Description of the dataset

Western-blot results from Synthetic-torpor experiments conducted in 2019-2020

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This dataset contains the original results for the western-blot (WB) determinations from experiments carried out in the "Physiological regulations in the wake-sleep cycle" lab, at the Department of Biomedical and Neuromotor Sciences - University of Bologna, Italy. The WB procedure, the bands acquisitions and their intensity quantifications were conducted at the “Centre for Applied Biomedical Research – CRBA” - University of Bologna, St. Orsola Hospital, Italy.

Part of these data are published on [1], where it is possible to find the complete descriptions and definitions of the experimental conditions and methods used. A brief description of the acronyms used for these conditions could be also found in the file “Experimental conditions”.

Terms of Use

Description

The WB analysis were conducted on Parietal Cortex and Hippocampus samples, taken from Sprague-Dawley albino rats.

The dataset shows the same data represented in two different ways:

- Showing the complete original gels, obtained after running the WB technique.

- Showing the protein bands correspondent for each single specific result and grouped for each experimental condition.

Brief description of the WB method used

The protein concentration was determined using the Bio-Rad DC protein assay kit (Bio-Rad Laboratories). Aliquots were thawed on ice and then denatured in a sample buffer (containing: 500mM DTT, lithium dodecyl sulfate [LDS]) with Coomassie G250 and phenol red (Invitrogen™ NuPAGE) at 65 °C for 10 min. Then, protein samples (20 µg) were loaded and separated electrophoretically using a 1.0 mm thick 4 to 12 % Bis-Tris 10-wells gel together with NuPAGE MOPS SDS Running Buffer (both by Invitrogen™ NuPAGE). The gels were then electrotransferred onto nitrocellulose membranes (Hybond C Extra, Amersham Pharmacia) via wet transfer. Membranes were blocked using 5 % (w/v) not-fat dry milk in 0,1% (v/v) tween-20 in PBS (PBST) for at least 40 min at room temperature, and then incubated overnight at 4 °C with the different primary antibodies used. Bound antibodies were detected using horseradish peroxidase-conjugated Anti-

rabbit and Anti-mouse secondary antibodies. The uniformity of sample loading was confirmed via Ponceau S staining and immunodetection of β-actin, used as a loading control. ChemiDoc™XRS+ (Image Lab™Software, Bio-Rad) was used to acquire digital images through a chemiluminescence reaction (ECL reagents, Amersham). A semi-quantitative measurement of the band intensity was performed using the same computer software and expressed as a ratio of band intensity with respect to the loading control, normalizing the different gels according to a randomly chosen sample used as an internal control (i.e., a sample taken from a single rat that was run on every gel for the different determinations).

Ethical approval

All the experiments were conducted following approval by the National Health Authority (decree: No.262/2020-PR - AEDB0.17.EXT.10), in accordance with the DL 26/2014 and the European Union Directive 2010/63/EU, and under the supervision of the Central Veterinary Service of the University of Bologna. All efforts were made to minimize the number of animals used and their pain and distress.

References

[1] (https://www.biorxiv.org/content/10.1101/2022.03.25.485745v1) Squarcio F, Hitrec T, Piscitiello E, Cerri M, Giovannini C, Martelli D, Occhinegro A, Taddei L, Tupone D, Amici R and Marco L. Synthetic torpor triggers a neuroprotective and regulated mechanism in the rat brain, leading to the reversibility of Tau protein hyperphosphorylation. BioRxive 2022, doi: 10.1101/2022.03.25.485745.