Pushing the Limits of Microfocus X-ray Sources for Biological Crystallography

J. Wiesmann[1], J. Graf[1], S. Freisz[2], V. Smith[2], M. Benning[3], A. Kleine[1], C. Michaelsen[1]

[1] Incoatec GmbH, 21502 Geesthacht, Germany; [2] Bruker AXS GmbH, 76187 Karlsruhe, Germany; [3] Bruker AXS Inc., Madison (WI), USA

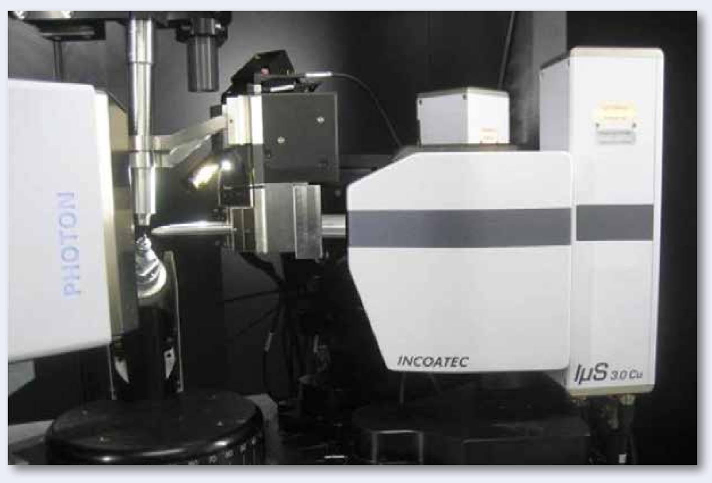
Introduction

Modern low power microfocus X-ray sources, such as the Incoatec Microfocus Soure IμS or the METALJET X-ray source, define the state-of-the-art for most in-house applications in X-ray diffraction. These sources deliver a brightness beyond that of comparable traditional X-ray sources at power settings below 1 kW. They are usually combined with multilayer X-ray mirrors which are excellent X-ray optical devices for beam shaping and preserving the brightness of the source.

The Incoatec Microfocus Source IμS

Since its introduction in 2006, the Incoatec Microfocus Source IμS has become the gold standard for low-power low-maintenance X-ray sources for the home-lab. The IμS combines a low-power microfocus X-ray sealed tube with dedicated Montel multilayer mirrors and delivers intensities beyond those of traditional rotating anode sources. With more than 700 sources sold world-wide, the IμS is the market-leading microfocus source for X-ray diffraction applications, such as single crystal diffraction on small molecule and protein crystals as well as small angle scattering.

- Low-power microfocus sealed tube
- Air-cooled
- Operated typically at 50 W
- Power load ~ 5 kW/mm²
- Montel multilayer mirrors for focused or collimated beam applications
- Available for Cu-Kα, Mo-Kα, Ag-Kα (others on request)
- Single port source



The IμS 3.0 - The New Micorofocus X-ray Tube with IXT

The latest generation of the IμS, the air-cooled IμS 3.0 source, contains the new Incoatec X-ray Tube *IXT* and is the first and only microfocus X-ray source that is designed for X-ray diffraction. Numerous small improvements make the IμS 3.0 the most user-friendly, yet most powerful microfocus X-ray source ever.

IμS 3.0

Incoatec Microfocus Source

NEW *with IXT*

- New X-ray tube IXT designed by Incoatec
- First microfocus source optimized for X-ray diffraction
- New beam path concept with true downstream alignment
- He filled optics housing and pre-aligned optics
- Swappable optics (Quick-lock concept)
- Improved user-friendliness
- New high-performance HV generator
- For Cu, Mo and Ag radiation
- 3 years warranty

30% more intensity

Improved Data Quality by the Cu-IμS 3.0

Absolute structure determination for a real life example: Diphenyloxazolidin-2-on Derivate $C_{25}H_{31}NO_5$, 0.12 x 0.10 x 0.09 mm³

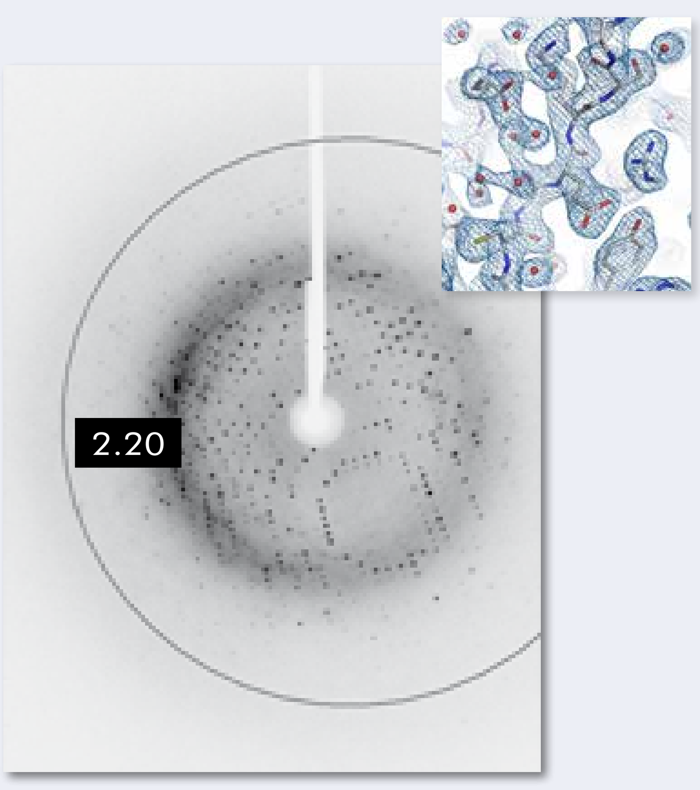
Source	Cu-IμS High Brilliance	Cu-IμS 3.0
Detector	Photon 100	Photon 100
Exposure time [s/°]	4 - 12; 5 h	4 - 12; 5 h
Resolution [Å]	0.80 (0.90 - 0.80)	0.80 (0.90 - 0.80)
Multiplicity	10.8 (7.5)	10.7 (7.5)
<I/σ>	27.7 (11.7)	32.2 (14.8)
*R*1, *wR*2 [%]	3.88, 9.88	3.61, 9.20
Parsons *z*(*v*)	0.07(9)	0.05(7)
Flack *x*(*u*)	0.00(24)	0.04(22)

Data statistics (left) and typical diffraction pattern (above) of the diphenyloxazolidin-2-on derivate ($P2_12_12_1$, a = 8.4167(17) Å, b = 13.761(3) Å, c = 19.304(4) Å, Z = 4).

Protein Crystallography with the Cu-IμS 3.0

215° data collection on a thin crystal of human NEIL1 (Endonuclease VIII-like protein)

Crystal Size [mm³]	0.15 x 0.12 x 0.02
Exposure time [s/0.5°]	100
Total time [h]	12
Resolution [Å]	2.25 (2.32 - 2.25)
Completeness [%]	99.6 (99.7)
Multiplicity	5.1 (3.5)
<I/σ(I)>	13.2 (2.6)
R_{merge}	0.0707 (0.3234)
$CC_{1/2}$ at High res [%]	89
R; R_{free}	0.196; 0.250

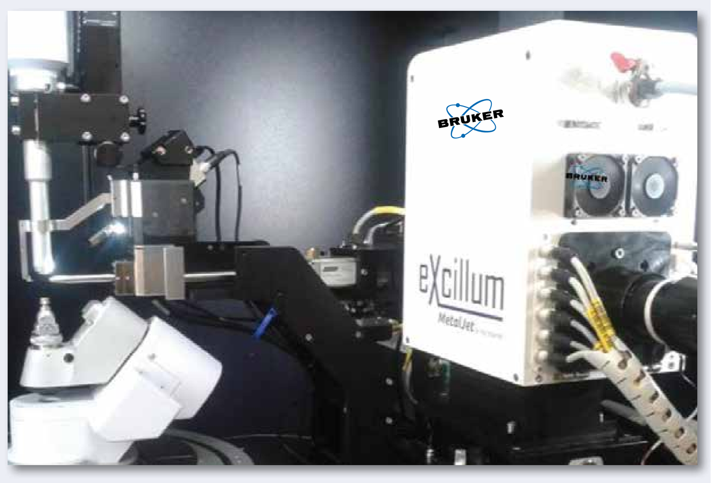


Data statistics (above, left), typical diffraction pattern (above, right) and section of the electron density map (right) of the human NEIL1 protein after structure solution by molecular replacement and refinement with REFMAC.

The METALJET X-ray Source

The brilliance of conventional X-ray sources using a solid metal target is limited by the maximum power load that can be applied without melting the anode material. The METALJET X-ray source technology has overcome this limitation, as it uses molten alloys containing Gallium and Indium as a high-speed liquid metal-jet target instead of fast spinning solid metal targets. Therefore, power loads of more than 100 kW/mm² in a spot size of 20 μm or smaller can be applied, which are an order of magnitude larger than those of modern microfocus rotating anodes.

- High-speed liquid metal-jet target
- Air-cooled
- Operated typically at 200 W
- Power load > 100 kW/mm²
- Montel multilayer mirrors for focused or collimated beam applications
- Available for Ga-Kα (1.34 Å), In-Kα (0.51 Å)
- Dual port source



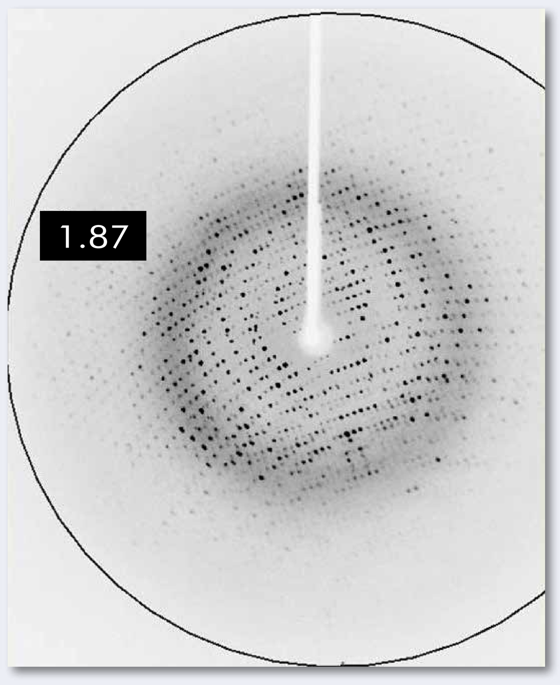
For preserving the extreme brightness of the source, multilayer mirrors of highest quality are needed, since a smaller source requires greater precision in the manufacturing of the multilayers. The Montel mirrors for Ga-Kα (9.3 keV) and In-Kα (24.2 keV) are synchrotron-class optics tailor-made for the METALJET source for applications, such as single crystal diffraction or small angle scattering. As the energy of the Ga-Kα line is close to the Cu-Kα line, the METALJET using a Gallium rich alloy as target is the source of choice for ultimate performance in the home lab for applications such as protein and pharmaceutical crystallography, as well as small angle scattering. The Montel optics for Ga-Kα radiation deliver an intense X-ray beam that is much brighter than what is currently achieved with microfocus rotating anode sources.

By using an Indium rich target, the yield from the In-Kα line, which is close to the Ag-Kα line, can be maximized, giving highest performance for diffraction applications on absorbing materials and for high-pressure experiments using DAC's.

Fast Data Collection for High Throughput Screening

100° Data Set from a Protein Complex within 4 min

Crystal Size [mm³]	0.10 x 0.80 x 0.05
Exposure time [s/°]	2
Resolution [Å]	1.95 (2.05 - 1.95)
Completeness (%)	97.5 (92.2)
Multiplicity	3.68 (2.34)
<I/σ(I)>	10.75 (2.77)
R_{merge} (%)	6.58 (23.91)
R_{sigma} (%)	5.99 (25.36)
R_{pim} (%)	3.58 (17.95)
CC(½) at 2.00 Å	86 %

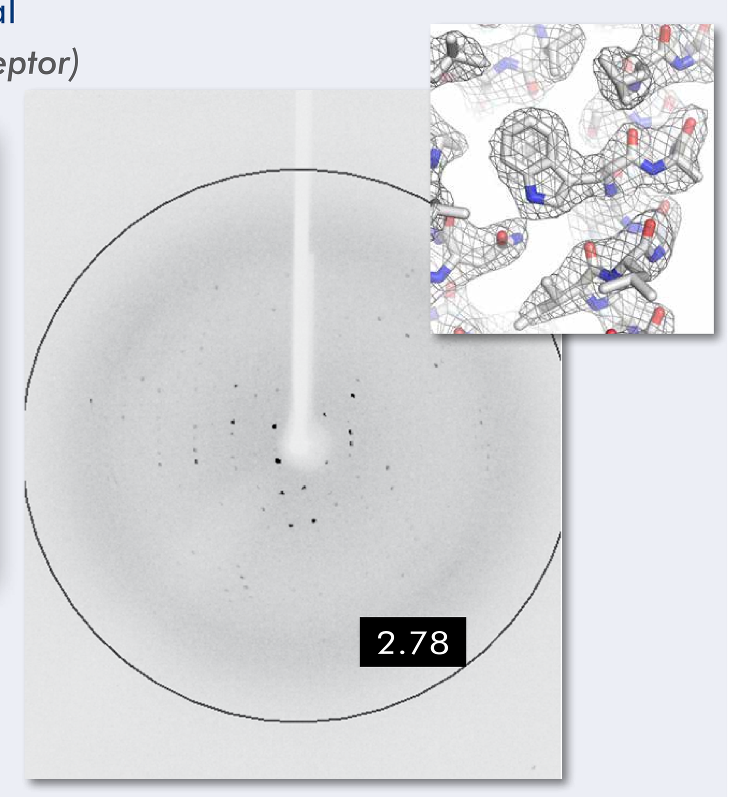


Data statistics and typical diffraction pattern of a protein complex, recorded with a D8 VENTURE 2nd Gen. and the METALJET X-ray source using a Ga rich alloy. The good data quality allowed for a structure solution by molecular replacement.

Data from a Small and Weak Diffracting Protein Crystal

2.5 data collection on a small crystal of a GPCR (Orexin receptor)

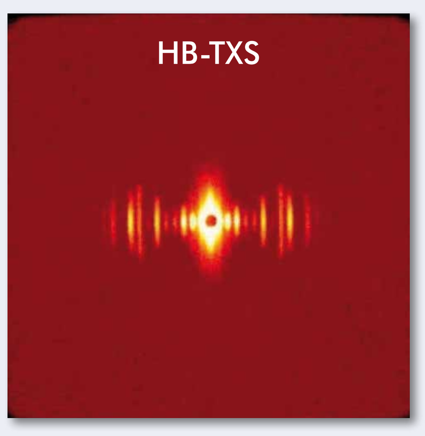
Crystal Size [mm³]	0.08 x 0.08 x 0.05
Exposure time [s/°]	60
Resolution [Å]	2.77 (2.87 - 2.77)
Completeness [%]	99.3 (97.9)
Multiplicity	3.2 (2.6)
<I/σ>	8.1 (1.1)
R_{pim} [%]	7.62(45.68)
$CC_{1/2}$ at High res [%]	89
R; R_{free}	0.244; 0.272



Data statistics (above, left), typical diffraction pattern (above, right) and section of the electron density map (right) of the GPCR crystal after structure solution by molecular replacement and refinement with REFMAC.

SAXS Data from a Thin Fiber of a Rat Tail Tendon

Performance comparison with METALJET and a microfocus rotating anode



SAXS scattering plots of a very thin fiber from a rat tail tendon, measured with a Bruker AXS NANOSTAR equipped with the METALJET X-ray source (200 W at 70 kV) and the microfocus rotating anode TXS (2.5 W at 50 kV). Both use the same type of parallel Montel optics. The METALJET delivers 3x higher intensity.

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