[{
16              RowBox[{
17                RowBox[{"-", "size"}], "/", "2"}], " ",
18                RowBox[{"size", "/", "2"}]}], "}"}}], ";"}], "\n",
19      RowBox[{
20        RowBox[{
21          RowBox[{"{",
22            RowBox[{"ymin", " ", "ymax"}], "}"}, "=",
23          RowBox[{"{",
24            RowBox[{
25              RowBox[{
26                RowBox[{"-", "size"}], "/", "2"}], " ",
27                RowBox[{"size", "/", "2"}]}], "}"}}], ";"}], "\[IndentingNewLine]",
28      "\[IndentingNewLine]",
29      RowBox[{{"(*",
30        RowBox[{"Thermal", " ", "diffusivities", " ", "in", " ",
31          RowBox[{
32            RowBox[{"\[Micro]m", "^", "2"}], "/", "ms"}]}], "*)"}],
33      "\[IndentingNewLine]",
34      RowBox[{
35        RowBox[{"\[Alpha]XI", "=", "140"}], ";"}], "\[IndentingNewLine]",
36      RowBox[{
37        RowBox[{"\[Alpha]CTX", "=", "140"}], ";"}], "\[IndentingNewLine]",
38      "\[IndentingNewLine]",
39      RowBox[{{"(*",

```

```

40     RowBox[{"Exposure", " ", "times", " ", "in", " ", "ms"}], "*)"}], "\n",
41 RowBox[{
42     RowBox[{"tExpXI", "=", "7"}], ";"}], "\[IndentingNewLine]",
43 RowBox[{
44     RowBox[{"tExpCTX", "=", "5"}], ";"}], "\[IndentingNewLine]", "\n",
45 RowBox[{"(*",
46     RowBox[{
47         RowBox[{"Start", "/", "Stop"}], " ", "time", " ", "in", " ", "ms"}],
48     "*)"}], "\n",
49 RowBox[{
50     RowBox[{"tStart", "=",
51         RowBox[{"-", "1"}]}], ";"}], "\[IndentingNewLine]",
52 RowBox[{
53     RowBox[{"tStop", "=", "15"}], ";"}], "\[IndentingNewLine]",
54 "\[IndentingNewLine]",
55 RowBox[{"(*",
56     RowBox[{
57         RowBox[{
58             "Beam", " ", "spatial", " ", "and", " ", "temporal", " ", "parameters"}],
59             ",", " ",
60             RowBox[{
61                 "conversion", " ", "from", " ", "FWHM", " ", "to", " ", "sigma"}]}],
62     "*)"}], "\[IndentingNewLine]",
63 RowBox[{
64     RowBox[{"sigmaX", "=",
65         RowBox[{"30", "/",
66             RowBox[{"(",
67                 RowBox[{"2",
68                     RowBox[{"Sqrt", "[",
69                         RowBox[{"2",
70                             RowBox[{"Log", "[", "2.", "]"}}}], "]"}}}], ")]"}], ";"}], "\n",
71 RowBox[{
72     RowBox[{"sigmaY", "=",
73         RowBox[{"10", "/",
74             RowBox[{"(",
75                 RowBox[{"2",
76                     RowBox[{"Sqrt", "[",
77                         RowBox[{"2",
78                             RowBox[{"Log", "[", "2.", "]"}}}], "]"}}}], ")]"}], ";"}], "\n",
79 RowBox[{
80     RowBox[{"aperture", "=", "10"}], ";"}], "\[IndentingNewLine]",

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81     "\[IndentingNewLine]",
82     RowBox[{{"(*",
83         RowBox[{{"Max", " ", "temperature", " ", "increase", " ", "at", " ", "center"}},
84         "*)"}], "\n",
85     RowBox[{{"deltaTempXI", "=", "20.12"}], ";"}], "\[IndentingNewLine]",
86     RowBox[{{"deltaTempCTX", "=", "11.24"}], ";"}], "\[IndentingNewLine]",
87     "\[IndentingNewLine]",
88     RowBox[{{"(*",
89         RowBox[{{"Smooth", " ", "rectangular", " ", "window", " ", "using", " ", "tanh",
90         RowBox[{{"(", ")"}]}], " ", "*)"}], "\[IndentingNewLine]",
91     RowBox[{{"SmoothWindow", "[",
92         RowBox[{{"x_", " ", "w_", " ", "s_"}], "]"}, ":=",
93     RowBox[{{"Tanh", "[",
94         RowBox[{{"(",
95         RowBox[{{"x", "+",
96         RowBox[{{"w", "/", "2"}]}], ")"}], "/", "s"}], "]"}, "/", "2"}],
97     "-",
98     RowBox[{{"Tanh", "[",
99         RowBox[{{"(",
100         RowBox[{{"x", "-",
101         RowBox[{{"w", "/", "2"}]}], ")"}], "/", "s"}], "]"}, "/",
102     "2"}]}], ";"}], "\[IndentingNewLine]",
103     RowBox[{{"(*",
104         RowBox[{{"Sharp", " ", "rectangular", " ", "window"}], ",", " ",
105         RowBox[{{"not", " ", "used"}]}], " ", "*)"}], "\n",
106     RowBox[{{"SharpWindow", "[",
107         RowBox[{{"x_", " ", "w_", " ", "s_"}], "]"}, ":=",

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122     RowBox[{ "DirichletWindow", "[",
123         RowBox[{ "x", "/", "w"}], "]" }], ";", " ", "\[IndentingNewLine]",
124     "\[IndentingNewLine]",
125     RowBox[{ "(*",
126         RowBox[{ "Time", "-",
127             RowBox[{ "and", " ", "space"}], "-",
128             RowBox[{ "dependent", " ", "beam"}]}], "*)" }, "\[IndentingNewLine]",
129     RowBox[{
130         RowBox[{
131             RowBox[{ "beamRaw", "[",
132                 RowBox[{ "x_", " ", "y_"}], "]" }, ":",
133             RowBox[{
134                 RowBox[{ "Exp", "[",
135                     RowBox[{
136                         RowBox[{ "-",
137                             RowBox[{ "x", "^", "2"}]}], "/",
138                         RowBox[{ "(",
139                             RowBox[{ "2", " ",
140                                 RowBox[{ "sigmaX", "^", "2"}]}], ")]"}], ")",
141                     RowBox[{ "Exp", "[",
142                         RowBox[{
143                             RowBox[{ "-",
144                                 RowBox[{ "y", "^", "2"}]}], "/",
145                             RowBox[{ "(",
146                                 RowBox[{ "2", " ",
147                                     RowBox[{ "sigmaY", "^", "2"}]}], ")]"}], ")]"}], ")",
148                     "\[IndentingNewLine]",
149                     RowBox[{
150                         RowBox[{
151                             RowBox[{ "beamAperture", "[",
152                                 RowBox[{ "x_", " ", "y_"}], "]" }, ":",
153                             RowBox[{
154                                 RowBox[{ "beamRaw", "[",
155                                     RowBox[{ "x", " ", "y"}], "]" }, ")",
156                                 RowBox[{ "SmoothWindow", "[",
157                                     RowBox[{
158                                         RowBox[{ "Sqrt", "[",
159                                             RowBox[{
160                                                 RowBox[{ "x", "^", "2"}], "+",
161                                                 RowBox[{ "y", "^", "2"}]}], ")]", " ", "aperture", " ", "0.2"}],
162                                     "]" }], ")",

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163 RowBox[{
164   RowBox[{
165     RowBox[{"temporalProfile", "["],
166     RowBox[{"t_", ",", "tExp_"}], "]"}, ":",
167     RowBox[{"SmoothWindow", "["],
168     RowBox[{
169       RowBox[{"t", "-"},
170       RowBox[{"tExp", "/", "2"}]}, ",", "tExp", ",", "0.05"}], "]"}}],
171 ";"], "\[IndentingNewLine]", "\[IndentingNewLine]",
172 RowBox[{"(*",
173   RowBox[{"Beam", " ", "shape", " ", "visualizations"}], "*)"},
174 "\[IndentingNewLine]",
175 RowBox[{
176   RowBox[{"P1", "="},
177   RowBox[{"Plot", "["],
178   RowBox[{
179     RowBox[{"{",
180     RowBox[{
181       RowBox[{"beamRaw", "["],
182       RowBox[{"x", ",", "0"}], "]"}, ",",
183       RowBox[{"beamAperture", "["],
184       RowBox[{"x", ",", "0"}], "]"}}], "}"}, ",",
185     RowBox[{"{",
186     RowBox[{"x", ",",
187       RowBox[{"-", "15"}], ",", "15"}], "}"}, ",", "\[IndentingNewLine]",
188     RowBox[{
189       "PlotLabel", "\[Rule]",
190       "\<Normalized beam profile along x at y=0>\\"}, ",",
191       RowBox[{"AxesLabel", "\[Rule]",
192       RowBox[{"{",
193       RowBox[{"\"\

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204         RowBox[{"\"\\<Raw beam\\>\"", ",", "\"\\<With aperture\\>\"", "}"}],
205         ",",
206         RowBox[{"Scaled", "["],
207         RowBox[{"{",
208         RowBox[{"{0.85", ",", "0.33"}}, "}"}, "]}]]], ",",
209         "\\[IndentingNewLine]",
210         RowBox[{"ImageSize", "\\[Rule]", "400"}]], "\\[IndentingNewLine]",
211         "]}]]], ";"}, "\\[IndentingNewLine]",
212 RowBox[{
213 RowBox[{"P2", "=",
214 RowBox[{"Plot", "[",
215 RowBox[{"{",
216 RowBox[{"{",
217 RowBox[{"{",
218 RowBox[{"beamRaw", "[",
219 RowBox[{"{0", ",", "y"}}, "]", ",",
220 RowBox[{"beamAperture", "[",
221 RowBox[{"{0", ",", "y"}}, "]}]]], "}"}, ",",
222 RowBox[{"{",
223 RowBox[{"y", ",",
224 RowBox[{"-", "15"}}, "]", ",", "15"}}, "]}]]], ",", "\\[IndentingNewLine]",
225 RowBox[{"{",
226 "PlotLabel", "\\[Rule]",
227 "\"\\<Normalized beam profile along y at x=0\\>\"", ",",
228 RowBox[{"AxesLabel", "\\[Rule]",
229 RowBox[{"{",
230 RowBox[{"{\\<y\\>", ",", "\\<Beam(0,y)\\>\"", "}"}, "]}]]], ",",
231 "\\[IndentingNewLine]",
232 RowBox[{"PlotRange", "\\[Rule]", "All"}], ",",
233 RowBox[{"PlotStyle", "\\[Rule]",
234 RowBox[{"{",
235 RowBox[{"{Dashed", ",", "Thick"}}, "]}]]], ",",
236 "\\[IndentingNewLine]",
237 RowBox[{"PlotLegends", "\\[Rule]",
238 RowBox[{"Placed", "[",
239 RowBox[{"{",
240 RowBox[{"{",
241 RowBox[{"{\\<Raw beam\\>", ",", "\"\\<With aperture\\>\"", "}"}],
242 ",",
243 RowBox[{"Scaled", "["],
244 RowBox[{"{",

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245         RowBox[{"0.85", ",", "0.66"}], "}]"}], "]}]]], ",",
246     "\[IndentingNewLine]",
247     RowBox[{"ImageSize", "\[Rule]", "400"}]], "\[IndentingNewLine]",
248     "]}]]], ";"}], "\n",
249 RowBox[{
250     RowBox[{"P3", "=",
251     RowBox[{"Plot", "[",
252     RowBox[{
253         RowBox[{"{",
254         RowBox[{
255             RowBox[{"temporalProfile", "[",
256             RowBox[{"t", ",", "tExpXI"}], "]", ",",
257             RowBox[{"temporalProfile", "[",
258             RowBox[{"t", ",", "tExpCTX"}], "]}]]], "}"}, ",",
259         RowBox[{"{",
260         RowBox[{"t", ",",
261         RowBox[{"-", "1"}], ",", "11"}], "}"}, ",", "\[IndentingNewLine]",
262         RowBox[{
263             "PlotLabel", "\[Rule]",
264             "\"\<Normalized temporal beam profile at center\>\"", ",",
265             "\[IndentingNewLine]",
266             RowBox[{"AxesLabel", "\[Rule]",
267             RowBox[{"{",
268             RowBox[{"\"\\<t>\"", ",", "\"\\<Beam(0,0,t)>\"", "}"}, "}"}, ",",
269             "\[IndentingNewLine]",
270             RowBox[{"AxesOrigin", "\[Rule]",
271             RowBox[{"{",
272             RowBox[{"tStart", ",", "0"}], "}"}, "}"}, ",", "\[IndentingNewLine]",
273             RowBox[{"PlotLegends", "\[Rule]",
274             RowBox[{"Placed", "[",
275             RowBox[{
276             RowBox[{"{",
277             RowBox[{"\"\\<XI: 7 ms>\"", ",", "\"\\<CTX: 5 ms>\"", "}"}, "}"}, ",",
278             RowBox[{"Scaled", "[",
279             RowBox[{"{",
280             RowBox[{"0.85", ",", "0.66"}], "}"}, "]}]]], "]}]]], ",",
281         "\[IndentingNewLine]",
282         RowBox[{"ImageSize", "\[Rule]", "400"}]], "\[IndentingNewLine]",
283         "]}]]], ";"}], "\[IndentingNewLine]", "\[IndentingNewLine]",
284 RowBox[{"(*",
285     RowBox[{"Pure", " ", "source", " ", "accumulation", " ",

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286     RowBox[{("(",
287         RowBox[{"no", " ", "diffusion"}], ")"}]], "*)"},
288     "\[IndentingNewLine]",
289     RowBox[{
290         RowBox[{"solutionXIstat", "=",
291             RowBox[{"NDSolveValue", "[",
292                 RowBox[{
293                     RowBox[{{"(",
294                         RowBox[{
295                             RowBox[{
296                                 RowBox[{"D", "[",
297                                     RowBox[{
298                                         RowBox[{"u", "[",
299                                             RowBox[{"x", " ", "y", " ", "t"}], "]"}], " ", "t"}], "]"}],
300                                         "\[Equal]",
301                                         RowBox[{
302                                             RowBox[{"beamAperture", "[",
303                                                 RowBox[{"x", " ", "y"}], "]"}], "*",
304                                                 RowBox[{"temporalProfile", "[",
305                                                     RowBox[{"t", " ", "tExpXI"}], "]"}],
306                                                 RowBox[{"deltaTempXI", "/", "tExpXI"}]]}], " ",
307                                         RowBox[{
308                                             RowBox[{"u", "[",
309                                                 RowBox[{"x", " ", "y", " ", "tStart"}], "]"}], "\[Equal]", "0"}]]],
310                                         ")}], " ", "u", " ",
311                                         RowBox[{{"(",
312                                             RowBox[{"x", " ", "xmin", " ", "xmax"}], ")}], " ",
313                                             RowBox[{{"(",
314                                                 RowBox[{"y", " ", "ymin", " ", "ymax"}], ")}], " ",
315                                                 RowBox[{{"(",
316                                                     RowBox[{"t", " ", "tStart", " ", "tStop"}], ")}]]], ")}]]], ";"},
317                                         "\[IndentingNewLine]",
318                                         RowBox[{
319                                             RowBox[{"solutionCTXstat", "=",
320                                                 RowBox[{"NDSolveValue", "[",
321                                                     RowBox[{
322                                                         RowBox[{{"(",
323                                                             RowBox[{
324                                                                 RowBox[{"D", "[",
325                                                                     RowBox[{

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327         RowBox[{"u", "["],
328         RowBox[{"x", ",", "y", ",", "t"}], "]"}, ",", "t"}], "]"},
329 "\[Equal]",
330 RowBox[{
331   RowBox[{"beamAperture", "["],
332   RowBox[{"x", ",", "y"}], "]"}, "*]",
333   RowBox[{"temporalProfile", "["],
334   RowBox[{"t", ",", "tExpCTX"}], "]"},
335   RowBox[{"deltaTempCTX", "/", "tExpCTX"}]]], ",",
336 RowBox[{
337   RowBox[{"u", "["],
338   RowBox[{"x", ",", "y", ",", "tStart"}], "]"}, "\[Equal]", "0"}]],
339 "}]"], ",", "u", ",",
340 RowBox[{"{",
341   RowBox[{"x", ",", "xmin", ",", "xmax"}], "}"}, ",",
342   RowBox[{"{",
343   RowBox[{"y", ",", "ymin", ",", "ymax"}], "}"}, ",",
344   RowBox[{"{",
345   RowBox[{"t", ",", "tStart", ",", "tStop"}], "}"}}], "]"}}], ";"}],
346 "\[IndentingNewLine]", "\[IndentingNewLine]",
347 RowBox[{"(*",
348   RowBox[{"Heat", " ", "diffusion", " ", "with", " ", "source"}], "*)"},
349 "\[IndentingNewLine]",
350 RowBox[{
351   RowBox[{"solutionXIdyn", "=",
352   RowBox[{"NDSolveValue", "["],
353   RowBox[{
354   RowBox[{"{",
355   RowBox[{
356   RowBox[{
357   RowBox[{"D", "["],
358   RowBox[{
359   RowBox[{"u", "["],
360   RowBox[{"x", ",", "y", ",", "t"}], "]"}, ",", "t"}], "]"},
361 "\[Equal]",
362   RowBox[{
363   RowBox[{"\[Alpha]XI", "*]",
364   RowBox[{"Laplacian", "["],
365   RowBox[{
366   RowBox[{"u", "["],
367   RowBox[{"x", ",", "y", ",", "t"}], "]"}, ",",

```

```

368         RowBox[{"{",
369             RowBox[{"x", ",", "y"}], "}"}, {"x", "y"}], "+",
370     RowBox[{
371         RowBox[{"beamAperture", "["],
372         RowBox[{"x", ",", "y"}], "]", "*"},
373         RowBox[{"temporalProfile", "["],
374         RowBox[{"t", ",", "tExpXI"}], "]",
375         RowBox[{"deltaTempXI", "/", "tExpXI"}]}], ",",
376     RowBox[{
377         RowBox[{"u", "["],
378         RowBox[{"x", ",", "y", ",", "tStart"}], "]", "\[Equal]", "0"}],
379     ",", " ",
380     RowBox[{"DirichletCondition", "["],
381     RowBox[{
382         RowBox[{
383             RowBox[{"u", "["],
384             RowBox[{"x", ",", "y", ",", "t"}], "]", "\[Equal]", "0"}], ",",
385         RowBox[{
386             RowBox[{"Abs", "["],
387             RowBox[{
388                 RowBox[{"x", "^", "2"}], "+",
389                 RowBox[{"y", "^", "2"}], "-"},
390             RowBox[{
391                 RowBox[{"(",
392                 RowBox[{"size", "/", "2"}], ")", "^", "2"}], ")",
393                 "0.1"}], "]", "u", ",",
394         RowBox[{"{",
395             RowBox[{"x", ",", "xmin", ",", "xmax"}], "}"}, " ",
396         RowBox[{"{",
397             RowBox[{"y", ",", "ymin", ",", "ymax"}], "}"}, " ",
398         RowBox[{"{",
399             RowBox[{"t", ",", "tStart", ",", "tStop"}], "}"}, " ",
400         RowBox[{"Method", "\[Rule]",
401         RowBox[{"{",
402             RowBox[{"\<MethodOfLines\>", ",",
403             RowBox[{"\<SpatialDiscretization\>", "\[Rule]",
404             RowBox[{"{",
405             RowBox[{"\<FiniteElement\>", ",",
406             RowBox[{"\<MeshOptions\>", "\[Rule]",
407             RowBox[{"{",
408             RowBox[{"\<MaxCellMeasure\>", "\[Rule]", "0.5"}],

```

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409         "}}}}}}", "}}}}}}", "}}}}}}", "}}}}", ";"},
410     "\[IndentingNewLine]",
411     RowBox[{
412         RowBox[{"solutionCTXdyn", "=",
413         RowBox[{
414             RowBox[{
415                 RowBox[{"{",
416                 RowBox[{
417                     RowBox[{
418                         RowBox[{"D", "[",
419                         RowBox[{
420                             RowBox[{"u", "[",
421                             RowBox[{"x", ",", "y", ",", "t"}], ""]}, ",", "t"}], ""]},
422                     "\[Equal]",
423                     RowBox[{
424                         RowBox[{"\[Alpha]CTX", "*",
425                         RowBox[{"Laplacian", "[",
426                         RowBox[{
427                             RowBox[{"u", "[",
428                             RowBox[{"x", ",", "y", ",", "t"}], ""]}, ",",
429                             RowBox[{"{",
430                             RowBox[{"x", ",", "y"}], "}}}}], ""]}}}, "+",
431                         RowBox[{
432                             RowBox[{"beamAperture", "[",
433                             RowBox[{"x", ",", "y"}], ""]}, "*",
434                             RowBox[{"temporalProfile", "[",
435                             RowBox[{"t", ",", "tExpCTX"}], ""]},
436                             RowBox[{"deltaTempCTX", "/", "tExpCTX"}]}]}]}], ",",
437                     RowBox[{
438                         RowBox[{"u", "[",
439                         RowBox[{"x", ",", "y", ",", "tStart"}], ""]}, "\[Equal]", "0"}],
440                     ",",
441                     RowBox[{"DirichletCondition", "[",
442                     RowBox[{
443                         RowBox[{
444                             RowBox[{"u", "[",
445                             RowBox[{"x", ",", "y", ",", "t"}], ""]}, "\[Equal]", "0"}], ",",
446                         RowBox[{
447                             RowBox[{"Abs", "[",
448                             RowBox[{"x", "^", "2"}], ""]}, "+",

```

```

450         RowBox[{"y", "^", "2"}], "-",
451         RowBox[{
452             RowBox[{"(",
453                 RowBox[{"size", "/", "2"}], ")"}], "^", "2"}], "]", "<",
454             "0.1"}], "}")], "u", "",
455         RowBox[{"{",
456             RowBox[{"x", "", "xmin", "", "xmax"}], "}"}, "",
457         RowBox[{"{",
458             RowBox[{"y", "", "ymin", "", "ymax"}], "}"}, "",
459         RowBox[{"{",
460             RowBox[{"t", "", "tStart", "", "tStop"}], "}"}, "",
461         RowBox[{"Method", "\[Rule]",
462             RowBox[{"{",
463                 RowBox[{"\"\\<MethodOfLines\\>\"", "",
464                     RowBox[{"\"\\<SpatialDiscretization\\>\"", "\[Rule]",
465                         RowBox[{"{",
466                             RowBox[{"\"\\<FiniteElement\\>\"", "",
467                                 RowBox[{"\"\\<MeshOptions\\>\"", "\[Rule]",
468                                     RowBox[{"{",
469                                         RowBox[{"\"\\<MaxCellMeasure\\>\"", "\[Rule]", "0.5"}],
470                                         "}"}}]]], "}"}}]]], "}"}}], ";"}],
471         "\[IndentingNewLine]", "\[IndentingNewLine]",
472         RowBox[{"(*", "Export", "*)"}, "\[IndentingNewLine]",
473         RowBox[{
474             RowBox[{"Export", "["},
475             RowBox[{
476                 RowBox[{"FileNameJoin", "["},
477                 RowBox[{"{",
478                     RowBox[{
479                         RowBox[{"NotebookDirectory", "["}, "]", "]", "",
480                         "\"\\<beam profile X.txt\\>\"", " ", "}"}, "]", "]", "",
481                     RowBox[{"N", "@",
482                         RowBox[{"Round", "["},
483                         RowBox[{
484                             RowBox[{"Table", "["},
485                             RowBox[{
486                                 RowBox[{"{",
487                                     RowBox[{"x", "",
488                                         RowBox[{"beamRaw", "["},
489                                         RowBox[{"x", "", "0"}], "]", "]", "",
490                                         RowBox[{"beamAperture", "["},

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491         RowBox[{"x", ",", "0"}], ""]}], "}], "],
492     RowBox[{"{",
493         RowBox[{"x", ",",
494             RowBox[{"-", "15"}], ",", "15", ",", "0.01"}], ""]}],
495     ",",
496     RowBox[{"1", "/", "1000"}]], ""]}], ""];
497 "\[IndentingNewLine]",
498 RowBox[{
499     RowBox[{"Export", "[",
500         RowBox[{
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502                 RowBox[{"{",
503                     RowBox[{"NotebookDirectory", "[", "]", "],",
504                     "\<beam profile Y.txt\>"}], " ", ""]}], "],",
505             RowBox[{"N", "@",
506                 RowBox[{"Round", "[",
507                     RowBox[{
508                         RowBox[{"Table", "[",
509                             RowBox[{
510                                 RowBox[{"{",
511                                     RowBox[{"{",
512                                         RowBox[{"y", ",",
513                                             RowBox[{"beamRaw", "[",
514                                                 RowBox[{"0", ",", "y"}], "]", "],",
515                                                 RowBox[{"beamAperture", "[",
516                                                     RowBox[{"0", ",", "y"}], ""]}], ""]}], "],",
517                                     RowBox[{"{",
518                                         RowBox[{"y", ",",
519                                             RowBox[{"-", "15"}], ",", "15", ",", "0.01"}], ""]}],
520                                     ",",
521                                     RowBox[{"1", "/", "1000"}]], ""]}], ""];
522 "\[IndentingNewLine]",
523 RowBox[{
524     RowBox[{"Export", "[",
525         RowBox[{
526             RowBox[{"FileNameJoin", "[",
527                 RowBox[{"{",
528                     RowBox[{"NotebookDirectory", "[", "]", "],",
529                     "\<beam profile t.txt\>"}], " ", ""]}], "],",
530             RowBox[{"N", "@",

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532     RowBox[{"Round", "["],
533     RowBox[{
534       RowBox[{"Table", "["],
535       RowBox[{
536         RowBox[{"{",
537         RowBox[{"t", ",",
538         RowBox[{"temporalProfile", "[",
539         RowBox[{"t", ",", "tExpXI"}], ""]}, ",",
540         RowBox[{"temporalProfile", "[",
541         RowBox[{"t", ",", "tExpCTX"}], ""]}]}, "}]"}, ",",
542       RowBox[{"{",
543       RowBox[{"t", ",",
544       RowBox[{"-", "1"}], ",", "11", ",", "0.01"}], ""]}]}, ""]},
545       ",",
546       RowBox[{"1", "/", "1000"}]}}], ""]}]}, ""]}, ";"},
547     "\[IndentingNewLine]",
548     RowBox[{
549       RowBox[{"Export", "["],
550       RowBox[{
551         RowBox[{"FileNameJoin", "["],
552         RowBox[{"{",
553         RowBox[{
554           RowBox[{"NotebookDirectory", "[", ""]}, ",",
555           "\"\\<heatexchange_ms_K_alpha140.txt\\>\""}, " "], "}]"}, ",",
556         RowBox[{"N", "@"],
557         RowBox[{"Round", "["],
558         RowBox[{
559           RowBox[{"Table", "["],
560           RowBox[{
561             RowBox[{"{",
562             RowBox[{"t", ",",
563             RowBox[{"solutionXIstat", "[",
564             RowBox[{"0", ",", "0", ",", "t"}], ""]}, ",",
565             RowBox[{"solutionXIIdyn", "[",
566             RowBox[{"0", ",", "0", ",", "t"}], ""]}, ",",
567             RowBox[{"solutionCTXstat", "[",
568             RowBox[{"0", ",", "0", ",", "t"}], ""]}, ",",
569             RowBox[{"solutionCTXIdyn", "[",
570             RowBox[{"0", ",", "0", ",", "t"}], ""]}]}, "}]"}, ",",
571             RowBox[{"{",
572             RowBox[{"t", ",", "tStart", ",", "tStop", ",", "0.01"}], ""]}]},

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573         "]""]], ",",
574         RowBox[{{"1", "/", "1000"}]}], "]""]]]], ";""]]]], "Input",
575 CellChangeTimes->{{3.9569200916785126*^9, 3.956920101102765*^9}, {
576   3.9569201661364965*^9, 3.9569201929664497*^9}, {3.9569202292615585*^9,
577   3.956920258230592*^9}, 3.9569842602131667*^9, {3.9569843921419907*^9,
578   3.9569844036068506*^9}, {3.9569844549562674*^9,
579   3.9569844611040573*^9}, {3.956984613293849*^9, 3.956984615722562*^9}, {
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