MODELLING ANHEDONIA TO STUDY MECHANISMS OF RESILIENCE/VULNERABILITY TOWARDS ACUTE STRESS

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The response to stressful events is physiologically required to adapt to external challenges. However, the stress response can either have a pro-adaptive outcome, when the response is efficiently activated and then inactivated properly, or exert maladaptive effects, when the subject is vulnerable and the response is dysregulated (McEwen et al., 2005). In this context, stress is a risk factor for neuropsychiatric disorders, that represent one of the great therapeutic challenges for the 21st century. Unfortunately, the mechanisms addressing the individual towards stress resilience/vulnerability are still largely unknown.

Recently, we demonstrated that acute inescapable footshock (FS)-stress induces in prefrontal and frontal cortex (PFC/FC) a rapid and sustained increase (up to 24h) of depolarization-evoked glutamate release (Musazzi et al., 2016), together with rapid and sustained structural remodeling of excitatory neurons (Nava et al., 2017). However, it is still unclear whether these changes are required for physiological adaptation or may represent maladaptive alterations.

To dissect adaptive and maladaptive alterations underlying acute stress response, we aimed at comparing functional and molecular changes induced by FS-stress in PFC/FC of rats classified as resilient/vulnerable towards acute stress using sucrose intake (SI) test, a standard behavioral test for anhedonia (Christensen et al., 2011).

36 male rats were housed two per cage. After habituation (sucrose 1% for 2 h), baseline sucrose intake was established giving rats 1 bottle of water and 1 of sucrose 1% for 1 hour, once a week, for 5 weeks (Christensen et al., 2011). Then, 24 animals were assigned to FS-stress (Musazzi et al., 2016) while the others were left undisturbed in their home cages (controls). SI was measured 24h after start of FS-stress. We then evaluated changes in SI vs baseline in response towards FS-stress, and anhedonic animals, (i.e. showing at least a 25% within-subject decrease in SI), were classified as vulnerable (FS-V), while the others were defined as resilient (FS-R) (Christensen et al., 2011). According to the mentioned criteria, we could identify 11 FS-V and 13 FS-R. One-way ANOVA followed by Bonferroni post-hoc analysis showed significant differences of percent alteration of SI in FS-V vs. control ($p<0.0001$), and FS-R ($p<0.0001$), without any differences between controls and FS-R.

All the animals were sacrificed immediately after the test, and basal and depolarization-evoked glutamate release were measured in the PFC/FC, using the method of synaptic terminals in superfusion (Musazzi et al., 2016). We demonstrated that FS-stress increased basal glutamate release only in FS-V, while depolarization-evoked glutamate release was increased in both FS-V and FS-R, compared to controls. Post-hoc analysis didn't reveal any significant difference between FS-V and FS-R, although the increase of depolarization-evoked glutamate release was higher in FS-R.

Moreover, in nuclear fractions and synaptic membranes from the same brain areas, we measured changes in the expression and phosphorylation levels of molecular effectors regulating glutamate release/transmission, and involved in the stress response (i.e. glucocorticoid receptor, synapsin I, HDACs). We are now evaluating whether FS-stress induces specific changes in dendritic morphology of glutamatergic neurons in PFC/FC of vulnerable/resilient rats, taking advantage of Golgi-Cox staining.

Our results showed that SI test can identify behavioral vulnerability/resilience towards FS-stress. Moreover, we found selective functional and molecular alterations of glutamatergic synapses in FS-V vs. FS-R, that could be involved respectively in the maladaptive or adaptive stress response. Although more studies are required, our approach could be useful to characterize the mechanisms of acute stress response, and to find markers of stress vulnerability and putative targets for treatment.

References

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