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**SUIT reference protocol: RP2-Pfi**

**O2k high-resolution respirometry**

SUIT-RP2: FAO-CI substrate control 2016-01-25

D + mt: 1D 2Oct 3Mtit 3c (3NADH) 4P 5G 6S 7U 8Gp 9Rot 10Ama 11Tm 12Azd

|  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| *E* |  |  |  |  |  | **7U** | **8Gp** | **9Rot** | **10Ama** | **11Tm** | **12Azd** |
| *P* | **1D** | **2Oct** | **3M+c** | **4P** | **5G** | **6S** |  |  |  |  |  |
| *L* |  |  |  |  |  |  |  |  |  |  |  |
|  | ROX | FAO | FAO | CI  &FAO | CI  &FAO | CI&II  &FAO | CI&II  &FAO&Gp | CII  &Gp | ROX | CIV | ROX |

**Sample mt=Permeabilized fibres, RP2-Pfi:**

| **O2k and DatLab file: P**\_\_\_( A / B ) 2016-  **Experimental code:**  **Operator:**  **MiR:**  MiR05+CtlCr | | | | | | | | |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Event** | **Mark name** | ***LPE*** | **Final conc. 2 ml O2k** | **Stock**  **[mM]** | **Comment** | **Tit.**  **[µl]** | **A** | **B** |
| **MiR** |  |  |  |  |  |  |  |  |
| **O2** |  |  | ~450 µM |  |  |  |  |  |
| **D** |  |  | 7.5 mM | 500 |  | 30 |  |  |
| **mt** |  |  |  |  |  |  |  |  |
| **O2** | **1D** | ROX | ~450 µM |  |  |  |  |  |
| **Oct** | **2Oct** | *P* | 0.5 mM | 100 |  | 10 |  |  |
| **M.05** | **3M.05** | *P* | 0.05 mM | 50 |  | 2 |  |  |
| **M.1** | **3M.1** | *P* | 0.1 mM | 50 |  | 2 |  |  |
| **M2** | **3M2** | *P* | 2 mM | 400 |  | 9.5 |  |  |
| **c** | **3c** | *P* | 10 µM | 4 |  | 5 |  |  |
| **NADH** | **3NADH** | *P* | 2.8 mM | 280 | NADH only if *FCFc* >.1 | 20 |  |  |
| **P** | **4P** | *P* | 5 mM | 2000 |  | 5 |  |  |
| **G** | **5G** | *P* | 10 mM | 2000 |  | 10 |  |  |
| **S** | **6S** | *P* | 50 mM | 1000 |  | 100 |  |  |
| **U** | **7U** | *E* | Δ0.5 µM | 1 | CCCP | Δ1 µl |  |  |
| **Gp** | **8Gp** | *E* | 10 mM | 1000 |  | 20 |  |  |
| **Rot** | **9Rot** | *E* | 0.5 µM | 1 |  | 1 |  |  |
| **Ama** | **10Ama** | ROX | 2.5 µM | 5 |  | 1 |  |  |
| **O2** |  |  | ~450 µM |  |  |  |  |  |
| **As** |  |  | 2 mM | 800 |  | 5 |  |  |
| **Tm** | **11Tm** | *E* | 0.5 mM | 200 | ~20 min | 5 |  |  |
| **Azd** | **12Azd** | ROX | ≥100 mM | 4000 | ~10 min | 100 |  |  |
| **O2** | **13Azd** | ROX | ~450 µM |  | 400 -> 250 µM |  |  |  |