

**Experiment title:****Crystal structures of the human UDP-GlcNAc (UDP-GalNAc) pyrophosphorylases AGX1 and AGX2****Experiment****number:**

LS1657

**Beamline:**

BM14

**Date of experiment:**

from: 26-4-00 to: 27-4-00

**Date of report:**

Aug00

**Shifts:**

6

**Local contact(s):**

Andy THOMPSON

*Received at ESRF:***Names and affiliations of applicants (\* indicates experimentalists):**

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**Report:**

Full-length AGX1 and AGX2 were successfully expressed in *E.coli*, purified and crystallised in the monoclinic space group P2<sub>1</sub> in presence or absence of different substrates/products. The structure of AGX1 was solved at beamline BM14 (ESRF, Grenoble) using Shake'n' Bake from 2.9Å resolution Se-edge MAD data. This structure is presently under refinement using 1.9Å resolution data collected on beamline ID14-2. The structure of AGX2 was solved by molecular replacement using the AGX1 model and data collected on beamline ID14-3. Diffraction data from crystals of the different AGX1- and AGX2-product/substrate complexes were also collected and processed.

Table 1 Structural statistics

| Data collection<br>Protein            | Se-Methionine |           |        | AGX1      |               |                   | Native                  |                         | AGX2              |                         |
|---------------------------------------|---------------|-----------|--------|-----------|---------------|-------------------|-------------------------|-------------------------|-------------------|-------------------------|
|                                       | $f^*$ max     | $f^*$ min | Remote | AGX1      | AGX1-<br>-UTP | AGX1-<br>GlcNAc1P | AGX1-<br>UDP-<br>GlcNAc | AGX1-<br>UDP-<br>GalNAc | AGX2-<br>GlcNAc1P | AGX2-<br>UDP-<br>GlcNAc |
|                                       | BM14          | BM14      | BM14   | ID14-2    | ID14<br>-2    | ID14-2            | ID14-2                  | ID14-2                  | ID14-3            | ID14-3                  |
| Resolution (Å)                        | 2.9           | 2.9       | 2.9    | 1.9       | 2.5           | 2.1               | 1.9                     | 2.45                    | 2.5               | 2.4                     |
| $R_{\text{sym}}$ (%)                  | 10.4          | 9.6       | 12.6   | 6.2       | 6.4           | 6.9               | 6.5                     | 6.6                     | 7.1               | 6.2                     |
| $R_{\text{meas}}$ (%)                 | 9             | 8         | 9.3    | -         | -             | -                 | -                       | -                       | -                 | -                       |
| $1/\sigma$ (I)                        | 6.6           | 7.6       | 5.7    | 6.8       | 7.2           | 8.7               | 8.8                     | 9.1                     | 7.7               | 6.2                     |
| Completeness (%)                      | 98.1          | 96.2      | 93.9   | 94.8      | 90.5          | 96.9              | 95.9                    | 95.9                    | 93.4              | 99.8                    |
| Anomalous completeness                | 95.9          | 91.8      | 88.9   | -         | -             | -                 | -                       | -                       | -                 | -                       |
| Refinement statistics (underway)      |               |           |        |           |               |                   |                         |                         |                   |                         |
| Resolution range (Å)                  |               |           |        | 25-1.9    |               |                   |                         |                         | 25-2.4            |                         |
| $R_{\text{work}}/R_{\text{free}}$ (%) |               |           |        | 30.3/34.9 |               |                   |                         |                         | 30.3/35.1         |                         |

## References

- 1 - A. Wang-Gillam, I. Pastuszak & A. D. Elbein. (1998), *J. Biol Chem* **273** (42), 27055-57.
- 2 - K. Brown, F. Pompeo, S. Dixon, D. Mengin-Lecreulx, C. Cambillau & Y. Bourne (1999), *EMBO J.* **18** (15), 4096-4107.