Published in final edited form as:

Schizophr Res. 2015 December ; 169(1-3): 498–499. doi:10.1016/j.schres.2015.09.030.

Histone H3 phosphorylation is upregulated in PBMCs of schizophrenia patients in comparison to healthy controls*

Rajiv P. Sharma,

The Psychiatric Institute, University of Illinois at Chicago, 1601 W. Taylor St., Chicago, IL 60612, United States. Jesse Brown Veterans Affairs Medical Center, 820 South Damen Avenue (M/C 151), Chicago, IL 60612, United States

Benjamin Feiner, and

The Psychiatric Institute, University of Illinois at Chicago, 1601 W. Taylor St., Chicago, IL 60612, United States

Kayla A. Chase

The Psychiatric Institute, University of Illinois at Chicago, 1601 W. Taylor St., Chicago, IL 60612, United States

Keywords

H3S10phos; PBMC; Lymphocytes; Schizophrenia; Kinases; Phosphorylation; Epigenetics; Histone; Chromatin

Dear Editors

Phosphorylation of H3S10 is an epigenetic modification associated with open chromatin and transcriptional activity and is catalyzed by kinase cascades, emanating from a broad array of membrane receptors including cytokine or monoamine neurotransmitter receptors (Sawicka and Seiser, 2014). Stimuli impinging on interferon receptors (JAK-STAT pathways), G-protein coupled receptors (MAPK family) as well as Toll-like receptors (TLR along the NF- κ B system) in either lymphocytes or neurons can signal towards H3S10 phosphorylation (Crosio et al., 2003; Chwang et al., 2007; Tiwari et al., 2011). H3S10 is a direct target of all the major MAPK pathways (JNK, ERK, p38) in addition to parallel kinase pathways such as

☆Ethical conduct of research: The authors state that they have obtained appropriate institutional review board approval from the University of Illinois at Chicago IRB. In addition, informed consent has been obtained from the participants involved.

Corresponding author at: Psychiatric Institute, 1601 West Taylor Street, Chicago, IL 60612, United States. rsharma@psych.uic.edu.

Conflict of interest statement

There are no conflicts of interest.

Contributors

Rajiv P. Sharma (RPS), Benjamin Feiner (BF) and Kayla A. Chase (KAC).

Conception of study — RPS, BF and KAC.

Data collection — RPS, BF and KAC.

Interpretation of results, data analysis and manuscript drafting — RPS and BF.

Review of manuscript — RPS, BF and KAC.

Financial disclosure statement

There are no financial interests to disclose.

JAK-STAT and NF- κ B which also transduce signals from adipogenic and immunoreactive inputs. The browning lab has demonstrated the ability of clozapine to enhance signaling along this Ras-Raf-MEK-ERK-MSK pathway (Browning et al., 2005). Because subjects with schizophrenia can be characterized by both heightened immunoreactivity (Miller et al., 2011) as well as anti-psychotic treatments, we hypothesized that levels of H3S10phos in nucleated peripheral blood cells could be different between normal and schizophrenia subjects.

Research was initiated after UIC IRB approval and written informed consent. Healthy individuals ($n = 42$) with no major Axis I disorder (as assessed by a SCID interview) were recruited from the community, and schizophrenia subjects were recruited ($n = 37$) through the medical center. Of the 37 schizophrenia patients, 30 were on atypical antipsychotic medication, and 6 were taking typical antipsychotic medication. Clinical symptoms were measured using the PANSS.

An ELISA kit was used to measure H3S10phos (Active Motif #53111) as well as Total H3 (#53110) from acid histone extracts of PBMC from human subjects (Sharma et al., 2006; Sharma, 2012; Chase et al., 2015). Samples were measured in duplicate using a 96-well format with multiple controls: 1) a standard curve; 2) plate-control (invariant sample); and 3) randomized and blinded plate position. Normalization was performed in the following sequence: 1) standard curve measured in ng/ul; 2) scaled to a plate-control; and 3) normalized to total H3. H3S10phos protein levels were an average of 3.20 ng/ul (SD = 2.6) when measured against a commercial recombinant protein. Intra-assay coefficient of variation (within-plate) is 7–8%; inter-assay (between-plates) using the same plate-control is 8–10%.

We found no significant effect of storage time or number of freeze–thaw events, age, sex, race, BMI, or history of smoking on levels of H3S10phos. There were no significant differences, in total H3 levels, between normal controls and schizophrenia subjects.

Schizophrenia subjects had significantly higher levels of H3S10phos compared to normal controls (raw ratio 3.13 vs 6.27), by an approximate 50% increase in the average H3S10phos/total H3 ratios (Fig. 1). These results were significant using either parametric ($F = 4.5$, $df = 1, 78$, $p < 0.03$) or non-parametric Mann Whitney U tests ($U = 385.5$, $p < 0.001$). The PANSS total score ($\rho = -0.47$; $p < 0.004$), was significantly negatively correlated with H3S10phos in participants with schizophrenia.

Given the relationship to the total PANSS score, we could not observe an effect of the dose or duration of antipsychotic use on H3S10phos levels. Chlorpromazine equivalents also did not explain the significant differences seen in levels of H3S10phos (Gardner et al., 2010). Our patients were on stable ongoing treatment with antipsychotics, which Akbarian et al. (2005) has noted is not associated with increased H3S10 phosphorylation. While we did not find a relationship between antipsychotic CPZ equivalents, this question would nonetheless require a more controlled comparison of appropriate samples. Besides antipsychotics however, circulating PBMCs also are capturing immunoreactive signals such as elevated cytokine levels, which could provide an additional stimulus towards heightened intracellular

phosphorylation. Our results suggest that global histone modifications such as H3S10phos can be reliably measured with commercially available ELISA kits and provide another window to explore pharmacological or immunoreactive stimuli.

Acknowledgments

Role of funding source

This work was supported in part by PHS grant (NIH) R01MH094358 (R.P.S.).

References

- Akbarian S, Ruehl MG, Bliven E, Luiz LA, Peranelli AC, Baker SP, Roberts RC, Bunney WE Jr, Conley RC, Jones EG, Tamminga CA, Guo Y. Chromatin alterations associated with down-regulated metabolic gene expression in the prefrontal cortex of subjects with schizophrenia. *Arch Gen Psychiatry*. 2005; 62(8):829–840. [PubMed: 16061760]
- Browning JL, Patel T, Brandt PC, Young KA, Holcomb LA, Hicks PB. Clozapine and the mitogen-activated protein kinase signal transduction pathway: implications for antipsychotic actions. *Biol Psychiatry*. 2005; 57(6):617–623. [PubMed: 15780848]
- Chase KA, Rosen C, Rubin LH, Feiner B, Bodapati AS, Gin H, Hu E, Sharma RP. Evidence of a sex-dependent restrictive epigenome in schizophrenia. *J Psychiatr Res*. 2015; 65:87–94. [PubMed: 25935252]
- Chwang WB, Arthur JS, Schumacher A, Sweatt JD. The nuclear kinase mitogen- and stress-activated protein kinase 1 regulates hippocampal chromatin remodeling in memory formation. *J Neurosci*. 2007; 27(46):12732–12742. [PubMed: 18003853]
- Crosio C, Heitz E, Allis CD, Borrelli E, Sassone-Corsi P. Chromatin remodeling and neuronal response: multiple signaling pathways induce specific histone H3 modifications and early gene expression in hippocampal neurons. *J Cell Sci*. 2003; 116(Pt 24):4905–4914. [PubMed: 14625384]
- Gardner DM, Murphy AL, O'Donnell H, Centorrino F, Baldessarini RJ. International consensus study of antipsychotic dosing. *Am J Psychiatry*. 2010; 167(6):686–693. [PubMed: 20360319]
- Miller BJ, Buckley P, Seabolt W, Mellor A, Kirkpatrick B. Meta-analysis of cytokine alterations in schizophrenia: clinical status and antipsychotic effects. *Biol Psychiatry*. 2011; 70(7):663–671. [PubMed: 21641581]
- Sawicka A, Seiser C. Sensing core histone phosphorylation — a matter of perfect timing. *Biochim Biophys Acta*. 2014; 1839(8):711–718. [PubMed: 24747175]
- Sharma RP. Blood chromatin as a biosensor of the epigenetic milieu: a tool for studies in living psychiatric patients. *Epigenomics*. 2012; 4(5):551–559. [PubMed: 23130836]
- Sharma RP, Rosen C, Kartan S, Guidotti A, Costa E, Grayson DR, Chase K. Valproic acid and chromatin remodeling in schizophrenia and bipolar disorder: preliminary results from a clinical population. *Schizophr Res*. 2006; 88(1–3):227–231. [PubMed: 16996718]
- Tiwari VK, Stadler MB, Wirbelauer C, Paro R, Schubeler D, Beisel C. A chromatin-modifying function of JNK during stem cell differentiation. *Nat Genet*. 2011; 44(1):94–100. [PubMed: 22179133]

Fig. 1.

(Left) H3S10 phosphorylation levels (normalized to total H3 protein and log transformed) is significantly increased in subjects with schizophrenia ($n = 37$) compared to normal controls ($n = 42$). (Right) Correlations between phosphorylated H3S10 and clinical symptomology as measured by the Positive and Negative Syndrome Scale (PANSS).