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MEETING ABSTRACT

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Corticosterone induces DNA methyltransferases expression in rat cortical neurons

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Background: Corticosterone is the main glucocorticoid hormone involved in stress responses in rodents. It is established that corticosterone exerts its effects via glucocorticoid receptor (NR3C1) and mineralocorticoid receptor that regulate downstream gene expression during development and adulthood [1]. In our previous study, we have shown that maternal separation on postnatal day 15 (PND15) increases DNA methyltransferase (DNMT) 1, 3A and 3B expression levels in rat nucleus accumbens lasting into adulthood [2]. However, the exact mechanism how maternal separation alters DNMT expression is unclear.

We hypothesize that stress-induced NR3C1 stimulation may increase the expression levels of DNMT and alter long-term DNA methylation/demethylation balance in infant rat brain.

Objectives: Our aim is to evaluate the effect of corticosterone and maternal separation on the expression levels of DNMT in rat cortex and cortical neurons.

Methods: Wistar rats were separated from mothers and littermates on PND 2–14 for 15 minutes (MS15) or 180 minutes (MS180) per day. Animal-facility-reared (AFR) group animals were not separated. Rats were decapitated on PND15, and *Dnmt1*, *Dnmt3a* and *Dnmt3b* mRNA levels in rat prefrontal cortex were measured with RT-qPCR. Plasma corticosterone levels were measured with ELISA. Increased relative DNMT3A protein levels were detected by western blotting in MS180 rats.

Results: Increased plasma corticosterone levels were detected in maternally separated rats. Higher mRNA and protein levels of DNMT3A, and higher mRNA levels of DNMT1 and DNMT3B in rat cortex at postnatal day 15 suggest that elevated corticosterone upregulates DNMTs expression. In rat primary cortical neurons, corticosterone treatment increased mRNA levels of DNMT3A and DNMT3B, which was attenuated by the glucocorticoid receptor antagonist mifepristone. NR3C1 was enriched in *Dnmt3a* promoter after 1 h corticosterone treatment.

Conclusions: The following results suggest that elevated corticosterone upregulates *Dnmt3a* and *Dnmt3b* expression: (1) mRNA levels of *Dnmt3a* and *Dnmt3b*, and protein levels of *Dnmt3a* were increased in the prefrontal cortex of MS180 rats on postnatal day 15; (2) glucocorticoid receptor mediates *Dnmt3a* and *Dnmt3b* mRNA upregulation after corticosterone treatment in primary cortical neurons; (3) NR3C1 binding was detected at *Dnmt3a* promoter in primary cortical cells after corticosterone treatment.

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