Published in final edited form as:

Schizophr Res. 2015 October ; 168(1-2): 554–562. doi:10.1016/j.schres.2015.07.026.

Molecular evidence for decreased synaptic efficacy in the postmortem olfactory bulb of individuals with schizophrenia

Chijioke Egbujo^a, Duncan Sinclair^a, Karin Borgmann-Winter^{a,b}, Steven E Arnold^a, Bruce Turetsky^a, and Chang-Gyu Hahn^{a,#}

^aDepartment of Psychiatry, University of Pennsylvania, Philadelphia, PA

^bChildren's Hospital of Philadelphia, Philadelphia, PA

Abstract

Multiple lines of evidence suggest altered synaptic plasticity/connectivity as a pathophysiologic mechanism for various symptom domains of schizophrenia. Olfactory dysfunction, an endophenotype of schizophrenia, reflects altered activity of the olfactory circuitry, which conveys signals from olfactory receptor neurons to the olfactory cortex via synaptic connections in the glomeruli of the olfactory bulb. The olfactory system begins with intranasal olfactory receptor neuron axons synapsing with mitral and tufted cells in the glomeruli of the olfactory bulb, which then convey signals directly to the olfactory cortex. We hypothesized that olfactory dysfunction in schizophrenia is associated with dysregulation of synaptic efficacy in the glomeruli of the olfactory bulb. To test this, we employed semi-quantitative immunohistochemistry to examine the olfactory bulbs of 13 postmortem samples from schizophrenia and their matched control pairs for glomerular expression of 5 pre- and postsynaptic proteins that are involved in the integrity and function of synapses. In the glomeruli of schizophrenia cases compared to their matched controls, we found significant decreases in three presynaptic proteins which play crucial roles in vesicular glutamate transport- synapsin IIa (–18.05%, $p=0.019$), synaptophysin (–24.08% $p=0.0016$) and SNAP-25 (–23.9%, $p=0.046$). Two postsynaptic proteins important for spine formation and glutamatergic signaling were also decreased- spinophilin (–17.40%, $p=0.042$) and PSD-95 (–34.06%, $p=0.015$). These findings provide molecular evidence for decreased efficacy of synapses within the olfactory bulb, which may represent a synaptic mechanism underlying olfactory dysfunction in schizophrenia.

Corresponding author: Chang-Gyu Hahn, Neuropsychiatric Signaling Program, Center for Neurobiology and Behavior, University of Pennsylvania, 125 S 31st St, Philadelphia, PA, 19104 USA; phone 610 520 9131, fax 215-573-2041.

Publisher's Disclaimer: This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final citable form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

Contributions

CE, KBW, SEA, BT, and CGH designed the study together. CE, DS and KBW together conducted the experiments, analyzed and interpreted the data. CE, DS, SEA, BT and CGH wrote the manuscript.

Conflict of Interest

The authors declare no conflict of interest.

Keywords

schizophrenia; synapse; olfactory bulb; synaptophysin; SNAP-25; PSD-95

1. Introduction

Multiple lines of evidence implicate synaptic alterations in schizophrenia. A number of genes and pathways critical for synaptic function have been associated with schizophrenia in genetic studies (Fromer et al., 2014; Kirov et al., 2012; Schizophrenia Working Group of the Psychiatric Genomics, 2014). In postmortem studies, altered expression of genes and proteins involved in synaptic structure or function, such as synaptophysin, synapsin II, GAP-43, SNAP-25, Cdc42 and Duo, have been observed in the dorsolateral prefrontal cortex (DLPFC), temporal lobe, hippocampus and amygdala in schizophrenia (Eastwood et al., 1995; Eastwood et al., 2000; Eastwood and Harrison, 1995; Glantz and Lewis, 1997; Hill et al., 2006; Ide and Lewis, 2010; Karson et al., 1999; Sawada et al., 2002; Tan et al., 2014; Tcherepanov and Sokolov, 1997; Thompson et al., 1998; Varea et al., 2012; Webster et al., 2001). Furthermore, brain imaging studies indicate altered structural and functional connectivity in cortico-limbic and meso-limbic circuitry in schizophrenia (Allen et al., 2012; Alonso-Solis et al., 2014; Amad et al., 2014; Genzel et al., 2014; Whitfield-Gabrieli et al., 2009; Woodward et al., 2012), some of which may be modifiable by antipsychotic medications (Sarpal et al., 2014). As ultra-structural correlates of synaptic connectivity, decreased dendritic spine density observed in the layer 3 of DLPFC (Glantz and Lewis, 2000; Rosoklija et al., 2000) and auditory cortex (Sweet et al., 2009) further supports impaired synaptic connectivity in schizophrenia.

The neural circuitry underlying olfaction may serve as a useful system for studying synaptic efficacy and connectivity in schizophrenia. The essential circuitry consists of the olfactory epithelium (OE) – olfactory bulb (OB) – pyriform cortex. In the OE, odorants bind to odorant receptors generating action potentials in glutamatergic olfactory receptor neurons (ORNs), which synapse with mitral and tufted cells in the glomeruli of the OB (Gottfried, 2010). Within the glomeruli, OB neurons are modulated by relay cells, and axons of OB neurons then travel to the forebrain via the olfactory nerve. Some of these neurons project to the olfactory tubercle, while most of the fibers terminate at the pyriform cortex, where they form synapses with pyramidal neurons that in turn terminate in other parts of the forebrain and limbic system. Neural transmission between the OE and olfactory cortex therefore relies on the relatively simple OE – OB synaptic connection, which is highly concentrated in the olfactory glomeruli. As such, the connectivity/strength in glomerular synapses can affect sensitivity and cognition of olfactory signals (Gottfried, 2010), which can offer clues to synaptic dysregulations in patients with schizophrenia.

Increasing evidence suggests that olfactory dysfunction is an endophenotype of schizophrenia (Turetsky et al., 2008; Turetsky et al., 2003a). Individuals with schizophrenia and those at high risk exhibit deficits in multiple domains of olfactory function including deficits in odor identification (Moberg et al., 1997), odor detection threshold sensitivity (Rupp et al., 2005), odor discrimination (Rupp et al., 2005) and odor memory and odor

hedonic judgments (Moberg et al., 2014; Moberg and Turetsky, 2003; Nguyen et al., 2011; Turetsky et al., 2009; Turetsky et al., 2003b). Interestingly, a lesser degree of olfactory dysfunction also may be found among patients' family members (Moberg et al., 2014) but see (Compton and Chien, 2008; Kamath et al., 2013), suggesting a heritability of this phenotype. Together, olfactory dysfunction may be a heritable trait that co-segregates with the illness and thus meets the criteria for an endophenotype.

Structural and functional alterations in the olfactory circuitry have been reported in schizophrenia (Arnold et al., 2001; Kamath et al., 2011; Nguyen et al., 2011; Rioux et al., 2004). Olfactory event related potentials that are measured in response to odorant stimulation were found to be decreased in patients with schizophrenia compared to controls (Turetsky, 2003a) which indicates an overall decrease in odorant induced neural transmission. Such changes could be in part due to structural changes in key regions of the circuit, as evidenced by decreased size of the OB (Turetsky et al., 2003a; Turetsky et al., 2000) and olfactory sulci (Nguyen et al., 2011; Takahashi et al., 2013a; Takahashi et al., 2013b; Takahashi et al., 2014) in schizophrenia patients and individuals at high risk. In addition, histologic examination of the postmortem OE showed increased ratios of immature vs. mature (GAP-43 immunoreactive vs. OMP immunoreactive) neurons (Arnold et al., 2001), suggesting that altered neuronal maturation may impact olfactory neuron function in schizophrenia patients.

The olfactory glomerulus is a specialized structure in the OB where the axons of the olfactory receptor neurons and dendrites of the olfactory nerves synapse. As such, it permits an opportunity to examine synaptic efficacy with relative ease compared to other regions of the brain. We hypothesized that there would be a decrease in key synaptic proteins in the OB of subjects with schizophrenia, which could mirror similar alterations in other areas of the brain. The results of our study indicate that key presynaptic and postsynaptic proteins are strikingly decreased in the OB of schizophrenia cases compared to their matched controls.

2. Method

2.1 Postmortem samples

Olfactory bulbs were removed at autopsy from 13 prospectively assessed elderly subjects with schizophrenia and 13 non-psychiatric control subjects carefully matched for age, postmortem interval (PMI), gender and fixative. No significant differences in age at death or PMI were found between schizophrenia and control groups. Subjects with schizophrenia had been diagnosed prospectively according to DSM-III-R/DSM-IV criteria based on medical history, interview with caregivers and clinical examination of the patient. Subjects with schizophrenia had been elderly participants in a prospective clinicopathological studies program with post mortem tissues stored with the University of Pennsylvania brain bank. Written informed consent for antemortem evaluation and autopsy in the event of death were obtained from next of kin according to approved institutional review board protocols. The antipsychotic exposures of individuals with schizophrenia were converted to chlorpromazine equivalent doses using established methods (Davis, 1974; Woods, 2003). Brain tissues from non-neuropsychiatric elderly controls were obtained through the University of Pennsylvania's Center for Neurodegenerative Disease Research. While none of these control

subjects had undergone antemortem assessments, a review of their clinical histories found no evidence of any neurological or psychiatric illness.

2.2 Immunohistochemistry

Olfactory bulbs were fixed in formalin (3.7% formaldehyde in 100 mM Tris) or 70% ethanol for 24 h, paraffin embedded, and cut into 10 µm thick sections, and then mounted on poly-L-lysine coated slides. Immunohistochemistry experiments were conducted according to previously described procedures (Talbot et al., 2012), to assess the expression of the synaptic proteins. Briefly, sections were deparaffinized, rehydrated and then incubated for 20 min in 3% H₂O₂ in methanol at room temperature to remove endogenous peroxidase activity. Antigen retrieval was performed, involving boiling for 10 min in 1 mM EDTA, 0.1 M Tris, pH 8.0. After two washes in TTB (0.1M Tris Buffer with 0.01% Triton X-100 pH 7.6) sections were incubated in TTB containing 10% normal horse serum (NS) for 60 min at room temperature. The sections were then incubated overnight at 4°C in primary antibodies diluted in TTB containing 10% NS. Primary antibodies used were a mouse monoclonal anti synapsin-IIa antibody (1:2000, cat #610667, BD Biosciences, San Jose, CA, USA), a rabbit polyclonal anti-synaptophysin antibody (1:2000, cat # 180130, Invitrogen, Waltham, MA, USA), a mouse monoclonal anti-SNAP-25 antibody (1:400, H-1 [sc-376713], Santa Cruz, Santa Cruz, CA, USA), a rabbit polyclonal anti-spinophilin antibody (1:1200, ABA5669, Millipore, Billerica, MA, USA), a mouse monoclonal anti-PSD-95 antibody (1:200, clone K28/43, UC Davis/NIH NeuroMab Facility, Davis, CA, USA), a mouse monoclonal anti-GFAP antibody (1:100,000, MAB360 Millipore, Billerica, MA, USA) and a mouse monoclonal anti-β-actin antibody (1:1000, cat # A2228 Sigma-Aldrich, St. Louis, MO, USA). Slides were then rinsed three times with TTB and incubated in secondary antibody (1:500, Vector Labs, Burlingame, CA, USA) in 10% NS for 1h. After being rinsed two times with TTB, they were incubated for 1 h in Vectastain ABC reagent (1:500, Vector Labs) in TBS at room temperature. The slides were rinsed, and then incubated with the chromogen diaminobenzidine (DAB in 0.1 M Tris) and 0.03% H₂O₂ for 10 min. Sections were then rinsed, dehydrated and coverslipped with Cytoseal (Thermo-Fisher Scientific, Waltham, MA, USA). For PSD-95 quantification, only formaldehyde fixed samples (pairs 1-11) could be used. One section per subject was used in each run. For each protein including actin and GFAP, all cases were processed in a single, precisely timed run. Sections incubated without primary antibody or with an IgG of the same species than the primary antibody were run in parallel and served as negative controls. The concentration of the primary antibodies was based on manufacturers' recommendations and our titration studies.

2.3 Quantification and analysis

To determine the optical density (OD) of the DAB/AB reaction product in the glomeruli of the olfactory bulbs, we used customized Image pro 7.2 software (Media Cybernetics, Rockville, MD, USA) run on a desktop computer attached to a Leitz DMRB microscope (Leica, Wetzlar, Germany) and an Evolution OE camera (Media Cybernetics, Rockville, MD, USA). All the images were captured at the same scope, with the same illumination and settings for each antibody. For each subject a randomly selected slide section of the olfactory bulb was used, the entire section of the bulb was captured and all glomeruli contained in it were delineated. For fifteen subjects (6 cases, 9 controls), both left and right olfactory bulbs

were sectioned and analyzed, while for eleven subjects (7 cases, 4 controls) a single bulb was sectioned and analyzed. Glomeruli were readily identified as areas of increased density of pre- and post-synaptic protein immunostaining within the glomerular layer of the bulb. The gray value for each glomerulus within each section was recorded and converted to an OD value, from which was subtracted the background OD (obtained from non-stained portion of the bulb), yielding the mean OD of each protein across all glomeruli in the section (Balu et al., 2012; Rioux et al., 2005; Talbot et al., 2004). All background-corrected mean glomerular OD data were approximately normally distributed (skewness between -1 and 1). Diagnostic groups were compared using paired two-tailed T-tests, and the relationship between continuous variables and glomerular OD was determined by Pearson's correlations.

3. Results

3.1 Presynaptic protein expression in OB glomeruli

In schizophrenia cases and controls, we investigated the expression levels of three important markers of presynaptic function. Synapsin-IIa is a synaptic vesicle phosphoprotein that is abundantly expressed within the glomeruli, but found much less elsewhere in the olfactory bulb (Figure 1A). There was a significant decrease in the mean expression levels of synapsin-IIa within glomeruli in schizophrenia cases compared to matched control subjects. (-18.1% , $p=0.019$; Figure 1B).

Synaptophysin is a presynaptic protein localized to synaptic vesicle membranes (Wiedenmann and Franke, 1985). Synaptophysin was strongly expressed in the glomeruli of the bulbs, but much less expressed in the peri-glomerular area (Figure 1C). There was a significant decrease in the glomerular levels of synaptophysin in schizophrenia cases compared to normal controls (-24.1% $p=0.0016$; Figure 1D).

SNAP-25 (Soluble NSF Attachment Protein) is a SNARE protein which plays a major role in the fusion of synaptic vesicles with the presynaptic membrane for neurotransmitter release (Washbourne et al., 2002). We observed that SNAP-25 is highly expressed in the olfactory bulb glomeruli (Figure 1E). There was a significant decrease in mean glomerular SNAP-25 expression levels in schizophrenia cases compared to matched controls. (-23.9% , $p=0.046$; Figure 1F).

3.2 Postsynaptic protein expression in OB glomeruli

The glomerular expression of two postsynaptic proteins was also quantified. Firstly, spinophilin is a postsynaptic marker which is highly expressed in dendritic spines (Muly et al., 2004). Spinophilin immunostaining was evident in various regions of the bulb but more so in the glomeruli (Figure 2A). There was a significant decrease in spinophilin expression within the glomeruli in schizophrenia cases compared to matched controls. (-17.4% , $p=0.042$; Figure 2B).

PSD-95 is a core scaffolding protein located within the postsynaptic density of excitatory synapses (Kim and Sheng, 2004). PSD-95 was highly expressed in the olfactory bulb glomeruli, but sparsely expressed in the peri-glomerular area (Figure 2C). We observed a

significant decrease in the expression levels of PSD-95 in the glomeruli of schizophrenia subjects when compared to matched controls (-34.1% , $p=0.015$; Figure 2D).

To determine whether non-specific or possibly artifactual differences between groups may account for group differences in synaptic proteins, β -actin and GFAP were quantified as internal controls. Both GFAP and β -actin was not highly concentrated in glomeruli (Figure 3E), so glomerular and peri-glomerular areas were quantified together. There were no differences between schizophrenia and matched control subjects in glomerular and peri-glomerular GFAP and β -actin expression ([GFAP] $p=0.41$, β -actin [$p=0.64$]; Figure 3F for not shown and for).

Overall, the glomerular expression of each synaptic protein investigated in this study was significantly decreased in subjects with schizophrenia when compared to controls (Figure 3). We found no significant correlations between expression levels of the synaptic proteins studied with age or PMI within control and schizophrenia groups individually, or both groups combined. Expression levels also were not correlated with age of onset, duration of illness or antipsychotic dosage within the schizophrenia group.

4. Discussion

Given multiple reports of abnormal synaptic density and protein expression in various cortical and limbic brain regions in schizophrenia (Glantz and Lewis, 1997, 2000; Hill et al., 2006; Karson et al., 1999; Rosoklija et al., 2000; Sweet et al., 2009; Thompson et al., 1998), we hypothesized that altered synaptic connectivity in schizophrenia would be reflected in dysregulation of key synaptic proteins in the olfactory glomeruli of schizophrenia cases. Consistent with this hypothesis, we observed a significant decrease in the expression levels of pre- and post-synaptic proteins synapsin-IIa, synaptophysin, SNAP-25, spinophilin and PSD-95 in the glomeruli of the olfactory bulb in schizophrenia, but no decrease in GFAP or β -actin. These findings may reflect a decrease in synaptic connectivity within the glomeruli, which could contribute to the olfactory dysfunction observed in schizophrenia.

In this study, levels of three presynaptic proteins (synapsin-IIa, synaptophysin and SNAP-25) and two postsynaptic proteins (spinophilin and PSD-95) were decreased in the glomeruli of individuals with schizophrenia. In presynaptic terminals, these three proteins are involved in multiple stages of vesicular neurotransmitter release, including reserve pool regulation [synapsin-IIa; (Gitler et al., 2008)], vesicle fusion [SNAP-25; (Washbourne et al., 2002)] and endocytosis [synaptophysin; (Kwon and Chapman, 2011)]. Postsynaptically, PSD-95 stabilizes the synapse and modulates synaptic long-term potentiation (Nagura et al., 2012) while spinophilin regulates spine morphology and plasticity (Feng et al., 2000; Sarrouilhe et al., 2006).

While deficits in a subset of these proteins may have suggested dysregulation of specific genes or pathways, the concurrent decreases of all five proteins are likely to reflect overall changes in synaptic integrity, such as decreased number of synapses or diminished synaptic strength throughout olfactory glomeruli in schizophrenia. The spatial resolution of our methods did not allow us to distinguish between these two possibilities.

The origin of such overall changes in the olfactory synapses is unclear at present. It is possible that dysregulations either in the presynaptic or postsynaptic segments are primary, and the other segment is affected via trans-synaptic communication. It is also possible that fundamental neurobiological changes affect each segment separately and together disrupt the synaptic ultrastructure. In this regard, our previous observations of altered neuronal lineage in the postmortem OE may suggest a role of presynaptic alterations. In the OE of schizophrenia cases, we found decreased ratios of OMP immunoreactive (IR) cells with respect to GAP-43 IR cells, each representing mature and immature olfactory sensory neurons (OSNs) respectively (Arnold et al., 2001). Given that presynaptic molecules in the OB arise from OSNs, decreased proportions of mature neurons could account for decreased expression of presynaptic molecules in the OB, which in turn may compromise the integrity of postsynaptic segment.

Two previous studies have investigated synaptic protein expression in the OB in schizophrenia. One study reported decreased MAP2 expression in OB glomeruli in schizophrenia (Rioux et al., 2004), suggesting a possible decrease in glomerular dendritic arborization which would be consistent with our findings. However, in a follow-up study, the same group did not identify differences in pre- and post-synaptic glomerular proteins OMP, GAP-43, NCAM, calbindin, NFM/HP, β -tubulin III or TrkB, and although synaptophysin was decreased by about 13% it was not statistically significant (Rioux et al., 2005). Further work is required to determine to what extent pre- and postsynaptic deficits in OB glomeruli are a consistent feature of schizophrenia, and to clarify whether deficits are specific to key cellular processes such as vesicular transport, which was the predominant focus of this study.

Our observations of decreased expression of synaptic proteins are consistent with similar findings in various postmortem brain regions in schizophrenia, many of which demonstrated brain region- or cell layer-specificity. Decreased levels of synapsin I and pan synapsin proteins in the hippocampus (Browning et al., 1993; Vawter et al., 2002), and synapsin III in the DLPFC (Porton and Wetsel, 2007), have been reported in schizophrenia. Decreased SNAP-25 protein has also been described in the PFC (BA10) (Karson et al., 1999; Thompson et al., 2003) and hippocampus (Fatemi et al., 2001; Young et al., 1998), while fewer spinophilin-positive puncta have been described in the auditory cortex in schizophrenia (Sweet et al., 2009). Decreased PSD-95 in the anterior cingulate cortex in schizophrenia (Funk et al., 2009), but increased PSD-95 in the dorsomedial thalamus (Clinton et al., 2006), have been described. Finally, decreased synaptophysin has been reported in the PFC (BA9, 10, 46) (Glantz and Lewis, 1997; Karson et al., 1999; Rao et al., 2013), thalamus (Landen et al., 1999), hippocampus (Davidsson et al., 1999; Vawter et al., 1999) and cingulate cortex (Davidsson et al., 1999). These broad array of findings, which reveal decreases in the same proteins quantified in our study, support that altered synaptic connectivity in multiple brain regions may be a hallmark of schizophrenia. Although many of these findings have been replicated, other studies have failed to find differences in other brain regions implicated in schizophrenia (Barksdale et al., 2014; Gray et al., 2010; Halim et al., 2003; Vawter et al., 2002; Young et al., 1998). One reason for the divergence of these findings may be that, while widespread, synaptic deficits may still be localized to discrete brain regions and cell types within them. The glomerulus of the OB represents a subcellular

domain which has a concentration of synaptic inputs and can be extremely accurately demarcated, suggesting that it may be a useful brain region for further exploration of synaptic abnormalities in schizophrenia.

Given that all the individuals with schizophrenia had been on varying doses of antipsychotics for several years, it is important to consider the impact of antipsychotics on our findings. Consistent with previous findings with spinophilin in schizophrenia (Sweet et al., 2009), we did not observe a correlation of antipsychotic dosage with levels of pre- or post-synaptic molecules. This analysis, however, has inherent limitations because although coefficients used in converting different antipsychotics to their chlorpromazine equivalents have been empirically derived and are based on expert consensus, they do not take into account the distinct effects of individual drugs or the effects of polypharmacy. In addition, the doses used for conversion were the dosages that the patients were on at the time of death, and may not accurately reflect cumulative lifetime exposure. This however remains an area for further study to evaluate the long term effects of antipsychotics on the olfactory circuit given that significant neurogenesis takes place in this circuit as OSNs are constantly regenerated in adult life.

An additional possible confound in this study is smoking, which is more common among individuals with schizophrenia than normal controls (Dickerson et al., 2013). Smoking may impair olfaction [(Katotomichelakis et al., 2007; Schriever et al., 2013)] and could lead to decreased olfactory bulb volume (Schriever et al., 2013). In previous studies, olfactory bulb volume decreases in schizophrenia cases and first degree relatives were significant when covarying for smoking status and volume (Moberg et al., 2014; Turetsky et al., 2003a; Turetsky et al., 2000), suggesting deficits in schizophrenia independent of any effects of smoking.

5. Conclusion

We report decreased levels of pre- and postsynaptic proteins in the OB glomeruli of individuals with schizophrenia which is consistent with the hypothesis that abnormal synaptic efficacy is a feature of the illness. Future studies will need to integrate findings from the OB, OE and olfactory cortex to shed further light on the origins of synaptic and circuit abnormalities that underlie schizophrenia.

Acknowledgements

We thank Konrad Talbot for providing technical expertise and advice in the design and execution of the project, Hala Kazi for technical assistance, and John Robinson, Theresa Schuck and the PENN Brain Bank for assistance with tissues and demographic data.

Role of Funding Source

CE is supported by an ITMAT CTSA KL2 Grant, DS by an NHMRC (Australia) CJ Martin Fellowship (APP1072878), and CGH by NIH grants MH890140 and MH10028395.

References

- Allen P, Luigjes J, Howes OD, Egerton A, Hirao K, Valli I, Kambeitz J, Fusar-Poli P, Broome M, McGuire P. Transition to psychosis associated with prefrontal and subcortical dysfunction in ultra high-risk individuals. *Schizophr Bull.* 2012; 38(6):1268–1276. [PubMed: 22290265]
- Alonso-Solis A, Vives-Gilabert Y, Grasa E, Portella MJ, Rabella M, Sauras RB, Roldan A, Nunez-Marin F, Gomez-Anson B, Perez V, Alvarez E, Corripio I. Resting-state functional connectivity alterations in the default network of schizophrenia patients with persistent auditory verbal hallucinations. *Schizophr Res.* 2014
- Amad A, Cachia A, Gorwood P, Pins D, Delmaire C, Rolland B, Mondino M, Thomas P, Jardri R. The multimodal connectivity of the hippocampal complex in auditory and visual hallucinations. *Mol Psychiatry.* 2014; 19(2):184–191. [PubMed: 23318999]
- Arnold SE, Han LY, Moberg PJ, Turetsky BI, Gur RE, Trojanowski JQ, Hahn CG. Dysregulation of olfactory receptor neuron lineage in schizophrenia. *Arch Gen Psychiatry.* 2001; 58(9):829–835. [PubMed: 11545665]
- Balu DT, Carlson GC, Talbot K, Kazi H, Hill-Smith TE, Easton RM, Birnbaum MJ, Lucki I. Akt1 deficiency in schizophrenia and impairment of hippocampal plasticity and function. *Hippocampus.* 2012; 22(2):230–240. [PubMed: 21049487]
- Barksdale KA, Lahti AC, Roberts RC. Synaptic proteins in the postmortem anterior cingulate cortex in schizophrenia: relationship to treatment and treatment response. *Neuropsychopharmacology.* 2014; 39(9):2095–2103. [PubMed: 24603856]
- Browning MD, Dudek EM, Rapier JL, Leonard S, Freedman R. Significant reductions in synapsin but not synaptophysin specific activity in the brains of some schizophrenics. *Biol Psychiatry.* 1993; 34(8):529–535. [PubMed: 8274580]
- Clinton SM, Haroutunian V, Meador-Woodruff JH. Up-regulation of NMDA receptor subunit and post-synaptic density protein expression in the thalamus of elderly patients with schizophrenia. *J Neurochem.* 2006; 98(4):1114–1125. [PubMed: 16762023]
- Compton MT, Chien VH. No association between psychometrically-determined schizotypy and olfactory identification ability in first-degree relatives of patients with schizophrenia and non-psychiatric controls. *Schizophr Res.* 2008; 100(1-3):216–223. [PubMed: 18077137]
- Davidsson P, Gottfries J, Bogdanovic N, Ekman R, Karlsson I, Gottfries CG, Blennow K. The synaptic-vesicle-specific proteins rab3a and synaptophysin are reduced in thalamus and related cortical brain regions in schizophrenic brains. *Schizophr Res.* 1999; 40(1):23–29. [PubMed: 10541003]
- Davis JM. Dose equivalence of the antipsychotic drugs. *J Psychiatr Res.* 1974; 11:65–69. [PubMed: 4156792]
- Dickerson F, Stallings CR, Origoni AE, Vaughan C, Khushalani S, Schroeder J, Yolken RH. Cigarette smoking among persons with schizophrenia or bipolar disorder in routine clinical settings, 1999-2011. *Psychiatr Serv.* 2013; 64(1):44–50. [PubMed: 23280457]
- Eastwood SL, Burnet PW, Harrison PJ. Altered synaptophysin expression as a marker of synaptic pathology in schizophrenia. *Neuroscience.* 1995; 66(2):309–319. [PubMed: 7477874]
- Eastwood SL, Cairns NJ, Harrison PJ. Synaptophysin gene expression in schizophrenia. Investigation of synaptic pathology in the cerebral cortex. *Br J Psychiatry.* 2000; 176:236–242. [PubMed: 10755070]
- Eastwood SL, Harrison PJ. Decreased synaptophysin in the medial temporal lobe in schizophrenia demonstrated using immunautoradiography. *Neuroscience.* 1995; 69(2):339–343. [PubMed: 8552231]
- Fatemi SH, Earle JA, Stary JM, Lee S, Sedgewick J. Altered levels of the synaptosomal associated protein SNAP-25 in hippocampus of subjects with mood disorders and schizophrenia. *Neuroreport.* 2001; 12(15):3257–3262. [PubMed: 11711867]
- Feng J, Yan Z, Ferreira A, Tomizawa K, Liauw JA, Zhuo M, Allen PB, Ouimet CC, Greengard P. Spinophilin regulates the formation and function of dendritic spines. *Proc Natl Acad Sci U S A.* 2000; 97(16):9287–9292. [PubMed: 10922077]

- Fromer M, Pocklington AJ, Kavanagh DH, Williams HJ, Dwyer S, Gormley P, Georgieva L, Rees E, Palta P, Ruderfer DM, Carrera N, Humphreys I, Johnson JS, Roussos P, Barker DD, Banks E, Milanova V, Grant SG, Hannon E, Rose SA, Chambert K, Mahajan M, Scolnick EM, Moran JL, Kirov G, Palotie A, McCarroll SA, Holmans P, Sklar P, Owen MJ, Purcell SM, O'Donovan MC. De novo mutations in schizophrenia implicate synaptic networks. *Nature*. 2014; 506(7487):179–184. [PubMed: 24463507]
- Funk AJ, Rumbaugh G, Haroutunian V, McCullumsmith RE, Meador-Woodruff JH. Decreased expression of NMDA receptor-associated proteins in frontal cortex of elderly patients with schizophrenia. *Neuroreport*. 2009; 20(11):1019–1022. [PubMed: 19483657]
- Genzel L, Dresler M, Cornu M, Jager E, Konrad B, Adamczyk M, Friess E, Steiger A, Cizisch M, Goya-Maldonado R. Medial Prefrontal-Hippocampal Connectivity and Motor Memory Consolidation in Depression and Schizophrenia. *Biol Psychiatry*. 2014
- Gitler D, Cheng Q, Greengard P, Augustine GJ. Synapsin IIa controls the reserve pool of glutamatergic synaptic vesicles. *J Neurosci*. 2008; 28(43):10835–10843. [PubMed: 18945891]
- Glantz LA, Lewis DA. Reduction of synaptophysin immunoreactivity in the prefrontal cortex of subjects with schizophrenia. Regional and diagnostic specificity. *Archives of general psychiatry*. 1997; 54(10):943–952. [PubMed: 9337775]
- Glantz LA, Lewis DA. Decreased dendritic spine density on prefrontal cortical pyramidal neurons in schizophrenia. *Arch Gen Psychiatry*. 2000; 57(1):65–73. [PubMed: 10632234]
- Gottfried JA. Central mechanisms of odour object perception. *Nat Rev Neurosci*. 2010; 11(9):628–641. [PubMed: 20700142]
- Gray LJ, Dean B, Kronsbein HC, Robinson PJ, Scarr E. Region and diagnosis-specific changes in synaptic proteins in schizophrenia and bipolar I disorder. *Psychiatry Res*. 2010; 178(2):374–380. [PubMed: 20488553]
- Halim ND, Weickert CS, McClintock BW, Hyde TM, Weinberger DR, Kleinman JE, Lipska BK. Presynaptic proteins in the prefrontal cortex of patients with schizophrenia and rats with abnormal prefrontal development. *Molecular psychiatry*. 2003; 8(9):797–810. [PubMed: 12931207]
- Hill JJ, Hashimoto T, Lewis DA. Molecular mechanisms contributing to dendritic spine alterations in the prefrontal cortex of subjects with schizophrenia. *Mol Psychiatry*. 2006; 11(6):557–566. [PubMed: 16402129]
- Ide M, Lewis DA. Altered cortical CDC42 signaling pathways in schizophrenia: implications for dendritic spine deficits. *Biol Psychiatry*. 2010; 68(1):25–32. [PubMed: 20385374]
- Kamath V, Moberg PJ, Kohler CG, Gur RE, Turetsky BI. Odor hedonic capacity and anhedonia in schizophrenia and unaffected first-degree relatives of schizophrenia patients. *Schizophr Bull*. 2013; 39(1):59–67. [PubMed: 21616912]
- Kamath V, Turetsky BI, Moberg PJ. Identification of pleasant, neutral, and unpleasant odors in schizophrenia. *Psychiatry research*. 2011; 187(1-2):30–35. [PubMed: 21239063]
- Karson CN, Mrak RE, Schluterman KO, Sturmer WQ, Sheng JG, Griffin WS. Alterations in synaptic proteins and their encoding mRNAs in prefrontal cortex in schizophrenia: a possible neurochemical basis for 'hypofrontality'. *Mol Psychiatry*. 1999; 4(1):39–45. [PubMed: 10089007]
- Katotomichelakis M, Balatsouras D, Tripsianis G, Davris S, Maroudias N, Danielides V, Simopoulos C. The effect of smoking on the olfactory function. *Rhinology*. 2007; 45(4):273–280. [PubMed: 18085020]
- Kim E, Sheng M. PDZ domain proteins of synapses. *Nat Rev Neurosci*. 2004; 5(10):771–781. [PubMed: 15378037]
- Kirov G, Pocklington AJ, Holmans P, Ivanov D, Ikeda M, Ruderfer D, Moran J, Chambert K, Toncheva D, Georgieva L, Grozeva D, Fjodorova M, Wollerton R, Rees E, Nikolov I, van de Lagaat LN, Bayes A, Fernandez E, Olason PI, Bottcher Y, Komiyama NH, Collins MO, Choudhary J, Stefansson K, Stefansson H, Grant SG, Purcell S, Sklar P, O'Donovan MC, Owen MJ. De novo CNV analysis implicates specific abnormalities of postsynaptic signalling complexes in the pathogenesis of schizophrenia. *Mol Psychiatry*. 2012; 17(2):142–153. [PubMed: 22083728]
- Kwon SE, Chapman ER. Synaptophysin regulates the kinetics of synaptic vesicle endocytosis in central neurons. *Neuron*. 2011; 70(5):847–854. [PubMed: 21658579]

- Landen M, Davidsson P, Gottfries CG, Grenfeldt B, Stridsberg M, Blennow K. Reduction of the small synaptic vesicle protein synaptophysin but not the large dense core chromogranins in the left thalamus of subjects with schizophrenia. *Biol Psychiatry*. 1999; 46(12):1698–1702. [PubMed: 10624552]
- Moberg PJ, Doty RL, Turetsky BI, Arnold SE, Mahr RN, Gur RC, Bilker W, Gur RE. Olfactory identification deficits in schizophrenia: correlation with duration of illness. *American Journal of Psychiatry*. 1997; 154(7):1016–1018. [PubMed: 9210756]
- Moberg PJ, Kamath V, Marchetto DM, Calkins ME, Doty RL, Hahn CG, Borgmann-Winter KE, Kohler CG, Gur RE, Turetsky BI. Meta-analysis of olfactory function in schizophrenia, first-degree family members, and youths at-risk for psychosis. *Schizophr Bull*. 2014; 40(1):50–59. [PubMed: 23641047]
- Moberg PJ, Turetsky BI. Scent of a disorder: olfactory functioning in schizophrenia. *Curr Psychiatry Rep*. 2003; 5(4):311–319. [PubMed: 12857535]
- Muly EC, Smith Y, Allen P, Greengard P. Subcellular distribution of spinophilin immunolabeling in primate prefrontal cortex: localization to and within dendritic spines. *J Comp Neurol*. 2004; 469(2):185–197. [PubMed: 14694533]
- Nagura H, Ishikawa Y, Kobayashi K, Takao K, Tanaka T, Nishikawa K, Tamura H, Shiosaka S, Suzuki H, Miyakawa T, Fujiyoshi Y, Doi T. Impaired synaptic clustering of postsynaptic density proteins and altered signal transmission in hippocampal neurons, and disrupted learning behavior in PDZ1 and PDZ2 ligand binding-deficient PSD-95 knockin mice. *Molecular brain*. 2012; 5:43. [PubMed: 23268962]
- Nguyen AD, Pelavin PE, Shenton ME, Chilakamarri P, McCarley RW, Nestor PG, Levitt JJ. Olfactory sulcal depth and olfactory bulb volume in patients with schizophrenia: an MRI study. *Brain Imaging Behav*. 2011; 5(4):252–261. [PubMed: 21728040]
- Porton B, Wetsel WC. Reduction of synapsin III in the prefrontal cortex of individuals with schizophrenia. *Schizophr Res*. 2007; 94(1-3):366–370. [PubMed: 17540541]
- Rao JS, Kim HW, Harry GJ, Rapoport SI, Reese EA. Increased neuroinflammatory and arachidonic acid cascade markers, and reduced synaptic proteins, in the postmortem frontal cortex from schizophrenia patients. *Schizophr Res*. 2013; 147(1):24–31. [PubMed: 23566496]
- Rioux L, Gelber EI, Parand L, Kazi HA, Yeh J, Wintering R, Bilker W, Arnold SE. Characterization of olfactory bulb glomeruli in schizophrenia. *Schizophr Res*. 2005; 77(2-3):229–239. [PubMed: 15946825]
- Rioux L, Ruschinsky D, Arnold SE. Microtubule-associated protein MAP2 expression in olfactory bulb in schizophrenia. *Psychiatry Res*. 2004; 128(1):1–7. [PubMed: 15450909]
- Rosoklija G, Toomayan G, Ellis SP, Keilp J, Mann JJ, Latov N, Hays AP, Dwork AJ. Structural abnormalities of subicular dendrites in subjects with schizophrenia and mood disorders: preliminary findings. *Arch Gen Psychiatry*. 2000; 57(4):349–356. [PubMed: 10768696]
- Rupp CI, Fleischhacker WW, Kemmler G, Oberbauer H, Scholtz AW, Wanko C, Hinterhuber H. Various bilateral olfactory deficits in male patients with schizophrenia. *Schizophrenia bulletin*. 2005; 31(1):155–165. [PubMed: 15888433]
- Sarpal DK, Robinson DG, Lencz T, Argyelan M, Ikuta T, Karlsgodt K, Gallego JA, Kane JM, Szeszko PR, Malhotra AK. Antipsychotic Treatment and Functional Connectivity of the Striatum in First-Episode Schizophrenia. *JAMA psychiatry*. 2014
- Sarrouilhe D, di Tommaso A, Métayé T, Ladeveze V. Spinophilin: from partners to functions. *Biochimie*. 2006; 88(9):1099–1113. [PubMed: 16737766]
- Sawada K, Young CE, Barr AM, Longworth K, Takahashi S, Arango V, Mann JJ, Dwork AJ, Falkai P, Phillips AG, Honer WG. Altered immunoreactivity of complexin protein in prefrontal cortex in severe mental illness. *Mol Psychiatry*. 2002; 7(5):484–492. [PubMed: 12082566]
- Schizophrenia Working Group of the Psychiatric Genomics, C. Biological insights from 108 schizophrenia-associated genetic loci. *Nature*. 2014; 511(7510):421–427. [PubMed: 25056061]
- Schriever VA, Reither N, Gerber J, Iannilli E, Hummel T. Olfactory bulb volume in smokers. *Exp Brain Res*. 2013; 225(2):153–157. [PubMed: 23212471]

- Sweet RA, Henteleff RA, Zhang W, Sampson AR, Lewis DA. Reduced dendritic spine density in auditory cortex of subjects with schizophrenia. *Neuropsychopharmacology*. 2009; 34(2):374–389. [PubMed: 18463626]
- Takahashi T, Nakamura Y, Nakamura K, Ikeda E, Furuichi A, Kido M, Kawasaki Y, Noguchi K, Seto H, Suzuki M. Altered depth of the olfactory sulcus in first-episode schizophrenia. *Progress in neuro-psychopharmacology & biological psychiatry*. 2013a; 40:167–172. [PubMed: 23063493]
- Takahashi T, Nakamura Y, Nakamura K, Nishiyama S, Ikeda E, Furuichi A, Kido M, Noguchi K, Suzuki M. Altered depth of the olfactory sulcus in subjects at risk of psychosis. *Schizophr Res*. 2013b; 149(1-3):186–187. [PubMed: 23830683]
- Takahashi T, Wood SJ, Yung AR, Nelson B, Lin A, Yucel M, Phillips LJ, Nakamura Y, Suzuki M, Brewer WJ, Proffitt TM, McGorry PD, Velakoulis D, Pantelis C. Altered depth of the olfactory sulcus in ultra high-risk individuals and patients with psychotic disorders. *Schizophr Res*. 2014; 153(1-3):18–24. [PubMed: 24530137]
- Talbot K, Eidem WL, Tinsley CL, Benson MA, Thompson EW, Smith RJ, Hahn CG, Siegel SJ, Trojanowski JQ, Gur RE, Blake DJ, Arnold SE. Dysbindin-1 is reduced in intrinsic, glutamatergic terminals of the hippocampal formation in schizophrenia. *J Clin Invest*. 2004; 113(9):1353–1363. [PubMed: 15124027]
- Talbot K, Wang HY, Kazi H, Han LY, Bakshi KP, Stucky A, Fuino RL, Kawaguchi KR, Samoyedny AJ, Wilson RS, Arvanitakis Z, Schneider JA, Wolf BA, Bennett DA, Trojanowski JQ, Arnold SE. Demonstrated brain insulin resistance in Alzheimer's disease patients is associated with IGF-1 resistance, IRS-1 dysregulation, and cognitive decline. *The Journal of clinical investigation*. 2012; 122(4):1316–1338. [PubMed: 22476197]
- Tan ML, Dyck BA, Gabriele J, Daya RP, Thomas N, Sookram C, Basu D, Ferro MA, Chong VZ, Mishra RK. Synapsin II gene expression in the dorsolateral prefrontal cortex of brain specimens from patients with schizophrenia and bipolar disorder: effect of lifetime intake of antipsychotic drugs. *Pharmacogenomics J*. 2014; 14(1):63–69. [PubMed: 23529008]
- Tcherepanov AA, Sokolov BP. Age-related abnormalities in expression of mRNAs encoding synapsin 1A, synapsin 1B, and synaptophysin in the temporal cortex of schizophrenics. *J Neurosci Res*. 1997; 49(5):639–644. [PubMed: 9302085]
- Thompson PM, Egbufoama S, Vawter MP. SNAP-25 reduction in the hippocampus of patients with schizophrenia. *Progress in neuro-psychopharmacology & biological psychiatry*. 2003; 27(3):411–417. [PubMed: 12691775]
- Thompson PM, Sower AC, Perrone-Bizzozero NI. Altered levels of the synaptosomal associated protein SNAP-25 in schizophrenia. *Biol Psychiatry*. 1998; 43(4):239–243. [PubMed: 9513732]
- Turetsky B, Moberg PJ, Owzar K, Johnson SC, Doty RL, Gur RE. Physiologic impairment of olfactory stimulus processing in schizophrenia. *Biol Psychiatry*. 2003; 53:403–411. [PubMed: 12614993]
- Turetsky BI, Hahn CG, Borgmann-Winter K, Moberg PJ. Scents and nonsense: olfactory dysfunction in schizophrenia. *Schizophr Bull*. 2009; 35(6):1117–1131. [PubMed: 19793796]
- Turetsky BI, Kohler CG, Gur RE, Moberg PJ. Olfactory physiological impairment in first-degree relatives of schizophrenia patients. *Schizophrenia research*. 2008; 102(1-3):220–229. [PubMed: 18457935]
- Turetsky BI, Moberg PJ, Arnold SE, Doty RL, Gur RE. Low olfactory bulb volume in first-degree relatives of patients with schizophrenia. *Am J Psychiatry*. 2003a; 160(4):703–708. [PubMed: 12668359]
- Turetsky BI, Moberg PJ, Roalf DR, Arnold SE, Gur RE. Decrements in volume of anterior ventromedial temporal lobe and olfactory dysfunction in schizophrenia. *Arch Gen Psychiatry*. 2003b; 60(12):1193–1200. [PubMed: 14662551]
- Turetsky BI, Moberg PJ, Yousem DM, Doty RL, Arnold SE, Gur RE. Reduced olfactory bulb volume in patients with schizophrenia. *Am J Psychiatry*. 2000; 157(5):828–830. [PubMed: 10784482]
- Varea E, Guirado R, Gilabert-Juan J, Marti U, Castillo-Gomez E, Blasco-Ibanez JM, Crespo C, Nacher J. Expression of PSA-NCAM and synaptic proteins in the amygdala of psychiatric disorder patients. *J Psychiatr Res*. 2012; 46(2):189–197. [PubMed: 22099865]

- Vawter MP, Howard AL, Hyde TM, Kleinman JE, Freed WJ. Alterations of hippocampal secreted N-CAM in bipolar disorder and synaptophysin in schizophrenia. *Mol Psychiatry*. 1999; 4(5):467–475. [PubMed: 10523820]
- Vawter MP, Thatcher L, Usen N, Hyde TM, Kleinman JE, Freed WJ. Reduction of synapsin in the hippocampus of patients with bipolar disorder and schizophrenia. *Mol Psychiatry*. 2002; 7(6):571–578. [PubMed: 12140780]
- Washbourne P, Thompson PM, Carta M, Costa ET, Mathews JR, Lopez-Bendito G, Molnar Z, Becher MW, Valenzuela CF, Partridge LD, Wilson MC. Genetic ablation of the t-SNARE SNAP-25 distinguishes mechanisms of neuroexocytosis. *Nat Neurosci*. 2002; 5(1):19–26. [PubMed: 11753414]
- Webster MJ, Shannon Weickert C, Herman MM, Hyde TM, Kleinman JE. Synaptophysin and GAP-43 mRNA levels in the hippocampus of subjects with schizophrenia. *Schizophr Res*. 2001; 49(1-2): 89–98. [PubMed: 11343868]
- Whitfield-Gabrieli S, Thermenos HW, Milanovic S, Tsuang MT, Faraone SV, McCarley RW, Shenton ME, Green AI, Nieto-Castanon A, LaViolette P, Wojcik J, Gabrieli JD, Seidman LJ. Hyperactivity and hyperconnectivity of the default network in schizophrenia and in first-degree relatives of persons with schizophrenia. *Proc Natl Acad Sci U S A*. 2009; 106(4):1279–1284. [PubMed: 19164577]
- Wiedenmann B, Franke WW. Identification and localization of synaptophysin, an integral membrane glycoprotein of Mr 38,000 characteristic of presynaptic vesicles. *Cell*. 1985; 41(3):1017–1028. [PubMed: 3924408]
- Woods SW. Chlorpromazine equivalent doses for the newer atypical antipsychotics. *J Clin Psychiatry*. 2003; 64(6):663–667. [PubMed: 12823080]
- Woodward ND, Karbasforoushan H, Heckers S. Thalamocortical dysconnectivity in schizophrenia. *Am J Psychiatry*. 2012; 169(10):1092–1099. [PubMed: 23032387]
- Young CE, Arima K, Xie J, Hu L, Beach TG, Falkai P, Honer WG. SNAP-25 deficit and hippocampal connectivity in schizophrenia. *Cereb Cortex*. 1998; 8(3):261–268. [PubMed: 9617921]

Figure 1.
Cytoarchitecture and connections of the olfactory bulb.

Figure 2.

Expression of presynaptic proteins in the glomeruli of the olfactory bulb from individuals with schizophrenia and controls. A, C, E) Immunostaining in the entire olfactory bulb and in glomeruli (inset) of a representative matched pair (pair 9) for (A) synapsin IIa, (C) synaptophysin and (E) SNAP-25; B, D, F) comparison of protein expression between individuals with schizophrenia and controls within each matched pair for (B) synapsin IIa, (D) synaptophysin and (F) SNAP-25. SCZ- schizophrenia. Inset scale bars indicate 150µm.

Figure 3.

Expression of postsynaptic proteins and GFAP in olfactory bulb glomeruli from individuals with schizophrenia and controls. A, C, E) Immunostaining in the entire olfactory bulb and in glomeruli (inset) of a representative matched pair (pair 9) for (A) spinophilin, (C) PSD-95 and (E) GFAP; B, D, F) comparison of protein expression between individuals with schizophrenia and controls within each matched pair for (B) spinophilin, (D) PSD-95 and (F) GFAP. SCZ- schizophrenia. Inset scale bars indicate 150 μ m.

Figure 4.

Differences in glomerular expression of pre- and post-synaptic proteins in the olfactory bulb between individuals with schizophrenia and controls. For GFAP and β -actin, mean ODs included glomerular and peri-glomerular areas. SCZ- schizophrenia. * $p < 0.05$, ** $p < 0.005$, N.S.- non-significant. Error bars represent standard error of the mean.

Table 1

Demographic and clinical information for individuals whose olfactory bulbs were used in this study.

Pair #	Diagnosis	Age (years)	Age at diagnosis (years)	Sex	Race	PMI (hr)	Tissue age (years)	Antipsychotic dose / day	Chlorpromazine dose equivalence	Cause of death
1	Schizophrenia	90	35	Female	African American	7.5	10	Risperidone 4mg	400	Hypertensive vasculopathy
2	Schizophrenia	86	24	Female	Caucasian	7.5	12	Unknown	Unknown	Resp failure 2nd to MRSA
3	Schizophrenia	83	19	Female	Caucasian	7.5	11	Risperidone 3mg	300	Pneumonia complications
4	Schizophrenia	90	20	Female	Caucasian	16	18	Thioridazine dose unknown	Unknown	Resp failure
5	Schizophrenia	82	42	Female	Caucasian	12	12	Clozapine 250mg	250	Resp failure 2nd to anemia, DM2, COPD, CVD
6	Schizophrenia	81	19	Male	Caucasian	9	9	Olanzapine 7.5mg	225	Resp failure 2nd to vasculopathy and CVA
7	Schizophrenia	84	Unknown	Male	Caucasian	20	3	Risperidone 6mg	600	Resp failure 2nd to COPD, cardiomyopathy, anemia
8	Schizophrenia	81	19	Male	Caucasian	5	11	Thioridazine 450mg	400	Post fall & head injury
9	Schizophrenia	88	24	Female	Caucasian	7	14	Thioridazine 25mg	25	Chronic Renal Failure
10	Schizophrenia	39	16	Female	African American	17.5	15	Risperidone 8mg	800	Complications of HTN, cardiomyopathy, CHF,
11	Schizophrenia	72	Unknown	Male	Caucasian	26	1	Unknown	Unknown	Unknown
12	Schizophrenia	76	21	Female	Caucasian	10	15	Thiothixene 10mg	200	Metastatic cecal CA, anemia
13	Schizophrenia	70	17	Female	Caucasian	17.5	12	Unknown	Unknown	Resp failure 2nd to COPD
1	Control	92	-	Female	Caucasian	5	11	-		Endometrial CA
2	Control	84	-	Female	Caucasian	3	3	-		CAD/HTN, Leukemia, breast CA, DM
3	Control	75	-	Female	Caucasian	15	3	-		Cardiac arrest 2nd to aortic stenosis
4	Control	97	-	Female	Caucasian	15	3	-		CAD, HTN, heart block
5	Control	78	-	Female	Caucasian	19	2	-		Post-op complications following GI bleed.
6	Control	72	-	Female	Caucasian	13.5	2	-		Acute renal failure 2nd to AA dissection
7	Control	82	-	Male	Caucasian	18	6	-		Cardiac arrest
8	Control	80	-	Male	Caucasian	10.5	7	-		Myocardial Infarction
9	Control	82	-	Female	Caucasian	5	3	-		Unknown
10	Control	46	-	Female	Multiracial	12	2	-		Fulminant hepatic & multi organ failure
11	Control	70	-	Male	Caucasian	36	1	-		Complications of DM & HTN
12	Control	72	-	Female	Caucasian	4	23	-		Unknown
13	Control	71	-	Female	Caucasian	10.5	8	-		Abdominal aortic aneurysm

Author Manuscript

Author Manuscript

Author Manuscript

Author Manuscript

PMI- postmortem interval, Resp- respiratory, 2nd- secondary, MRSA- multidrug resistant staphylococcus aureus, PNA- pneumonia, COPD- chronic obstructive pulmonary disease, CVD- cardiovascular disease, HTN- hypertension, CHF- congestive heart failure, CA- cancer, CAD- coronary artery disease, DM- diabetes mellitus, GI- gastrointestinal, AA- aortic arch.

Table 2

Summary of demographic information.

	Schizophrenia	Control
Age	78.6 ($\pm$ 13.4)	77.5 ($\pm$ 12.3)
PMI	12.5 ($\pm$ 6.3)	12.4 ($\pm$ 9.0)
Tissue age	11 ($\pm$ 4.7)	5.7 ($\pm$ 6.0)

Data represented as group mean ($\pm$ standard deviation). PMI- postmortem interval.