



| The European Synchrotron



# cryo-EM at the ESRF

current status and future developments

Christoph Mueller-Dieckmann

## 13 Member states:

France	27,5 %
Germany	24 %
Italy	13,2 %
United Kingdom	10,5 %
Russia	6 %
Benesync (Belgium, The Netherlands)	5,8 %
Nordsync (Denmark, Finland, Norway, Sweden)	5 %
Spain	4 %
Switzerland	4 %

Contribution to the budget in %

## 9 Associate countries:

Israel	1,75 %
Austria	1,75 %
Centralsync (Czech Republic, Hungary, Slovakia)	1,05 %
Poland	1 %
Portugal	1 %
India	0,66 %
South Africa	0,3 %



**22 partner nations**

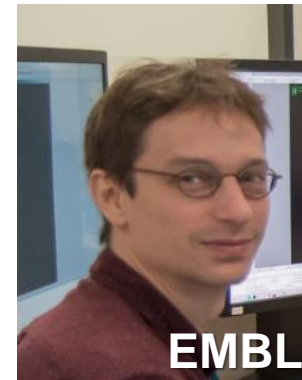
**Members of staff: 630 of 40 different nationalities**

**Legal status: Private civil company subject to French law**





Titan Krios G3  
Quantum LS energy filter  
K2 summit direct detector  
Volta phase plate  
300 kV





Operational since 11/2017

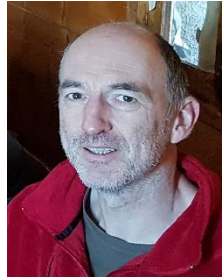
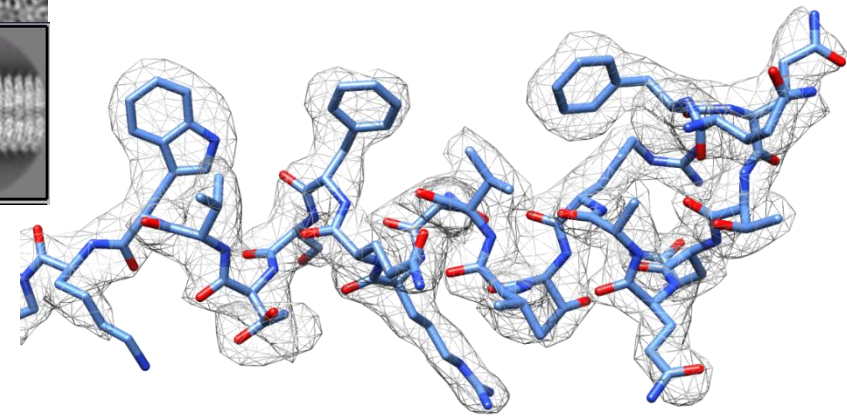
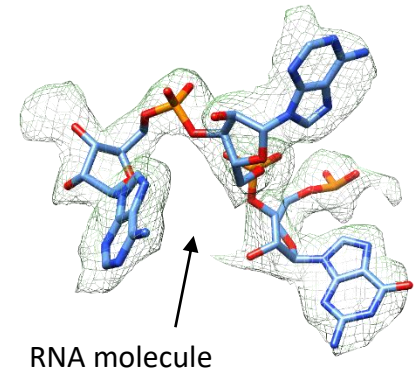
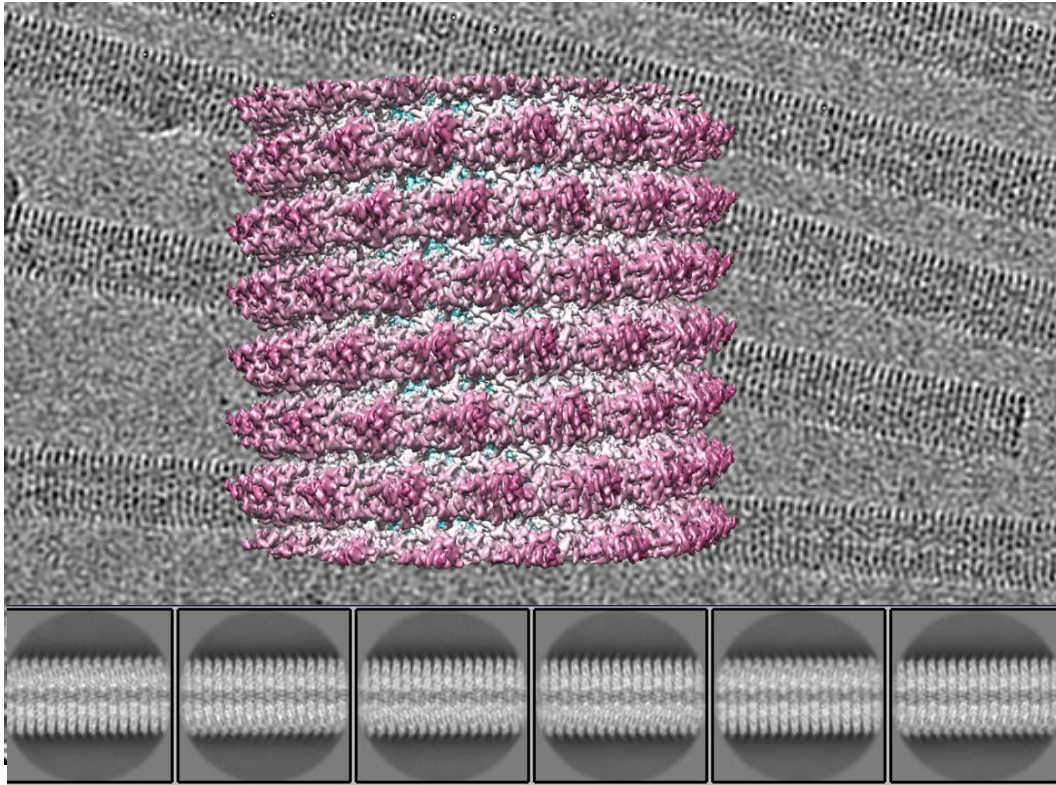
Complement to MX

SP only

In house Research programme

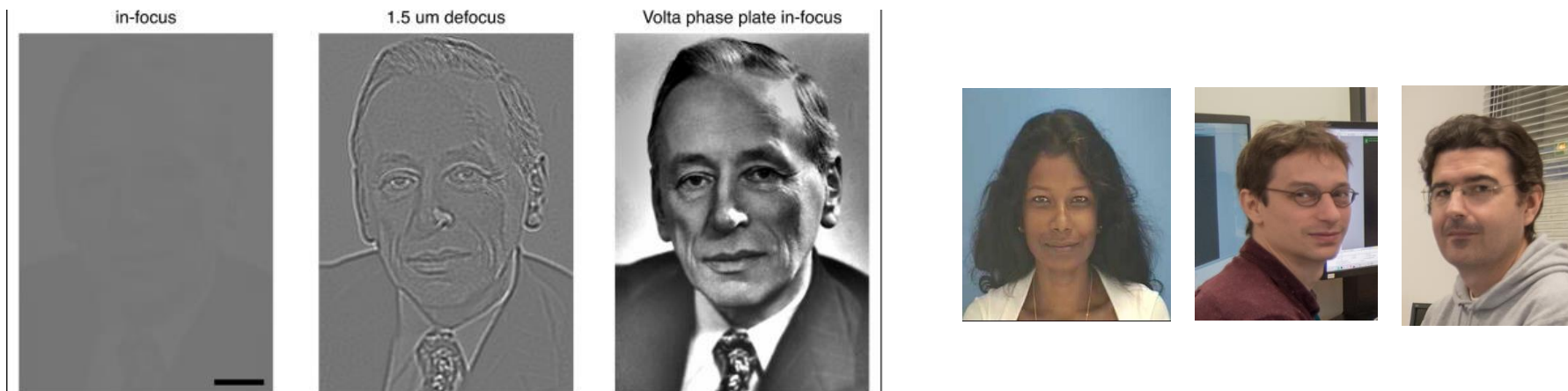


- **USER SUPPORT**
- In-house Research
  - Benchmarking of Titan Krios and its support with test samples to push the resolution limit to the maximum possible.
  - Characterization of Volta phase plate using standard proteins of smaller size, of the range of less than 100kDa.
  - Implementation and testing other software such as SerialEM for automatic data collection to improve high throughput data collection.

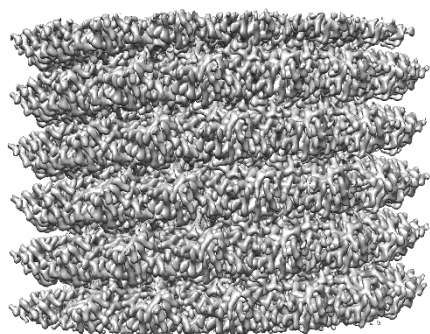




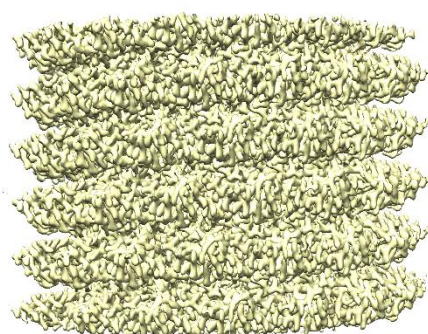
## Volta Phase Plate



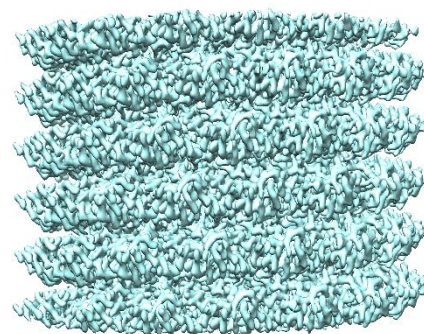
Danev & Baumeister (2017). *Current Opinion in Structural Biology* **46**, 87–94



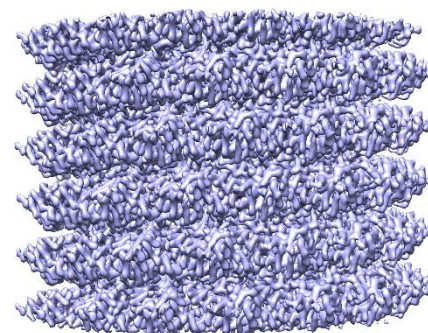
All micrographs  
78637 particles  
Res = 3.28Å



Mic 69 - 300  
26355 particles  
Res = 3.15Å



Mic 302 - 458  
26207 particles  
Res = 3.34Å



Mic 459 - 587  
26355 particles  
Res = 3.59Å



- Single particle experiments only
- Rolling access proposals at any time
- Beam time granted by BTAP
- Pre-characterised samples only
- 2 experiments per week
- 2 reimbursed user per experiment (or dewars)
- Experimental support by 3 scientists (ESRF, EMBL, IBS)
- [http://www.esrf.eu/home/UsersAndScience/Experiments/MX/About\\_our\\_beamlines/CM01.html](http://www.esrf.eu/home/UsersAndScience/Experiments/MX/About_our_beamlines/CM01.html)



User office  
BTAP  
Safety

External proposal



BTAP evaluation



Approved proposal



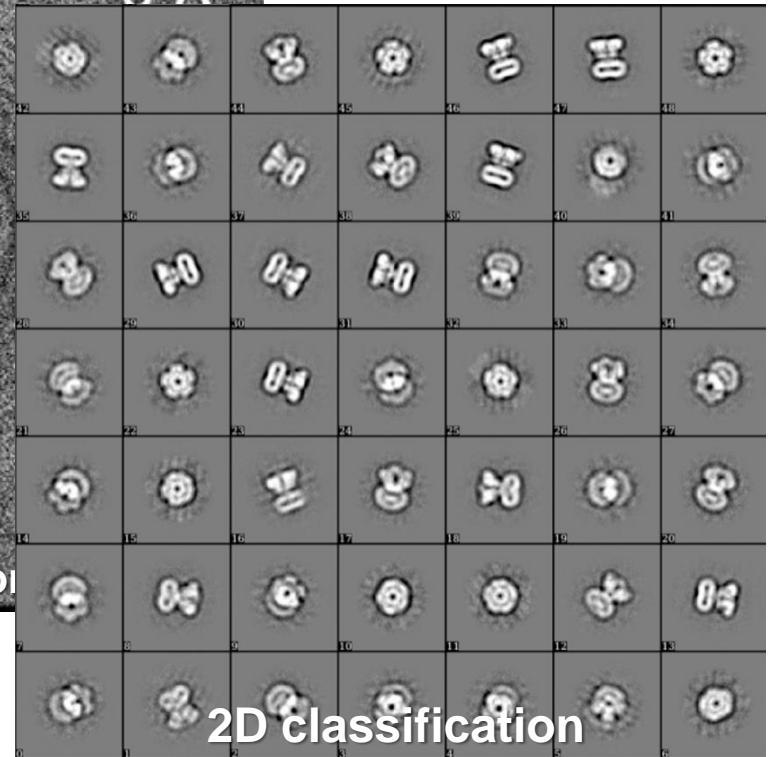
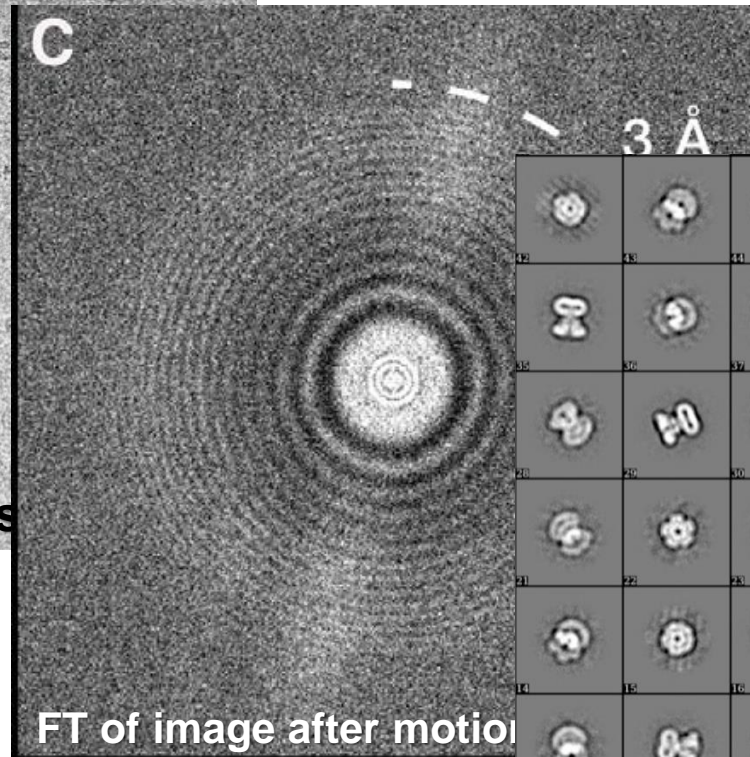
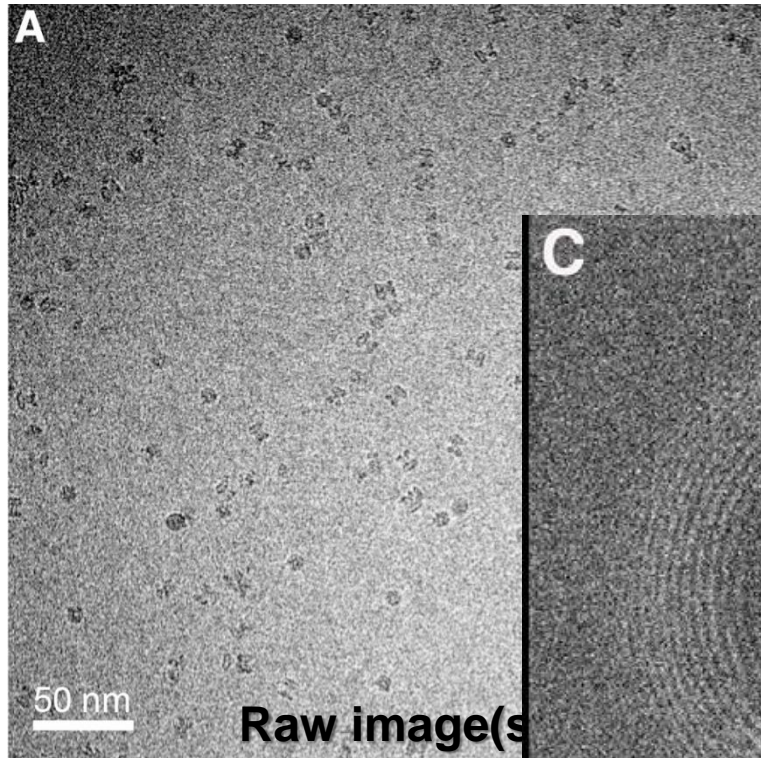
**Pre-evaluated project**  
(sample preparation  
established)



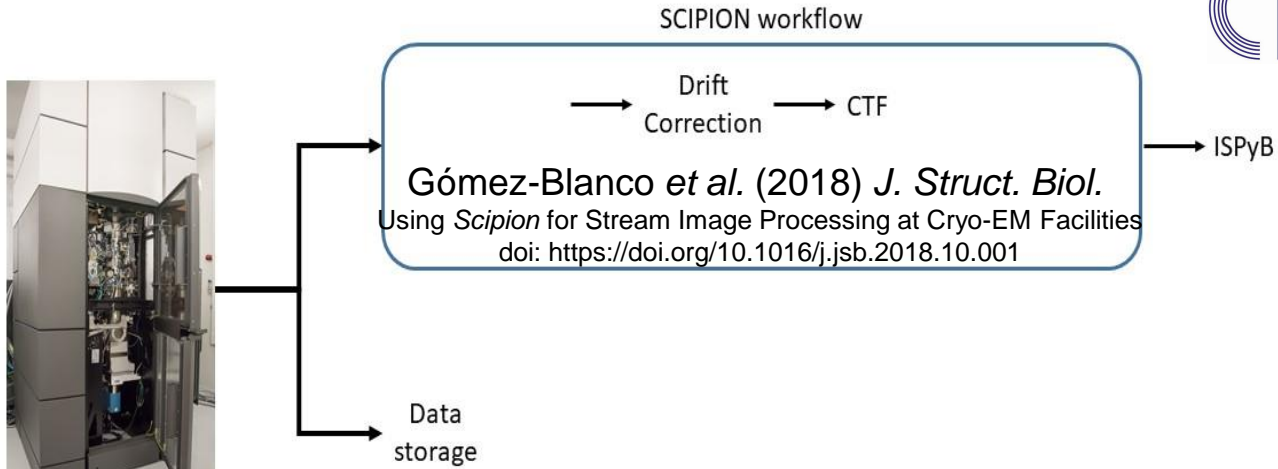
Grids  
available



KRIOS



# (CURRENT) AUTOMATIC DATA PRE-PROCESSING



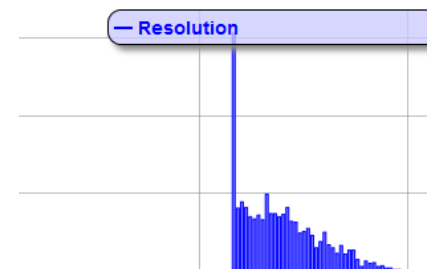
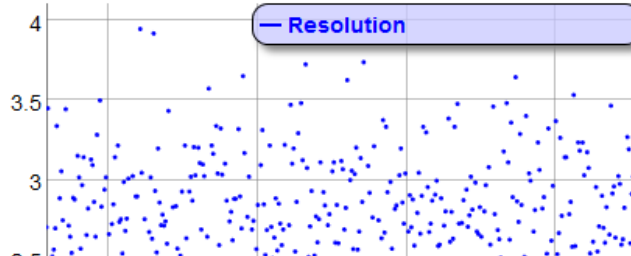
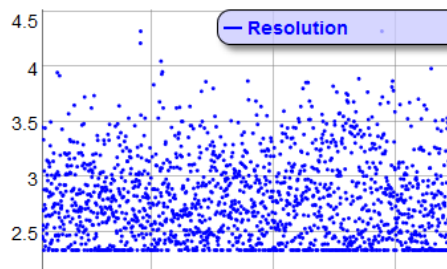
<p>Novia #30631</p> <p>Number: 30631</p> <p>X, Y: -0.000364, 0.000903</p>	<p>Motion Correction</p> <p>Total Motion: 51.2</p> <p>Avg. Motion/frame: 1.3</p> <p>Frame Range: 1 - 40</p> <p>Dose: 0.7500</p>	<p>CTF</p> <p>Resolution Limit: 2.936</p> <p>Correlation: 0.114407</p> <p>Defocus U: 13594.68</p> <p>Defocus V: 13852.07</p> <p>Angle: 19.90</p> <p>Estimated B factor: 89.48</p>
<p>Novia #30630 14-05-2018 02:18:31</p> <p>Number: 30630</p> <p>X, Y: -0.000368, 0.000905</p>	<p>Alignment based upon</p>	<p>Instrument MRC <a href="#">XML</a></p> <p>CTF</p> <p>Angle: 55.94</p> <p>Estimated B factor: 90.57</p>
<p>Novia #30629 14-05-2018 02:18:31</p> <p>Number: 30629</p> <p>X, Y: -0.000368, 0.000905</p>		<p>Instrument MRC <a href="#">XML</a></p> <p>CTF</p> <p>Angle: 26.07</p> <p>Estimated B factor: 89.40</p>

# STATS FOR AUTOMATIC DATA PRE-PROCESSING

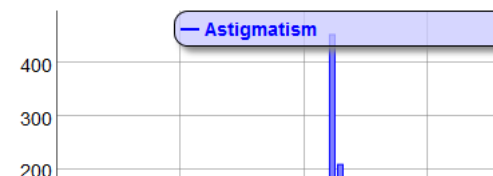
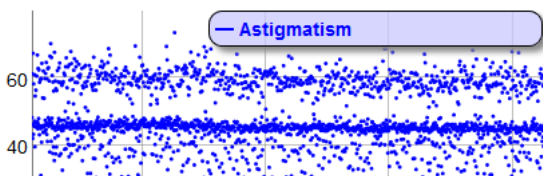
13 Data Collections

Stats

## Resolution



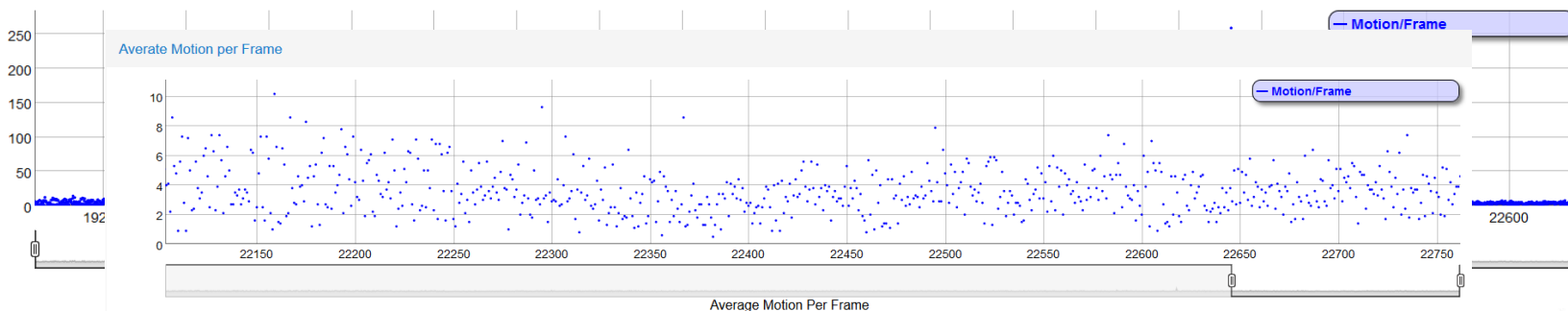
## Astigmatism



## Defocus



## Average Motion per Frame





- Robust sample preparation (e.g. blot-less techniques)
- On-the-fly data (pre-)processing and quality indicator(s)
- Automated pipelines as much as possible
- RA for experienced users
- Training
- Unified data policy

1<sup>st</sup> sample preparation workshop 18-19/09/2018







## Sample preparation

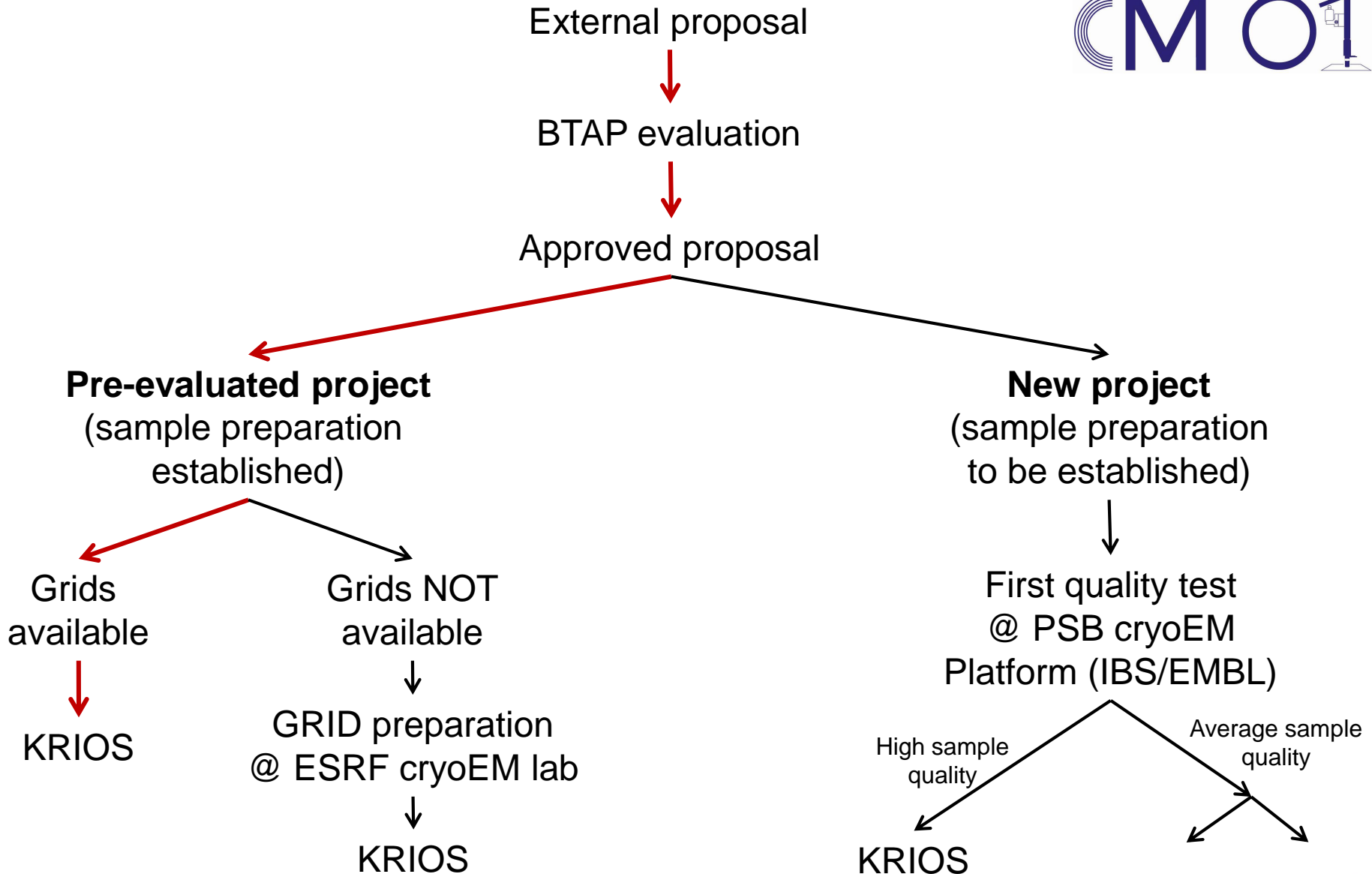


## Sample storage



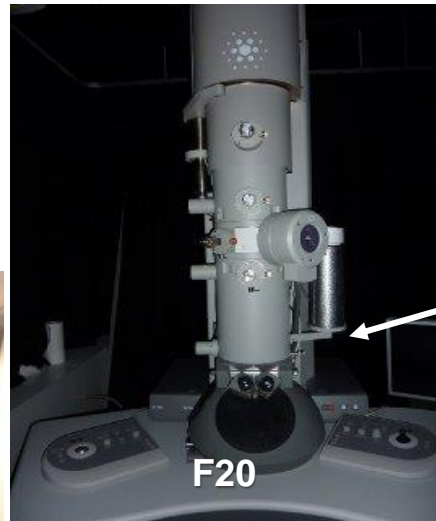
## Future developments

- Increase mail-in service; remote experiments
- Increase performance (speed)
- Extend data processing pipeline(s)
- Increase IT infrastructure
- Sample preparation
- Extend service



IBS

<http://www.ibs.fr/research/research-groups/methods-and-electron-microscopy-group/>



cryo-SP and tomo



cryo-SP

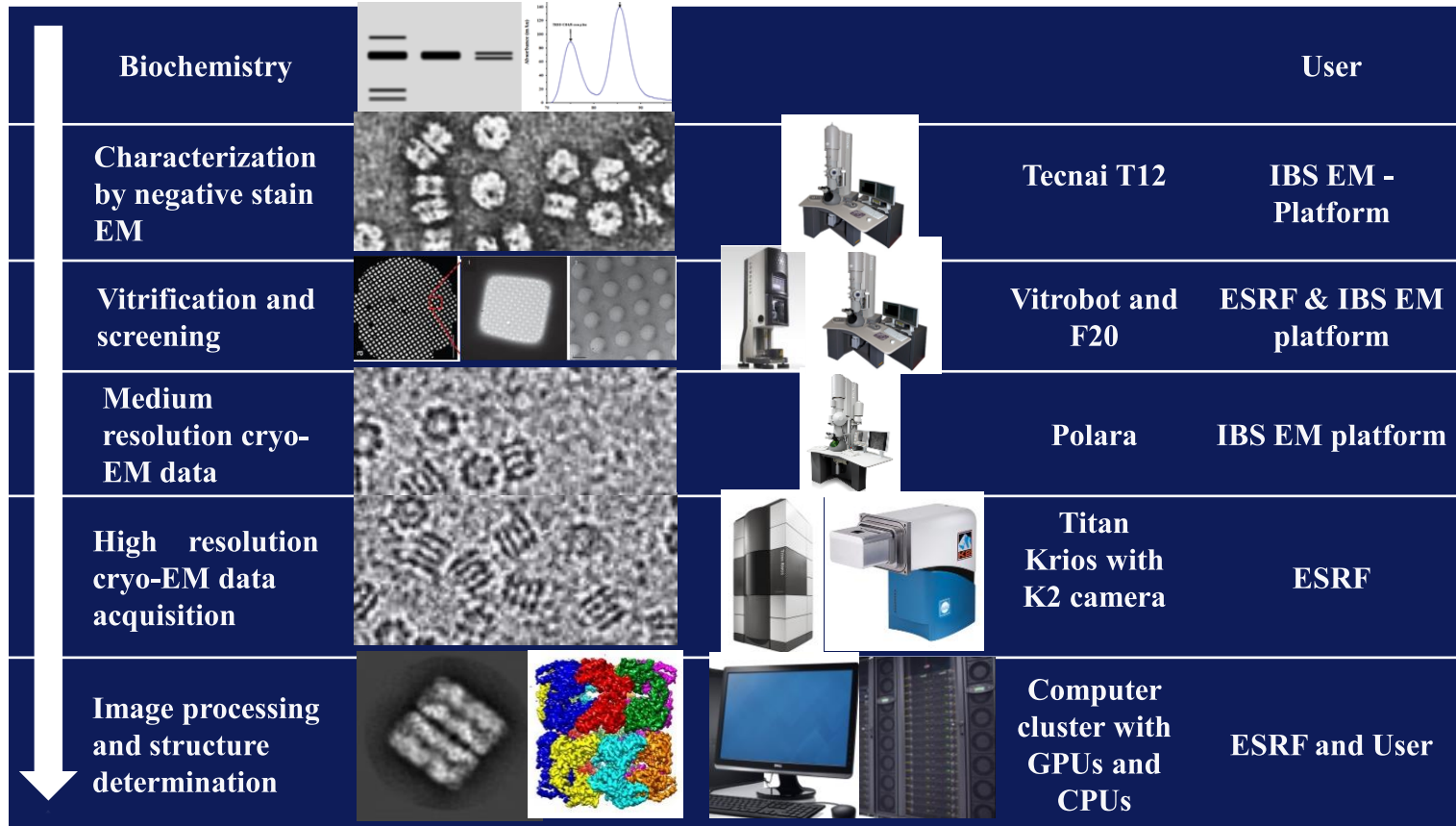
Negative staining



**Guy Schoehn**  
guy.schoehn@ibs.fr  
ibs-plateforme-em.contact@ibs.fr

and  
Cellular electron microscopy  
Correlative microscopy







ESRF – EBS shutdown

10.12.2018 – 08.2020

Cryo-EM facility will be running.