J Neural Transm (2012) 119:661–667
DOI 10.1007/s00702-011-0745-z

MK-801 alters Na^+, K^+-ATPase activity and oxidative status in zebrafish brain: reversal by antipsychotic drugs

Kelly Juliana Seibt · Renata da Luz Oliveira · Denis Broock Rosemberg · Luiz Eduardo Baggio Savio · Emilene B. S. Scherer · Felipe Schmitz · Angela T. S. Wyse · Carla Denise Bonan

Received: 6 September 2011 / Accepted: 20 November 2011 / Published online: 6 December 2011

Abstract Schizophrenia is a debilitating mental disorder with a global prevalence of 1% and its etiology remains poorly understood. In the current study we investigated the influence of antipsychotic drugs on the effects of MK-801 administration, which is a drug that mimics biochemical changes observed in schizophrenia, on Na^+, K^+-ATPase activity and some parameters of oxidative stress in zebrafish brain. Our results showed that MK-801 treatment significantly decreased Na^+, K^+-ATPase activity, and all antipsychotics tested prevented such effects. Acute MK-801 treatment did not alter reactive oxygen/nitrogen species by 2′7′-dichlorofluorscein (H2DCF) oxidation assay, but increased the levels of thiobarbituric acid reactive substances (TBARS), when compared with controls. Some antipsychotics such as sulpiride, olanzapine, and haloperidol prevented the increase of TBARS caused by MK-801. These findings indicate oxidative damage might be a mechanism involved in the decrease of Na^+, K^+-ATPase activity induced by MK-801. The parameters evaluated in this study had not yet been tested in this animal model using the MK-801, suggesting that zebrafish is an animal model that can contribute for providing information on potential treatments and disease characteristics.

Keywords Schizophrenia · Antipsychotics · MK-801 · Na^+, K^+-ATPase · TBARS · DCF

K. J. Seibt · R. da Luz Oliveira · C. D. Bonan (✉)
Programa de Pós-Graduação em Biologia Celular e Molecular Laboratório de Neuroquímica e Psicofarmacologia, Departamento de Biologia Celular e Molecular, Faculdade de Biociências, Pontifícia Universidade Católica do Rio Grande do Sul, Avenida Ipiranga, 6681, Porto Alegre, RS 90619-900, Brazil
e-mail: cbonan@pucrs.br

K. J. Seibt · R. da Luz Oliveira · C. D. Bonan
Instituto Nacional Translacional em Medicina (INCT-TM), Porto Alegre, RS 90035-003, Brazil

D. B. Rosemberg · L. E. B. Savio · E. B. S. Scherer · F. Schmitz · A. T. S. Wyse
Programa de Pós-Graduação em Bioquímica, Departamento de Bioquímica, Instituto de Ciências Básicas da Saúde, Universidade Federal do Rio Grande do Sul, Rua Ramiro Barcelos 2600-Anexo, Porto Alegre, RS 90035-003, Brazil

D. B. Rosemberg
Instituto Nacional de Ciência e Tecnologia em Excitotoxicidade e Neuroproteção (INCT-EN), Porto Alegre, RS 90035-003, Brazil

Introduction

Schizophrenia is a severe neuropsychiatric illness with a lifetime prevalence of approximately 1% whose etiology remains poorly understood (Boison et al. 2011; Yu et al. 2011). The disease is characterized by a broad range of mental and neuropsychological dysfunctions including (i) positive symptoms characterized by functional excesses such as delusions, hallucinations, and disorganized thinking; (ii) affective symptoms, such as depression or mania; (iii) negative symptoms, characterized by loss of normal functions, such as anhedonia, blunted affect, and social withdrawal, and (iv) cognitive symptoms, reflecting deterioration of memory, selective attention, and executive functions (Ross et al. 2006; van Os and Kapur 2009). One of the best characterized animal models of schizophrenia is based on NMDA hypofunction (Farber 2003). This model is based on observations that NMDA antagonists, such as phencyclidine and MK-801, can mimic the complexity of positive, negative, and cognitive symptoms of the disease (Li et al. 2011; Rujescu et al. 2006). MK-801 is a

non-competitive antagonist of NMDA subtype of glutamate receptors and acts by means of open-channel blockade (Stuchlík et al. 2009).

Although the pathophysiological mechanisms underlying this disorder remain unclear, many studies point towards an involvement of oxidative stress in the neuropathological processes of schizophrenia (Gama et al. 2008; Ng et al. 2008; Zhang et al. 2010). Oxidative stress, which results from an impaired redox balance, is supposed to be one of the major causes for schizophrenia (Mahadik and Scheffer 1996; Fenton et al. 2000). Shifting of the redox balance towards oxidative stress may occur due to excessive generation of free radicals, decreased antioxidant activities, or a combination of both (Singh et al. 2008). Recent findings indicate the role of changes of membrane phospholipids and fatty acids induced by oxidative stress in etiopathogenic mechanisms in schizophrenia (Fendri et al. 2006; Yao et al. 2001). Lipid peroxidation assessed by TBARS was shown to be increased in the plasma of drug-free and medicated schizophrenic patients (Akyol et al. 2002).

Na^+, K^+-ATPase (EC 3.6.1.37), also known as the sodium pump, is a major membrane protein responsible for generating the membrane potential. The active transport of Na^+ and K^+ ions in the central nervous system (CNS) is indispensable to regulate neuronal excitability and cellular volume (Kaplan 2002; Aperia 2007). This enzyme consumes about 40–60% of ATP produced in brain (Erecinska et al. 2004). Disturbance in Na^+, K^+-ATPase density and/or activity might induce significant damage on brain function. Studies show that its activity is altered in various brain disorders, such as ischemia (Wyse et al. 2000), neurodegenerative (Yu 2003; Vignini et al. 2007) and neuropsychiatric diseases (Goldstein et al. 2006). In addition, a decline of Na^+, K^+-ATPase activity has been attributed to induce lipid changes and associated oxidative damage (Chakraborty et al. 2003; Dencher et al. 2007).

Zebrafish represents an attractive vertebrate model in Developmental Biology, Genetics, Pharmacology, and Neuroscience (Gerlai et al. 2000; Grossman et al. 2010; Cachat et al. 2010). The zebrafish have many inherent advantages as a model organism, such as low cost, easy handling and maintenance as compared with other vertebrate models, and 70–80% genetic homology to humans (Barbazuk et al. 2000; Egan et al. 2009). Behavior-based chemical screens in zebrafish may improve our understanding of neurobiology and drug action and accelerate the pace of psychiatric drug discovery (Kokel and Peterson 2008; Rihel et al. 2010). Considering (i) oxidative stress is likely to be involved in the pathophysiology of schizophrenia, (ii) Na^+, K^+-ATPase activity is critical for normal brain function, being altered in many disorders, and (iii) zebrafish has been used as a model for studying several diseases, we investigate the effect of MK-801, a drug used for eliciting schizophrenia-like symptoms, on Na^+, K^+-ATPase activity and reactive oxygen/nitrogen species levels in zebrafish brain. The protective effect of antipsychotic drugs was also evaluated.

Materials and methods

Animals

Adult wild-type zebrafish strains (3–5 cm) of both sexes were obtained from a specialized commercial supplier (Redfish, RS, Brazil) and were of genetically heterogeneous (randomly bred) stock. The fish were acclimatized to the laboratory environment for at least 14 days and housed in a 50-L thermostated aquarium filled with continuously unchlorinated water at a targeted temperature of 28 ± 2°C, with constant filtration and aeration (7.20 mg O_2/L) and a density of up to five animals per liter (Westerfield 2007). Animals were kept on a day:night cycle of 14:10 h and fed twice a day with flaked fish food that was supplemented with live brine shrimp.

Fish were manipulated healthy and free of any signs of disease, according to the ‘‘Guide for the Care and Use of Laboratory Animals’’ published by the US National Institutes of Health (NIH publication No 85–23, revised 1996). The ethics committee of the Pontifical Catholic University of Rio Grande do Sul (PUCRS) approved the protocol under license number CEUA 09/00135.

Pharmacological treatments

A group of five animals were transferred and kept in 500-mL beakers for 30 min divided in two consecutive 15-min periods, as follows: (a) In the first 15-min exposure period, animals were exposed to tank water or 20 μM MK-801; (b) During the final 15-min-exposure, animals were treated with tank water or with one of the following antipsychotic drugs: 9 μM haloperidol (HAL), 100 μM olanzapine (OLA), or 250 μM sulpiride (SUL). Therefore, the following experimental groups were tested: (i) water plus water, (ii) water plus HAL, (iii) water plus OLA, (iv) water plus SUL, (v) MK-801 plus water, (vi) MK-801 plus HAL, (vii) MK-801 plus OLA (viii) MK-801 plus SUL. The MK-801 and haloperidol doses were chosen based on previous studies with zebrafish (Giacomini et al. 2006; Swain et al. 2004; Seibt et al. 2010). Moreover, our group showed that haloperidol, sulpiride, and olanzapine per se did not induce changes in the locomotion in zebrafish (Seibt et al. 2010).

Membrane preparation

Brain membranes were prepared according to the method previously described (Barnes et al. 1993). Briefly, whole zebrafish brains were homogenized in 60 volumes (v/w) of chilled Tris-citrate buffer (50 mM Tris, 2 mM EDTA, 2 mM EGTA, pH 7.4, adjusted with citric acid) in a glass-Teflon homogenizer. The homogenate was centrifuged at 1,000*g* for 10 min and the pellet was discarded. After removing the nuclear and cell debris, the supernatant was centrifuged for 25 min at 40,000*g*. The resultant pellet was frozen in liquid nitrogen, thawed, resuspended in Tris-citrate buffer, and centrifuged for 20 min at 40,000*g*. The final pellet was resuspended and used for the enzyme assays. The material was maintained at 2–4°C throughout preparation.

Protein determination

Protein was measured by the Coomassie Blue method (Bradford 1976), using bovine serum albumin as standard.

Na^+K^+ATPase activity

Na^+, K^+-ATPase activity was determined as previously reported (Tsakiris and Deliconstantinos 1984). The enzyme preparation (3–5 μg protein) was added to the reaction mixture containing 5 mM $MgCl_2$, 80 mM NaCl, 20 mM KCl, and 40 mM Tris–HCl buffer, pH 7.4, in a final volume of 200 μL. The reaction was started by the addition of ATP (vanadium-free disodium salt) to a final concentration of 3 mM and stopped after 5 min with addition of 200 μL trichloroacetic acid 10%. The control was assessed under the same conditions, except that 1 mM ouabain was added to the reaction medium. The Na^+, K^+-ATPase activity was calculated by determining the difference between these two assays. The released inorganic phosphate (Pi) was measured by the colorimetric Malachite Green method (Chan et al. 1986) and results were expressed as nmol Pi/min/mg protein.

Measurement of lipid peroxidation

Lipid peroxidation was measured through determination of thiobarbituric acid reactive substances (TBARS) according to the colorimetric assay previously described (Ohkawa et al. 1979). Briefly, samples and reagents were added in the following order: 100 μL of tissue supernatant; 25 μL of SDS 8.1%; 190 μL of 20% acetic acid in aqueous solution (v/v) pH 3.5; and 190 μL of 0.8% thiobarbituric acid. The mixture was vortexed and the reaction was carried out in a boiling water bath for 1 h. TBARS were determined by the absorbance at 535 nm and calculated as nmol of malondialdehyde (MDA) formed per milligram of protein.

Dichlorofluorescein (H2DCF) oxidation assay

Reactive oxygen/nitrogen species production was measured following the method based on 2′7′-dichlorofluorscein oxidation (Lebel et al. 1992). Briefly, homogenates from total zebrafish brain (60 μL) were incubated for 30 min at 37°C in the dark with 240 μL of 100 μM 2′7′-dichlorofluorscein diacetate solution in a 96-well plate. H2DCF-DA is cleaved by cellular esterases and H2DCF formed is eventually oxidized by ROS or RNS present in samples. The last reaction produces the fluorescent compound DCF, which was measured at 488 nm excitation and 525 nm emission, and the results were represented by nmol DCF/mg protein. A calibration curve was performed with purified DCF as standard.

Statistical analysis

Data were expressed as means ± SEM. The Na^+, K^+-ATPase activity and TBARS data were analyzed by two-way analysis of variance (ANOVA), followed by Bonferroni's multiple test range as post hoc. The DCFH data was analyzed by one-way analysis of variance (ANOVA), followed by Tukey's test. A value of $p < 0.05$ was considered significant.

Results

At first, we tested 20 μM MK-801 during 15 and 30 min of exposure in order to determine the time exposure to further experiments related to antipsychotic effects. Our results demonstrated that there was a decrease [52.5%; $F(2,9) = 34.56$; $p < 0.01$] of Na^+, K^+-ATPase activity in the group submitted to 15 min of exposure to MK-801 (195 ± 33 nmol Pi. min^{-1} mg^{-1} protein) in relation to the control group (410 ± 52 nmol Pi. min^{-1} mg^{-1} protein). However, there were no changes on Na^+, K^+-ATPase activity after 30-min MK-801 exposure (405 ± 13 nmol Pi. min^{-1} mg^{-1} protein) when compared with control group (410 ± 52 nmol Pi. min^{-1} mg^{-1} protein). Therefore, we have chosen 15 min-exposure to MK-801 for the subsequent experiments with antipsychotics.

We investigated the effect of acute administration of MK-801 and antipsychotic drugs on Na^+, K^+-ATPase activity. Figure 1 shows that MK-801/water significantly decreased Na^+, K^+-ATPase activity [$F(1,11) = 13.18$, $p < 0.002$]. Water/sulpiride and water/olanzapine alone did not alter this parameter. However, water/haloperidol treatment significantly increased Na^+, K^+-ATPase activity [$F(3,23) = 18{,}91$, $p < 0.001$]. In addition, the treatment with MK-801/olanzapine [$F(3,23) = 93.64$, $p < 0.0001$], MK-801/sulpiride [$F(3,23) = 25.64$, $p < 0.0001$], and

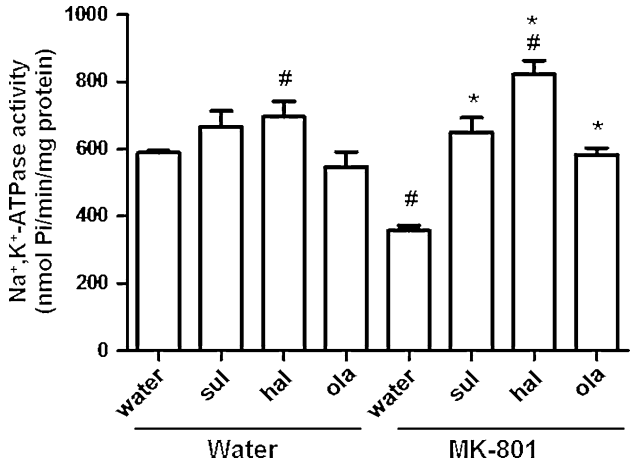

Fig. 1 Effect of MK-801 and antipsychotic drug treatment on Na^+, K^+-ATPase activity in zebrafish brain. Data are expressed as mean ± SEM for 5–7 animals in each group. #Significant difference compared to water/water. *Significant difference compared to MK-801/water

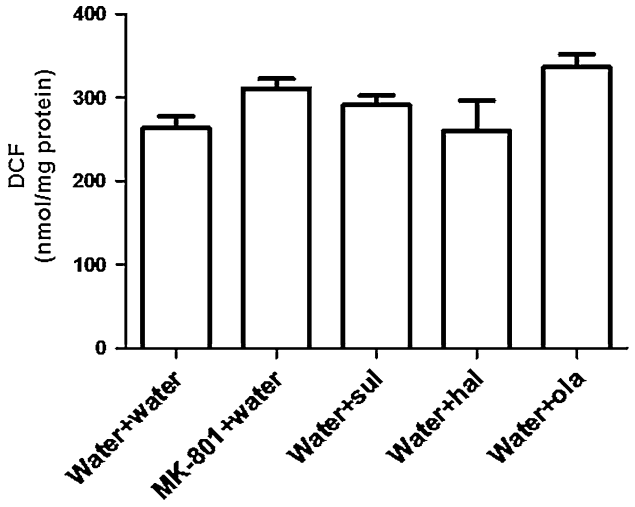

Fig. 2 Effect of acute administration of antipsychotic drugs and MK-801 on the reactive species levels in zebrafish brain. *Data* are expressed as mean ± SEM for 5–7 animals in each group. Results are expressed in nmol/mg protein

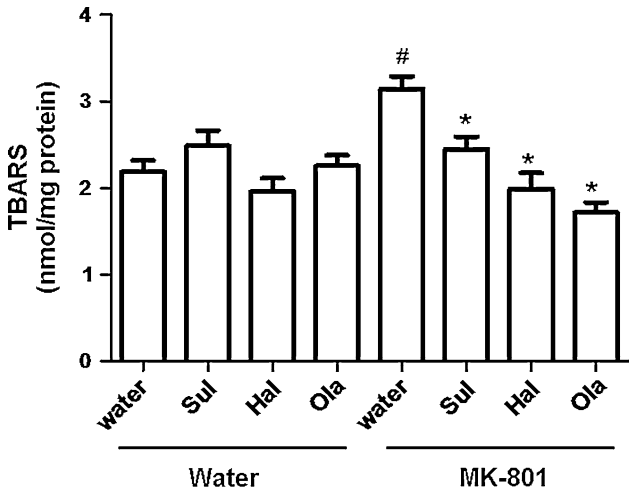

Fig. 3 In vivo effect of acute administration of antipsychotic drugs and MK-801 on TBARS in zebrafish brain. Data are expressed as mean ± SEM for 5–7 animals in each group. Results are expressed in nmol MDA/mg protein. #Significant difference compared with water/water. *Significant difference compared with MK-801/water

MK-801/haloperidol [$F(3,23) = 68.95$, $p < 0.0001$] reverted MK-801/water-induced effects on Na^+, K^+-ATPase activity. Only MK-801/olanzapine and MK-801/sulpiride maintained the enzyme activity at the same levels of control group. DMSO 5%, used as the vehicle to sulpiride, was tested alone and together with MK-801 and Na^+, K^+-ATPase activity was not altered (data not shown).

We also investigated the effect of MK-801/water on some parameters of oxidative stress, such as production of reactive oxygen/nitrogen species and TBARS in zebrafish brain. Figure 2 shows that MK-801/water and water/antipsychotic drugs did not change the levels of this marker in zebrafish brain [$F(4,27) = 2.750$; $P > 0.09$]. Figure 3 shows that acute MK-801/water administration significantly increased TBARS levels in zebrafish brain, when compared with controls (water-water treated) [$F(1,11) = 8.68$; $P < 0.01$]. Our data also showed that the treatment with MK-801/antipsychotic drugs, sulpiride [$F(3,23) = 10.31$; $P < 0.01$], haloperidol [$F(3,23) = 8.53$; $P < 0.01$], and olanzapine [$F(3,23) = 31.07$; $P < 0.001$], reverted this effect (Fig. 3).

Discussion

Schizophrenia is a brain disorder that has been intensively studied for over a century; however, its etiology and multifactorial pathophysiology remain a puzzle (Yao and Keshavan 2011). Previous studies have demonstrated that Na^+, K^+-ATPase activity may be altered in both neurodegenerative (Grisar 1984; Wyse et al. 2000) and psychiatric disorders (Kurup and Kurup 2002; Goldstein et al. 2006).

Na^+, K^+-ATPase is a membrane enzyme responsible for the generation of the membrane potential through the active transport of sodium and potassium ions in the CNS indispensable to maintain neuronal excitability, present at elevated concentrations in brain (Aperia 2007). Previous studies have shown a significant decrease in Na^+, K^+-ATPase activity in patients with schizophrenia (Rybakowski and Lehmann, 1994; Kurup and Kurup 2002). In the present study, we evaluated the effects of MK-801 and antipsychotic drugs on Na^+, K^+-ATPase activity in zebrafish brain. Our results showed that MK-801 decreased Na^+, K^+-ATPase activity and that antipsychotic

drugs (haloperidol, sulpiride and olanzapine) reverted this effect. Our findings have also shown that haloperidol administration increased Na^+, K^+-ATPase activity in zebrafish brain. These data are consistent with previous studies that showed haloperidol increased Na^+, K^+-ATPase activity in the rat brain (Wood et al. 1989). Other studies have demonstrated similar effects on other drugs, such as amphetamine (Zugno et al. 2009), fluoxetine (Zanatta et al. 2001), selegiline (Carageorgiou et al. 2003), carbamazepine, and lithium (Wood et al. 1989).

Oxidative stress occurs when there is an imbalance between reactive oxygen species (ROS) generation and antioxidant defenses in favor of the former. There are multiple pathological consequences of increased ROS production. Oxidative stress and the oxidative changes in different biomolecules may be involved in the pathology of schizophrenia (Berk et al. 2008). It has been verified that Na^+, K^+-ATPase is particularly susceptible to free radical attack since its inhibition has been correlated with changes in plasma membrane lipid composition (Dencher et al. 2007), and in other amino acid residues caused by free radicals or lipid peroxidation (Potts et al. 2006; Siems et al. 1996).

To verify whether reactive oxidative stress could play a role on MK-801-mediated effects, TBARS and H_2DCF assays were performed. Our results showed that acute administration of MK-801 and antipsychotic drugs did not alter the 2′,7′-Dichlorofluorescein oxidation. However, the results showed that MK-801 increased TBARS levels in brain from zebrafish treated with MK-801. We also observed that administration of antipsychotic drugs after MK-801 treatment significantly reversed the increase on TBARS levels. These data are consistent with previous studies showing that oxidative stress and alterations in antioxidant enzymes have long been described in the pathophysiology of schizophrenia (Reddy and Yao 1996; Grignon and Chianetta 2007). In addition, Dietrich-Muszalska et al has shown that the level of TBARS was significantly increased in plasma of patients with schizophrenia (Dietrich-Muszalska et al. 2005; Dietrich-Muszalska and Olas 2007), whereas the activities of antioxidant defense enzymes were diminished (Dietrich-Muszalska and Olas 2007). Other studies have shown that chronic treatment with antipsychotics increased free radical production and oxidative stress (Balijepalli et al. 2001). In addition, chronic use of antipsychotics is also reported to cause a decrease in the activity of antioxidant enzymes, superoxide dismutase, and catalase (Cadet et al. 1987).

In summary, we demonstrated that MK-801 administration decreased Na^+, K^+-ATPase activity in zebrafish brain and that distinct typical and atypical antipsychotics reverted this effect. The same effect is also evaluated when testing the TBARS parameter of oxidative stress. Together, these findings support the involvement of reactive oxygen/nitrogen species and/or lipid peroxidation in the MK-801-elicited effects on Na^+, K^+-ATPase. This data show that treatment with MK-801 in zebrafish might mimic some biochemical changes observed in schizophrenic patients, suggesting that the zebrafish is an animal model that can contribute for providing information about potential treatments and disease characteristics.

Acknowledgments This work was supported by Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) and Fundação de Amparo à Pesquisa do Estado do Rio Grande do Sul (FAPERGS, Proc. 1018580). And by DECIT/SCTIE-MS through CNPq and FAPERGS, (Proc. 10/0036-5 – PRONEX/Conv. 700545/2008).

References

Akyol O, Herken H, Uz E, Fadillioglu E, Unal S, Sogutt S, Ozyurt H, Savaş HÁ (2002) The indices of endogenous oxidative and antioxidative processes in plasma from schizophrenic patients. The possible role of oxidant/antioxidant imbalance. Prog Neuropsychopharmacol Biol Psychiatry 26:995–1005

Aperia A (2007) New roles for an old enzyme: Na, K-ATPase emerges as an interesting drug target. J Intern Med 261:44–52

Balijepalli S, Kenchappa RS, Boyd MR, Ravindranath V (2001) Protein thiol oxidation by haloperidol results in inhibition of mitochondrial complex I in brain regions: comparison with atypical antipsychotics. Neurochem Int 38:425–435

Barbazuk WB, Korf I, Kadavi C, Heyen J, Tate S, Wun E, Bedell JA, McPherson JD, Johnson SL (2000) The syntenic relationship of the zebrafish and human genomes. Genome Res 10:1351–1358

Barnes JM, Murphy PA, Kirkham D, Henley JM (1993) Interaction of guanine nucleotides with [3H]kainate and 6-[3H]cyano-7-nitroquinoxaline-2,3-dione binding in goldfish brain. J Neurochem 61:1685–1691

Berk M, Copolov D, Dean O, Lu K, Jeavons S, Schapkaitz I, Anderson-Hunt M, Judd F, Katz P, Ording-Jespersen S, Little J, Conus P, Cuenod M, Do KQ, Bush AI (2008) *N*-acetyl cysteine as a glutathione precursor for schizophrenia–A double-blind, randomized, placebo- controlled trial. Biol Psychiatry 64:361–368

Boison D, Singer P, Shen HY, Feldon J, Yee BK (2011) Adenosine hypothesis of schizophrenia—Opportunities for pharmacotherapy. Neuropharmacology (Epub ahead of print)

Bradford MM (1976) A rapid and sensitive method for the quantification of microgram quantities of protein utilizing the principle of protein-dye binding. Anal Biochem 72:218–254

Cachat J, Stewart A, Grossman L, Gaikwad S, Kadri F, Chung KM, Wu N, Wong K, Roy S, Suciu C, Goodspeed J, Elegante M, Bartels B, Elkhayat S, Tien D, Tan J, Denmark A, Gilder T, Kyzar E, Dileo J, Frank K, Chang K, Utterback E, Hart P, Kalueff AV (2010) Measuring behavioral and endocrine responses to novelty stress in adult zebrafish. Nat Protoc 5(11):1786–1799

Cadet JL, Lohr JB, Jeste DV (1987) Tardive dyskinesia and schizophrenic burnout: the possible involvement of cytotoxic free radicals. In: Henn FA, DeLisi LE (eds) Handbook of Schizophrenia: the neurochemistry and pharmacology of Schizophrenia. Amsterdam, Elsevier, pp 425–438

Carageorgiou H, Zarros A, Tsakiris S (2003) Selegiline long-term effects on brain acetylcholinesterase, Na^+, K^+-ATPase activities, antioxidant status and learning performance of aged rats. Pharmacol Res 48:245–251

Chakraborty H, Sen P, Sur A, Chatterjee U, Chakrabarti S (2003) Age-related oxidative inactivation of Na^+, K^+-ATPase in rat brain crude synaptosomes. Exp Gerontol 38:705–710

Chan KM, Delfer D, Junger KD (1986) A direct colorimetric assay for Ca2+-stimulated ATPase activity. Anal Biochem 157:375–380

Dencher NA, Frenzel M, Reifschneider NH, Sugawa M, Krause F (2007) Proteome alterations in rat mitochondria caused by aging, Ann. NY Acad Sci 1100:291–298

Dietrich-Muszalska A, Olas B (2007) Isoprostanes as indicators of oxidative stress in schizophrenia. World J Biol Psychiatry 4:1–6

Dietrich-Muszalska A, Olas B, Rabe-Jablonska J (2005) Oxidative stress in blood platelets from schizophrenic patients. Platelets 16:386–391

Egan RJ, Bergner CL, Hart PC, Cachat JM, Canavello PR, Elegante MF, Elkhayat SI, Bartels BK, Tien AK, Tien DH, Mohnot S, Beeson E, Glasgow E, Amri H, Zukowska Z, Kalueff AV (2009) Understanding behavioral and physiological phenotypes of stress and anxiety in zebrafish. Behav Brain Res 205(1): 38–44

Erecinska M, Cherian S, Silver IA (2004) Energy metabolism in mammalian brain during development. Prog Neurobiol 73:397–445

Farber NB (2003) The NMDA receptor hypofunction model of psychosis. Ann NY Acad Sci 1003:119–130

Fendri C, Mechri A, Khiari G, Othman A, Kerkeni A, Gaha L (2006) Oxidative stress involvement in schizophrenia pathophysiology. Encephale 32:244–252

Fenton WS, Hibbeln J, Knable M (2000) Essential fatty acids, lipid membrane abnormalities, and the diagnosis and treatment of schizophrenia. Biol Psychiatry 47:8–21

Gama CS, Salvador M, Andreazza AC, Lobato MI, Berk M, Kapczinski F, Belmonte-de-Abreu PS (2008) Elevated serum thiobarbituric acid reactive substances in clinically symptomatic schizophrenic males. Neurosci Lett 433(3):270–273

Gerlai R, Lahav M, Guo S, Rosenthal A (2000) Drinks like a fish: zebra fish (*Danio rerio*) as a behavior genetic model to study alcohol effects. Pharmacol Biochem Behav 67(4):773–782

Giacomini NJ, Rose B, Kobayashi K, Guo S (2006) Antipsychotics produce locomotor impairment in larval zebrafish. Neurotoxicol Teratol 28:245–250

Goldstein I, Levy T, Galili D, Ovadia H, Yirmiya R, Rosen H, Lichtstein D (2006) Involvement of Na^+, K^+-ATPase and endogenous digitalis-like compounds in depressive disorders. Biol Psychiatry 60:491–499

Grignon S, Chianetta JM (2007) Assessment of malondialdehyde levels in schizophrenia: a meta-analysis and some methodological considerations. Prog Neuropsychopharmacol Biol Psychiatry 31(2):365–369

Grisar T (1984) Glial and neuronal Na^+-K^+ pump in epilepsy. Ann Neurol 16:S128–S134

Grossman L, Utterback E, Stewart A, Gaikwad S, Chung KM, Suciu C, Wong K, Elegante M, Elkhayat S, Tan J, Gilder T, Wu N, Dileo J, Cachat J, Kalueff AV (2010) Characterization of behavioral and endocrine effects of LSD on zebrafish. Behav Brain Res 214(2):277–284

Jianfeng Yu, Dake Qi, Mengjuan Xing, Ruixi Li, Kaida Jiang, Yuwen Peng, Donghong Cui (2011) MK-801 induces schizophrenic behaviors through downregulating Wnt signaling pathways in male mice. Brain Res 1385:281–292

Kaplan JH (2002) Biochemistry of Na, K-ATPase. Annu Ver Biochem 71:511–535

Kokel D, Peterson RT (2008) Chemobehavioural phenomics and behaviour-based psychiatric drug discovery in the zebrafish. Brief Funct Genomic Proteomic 7(6):483–490

Kurup AR, Kurup PA (2002) Membrane Na^+, K^+-ATPase mediated cascade in bipolar mood disorder, major depressive disorder, and schizophrenia-relationship to hemispheric dominance. Int J Neurosci 112:965–982

Lebel CP, Ischiropoulos H, Bondy SC (1992) Evaluation of the probe 2′,7′-dichlorofluorescin as an indicator of reactive oxygen species formation and oxidative stress. Chem Res Toxicol 5:227–231

Li JT, Su YA, Guo CM, Feng Y, Yang Y, Huang RH et al (2011) Persisting cognitive deficits induced by low-dose, subchronic treatment with MK-801 in adolescent rats. Eur J Pharmacol 652(1–3):65–72

Mahadik SP, Scheffer RE (1996) Oxidative injury and potential use of antioxidants in schizophrenia. Prostaglandins Leukot Essent Fatty Acids 55:45–54

Ng F, Berk M, Dean O, Bush AI (2008) Oxidative stress in psychiatric disorders: evidence base and therapeutic implications. Int J Neuropsychopharmacol 11(6):851–876

Ohkawa H, Ohishi N, Yagi K (1979) Assay for lipid peroxides in animal tissues by thiobarbituric acid reaction. Anal Biochem 95:351–358

Potts MB, Koh SE, Whetstone WD, Walker BA, Yoneyama T, Claus CP, Manvelyan HM, Noble-Haeusslein LJ (2006) Traumatic injury to the immature brain: inflammation, oxidative injury, and iron-mediated damage as potential therapeutic targets. NeuroRx 3:143–153

Reddy RD, Yao JK (1996) Free radical pathology in schizophrenia: a review. Prostaglandins Leukot Essent Fatty Acids 55(1–2):33–43

Rihel J, Prober DA, Arvanites A, Lam K, Zimmerman S, Jang S, Haggarty SJ, Kokel D, Rubin LL, Peterson RT, Schier AF (2010) Zebrafish behavioral profiling links drugs to biological targets and rest/wake regulation. Science 327(5963):348–351

Ross CA, Margolis RL, Reading SA, Pletnikov M, Coyle JT (2006) Neurobiology of schizophrenia. Neuron 52(1):139–153 Review

Rujescu D, Bender A, Keck M, Hartmann AM, Ohl F, Raeder H et al (2006) A pharmacological model for psychosis based on N-methyl-d-aspartate receptor hypofunction: molecular, cellular, functional and behavioral abnormalities. Biol Psychiatry 59:721–729

Rybakowski JK, Lehmann W (1994) Decreased activity of erythrocyte membrane ATPases in depression and schizophrenia. Biol Psychiatry 30:11–14

Seibt KJ, Oliveira Rda L, Zimmermann FF, Capiotti KM, Bogo MR, Ghisleni G, Bonan CD (2010) Antipsychotic drugs prevent the motor hyperactivity induced by psychotomimetic MK-801 in zebrafish (*Danio rerio*). Behav Brain Res 214(2):417–422

Siems WG, Hapner SJ, van Kuijk F (1996) 4-hydroxynonenal inhibits Na(+)–K(+)-ATPase, Free Radic. Biol Med 20:215–223

Singh OP, Chakraborty I, Dasgupta A, Datta S (2008) A comparative study of oxidative stress and interrelationship of important antioxidants in haloperidol and olanzapine treated patients suffering from schizophrenia. Indian J Psychiatry 50(3):171–176

Stuchlík A, Petrásek T, Vales K (2009) Effect of alpha(1)-adrenergic antagonist prazosin on behavioral alterations induced by MK-801 in a spatial memory task in Long-Evans rats. Physiol Res 58:733–740

Swain HA, Sigstad C, Scalzo FM (2004) Effects of dizocilpine (MK-801) on circling behavior, swimming activity, and place preference in zebrafish (*Danio rerio*). Neurotoxicol Teratol 26:725–729

Tsakiris S, Deliconstantinos G (1984) Influence of phosphatidylserine on ($Na^+ + K^+$)-stimulated ATPase and acetylcholinesterase activities of dog brain synaptosomal plasma membranes. Biochem J 220(1):301–307

van Os J, Kapur S (2009) Schizophrenia. Lancet 374(9690):635–645

Vignini A, Nanetti L, Moroni C, Tanase L, Bartolini M, Luzzi S, Provinciali L, Mazzanti L (2007) Modifications of platelet from Alzheimer disease patients: a possible relation between

membrane properties and NO metabolites. Neurobiol Aging 28:987–994

Westerfield M (2007) The zebrafish book, 5th edn. University of Oregon Press, Eugene

Wood AJ, Elphick M, Grahame-Smith DG (1989) Effect of lithium and of other drugs used in the treatment of manic illness on the cation-transporting properties of Na^+, K^+-ATPase in mouse brain synaptosomes. J Neurochem 52(4):1042–1049

Wyse ATS, Streck EL, Worm P, Wajner A, Ritter F, Netto CA (2000) Preconditioning prevents the inhibition of Na^+, K^+-ATPase activity after brain ischemia. Neurochem Res 25:969–973

Yao JK, Keshavan MS (2011) Antioxidants, redox signaling, and pathophysiology in schizophrenia: an integrative view. Antioxid Redox Signal 15(7):2011–2035

Yao JK, Reddy RD, van Kammen DP (2001) Oxidative damage and schizophrenia: an overview of the evidence and its therapeutic implications. CNS Drugs 15:287–310

Yu SP (2003) Na^+, K^+-ATPase: the new face of an old player in pathogenesis and apoptotic/hybrid cell death. Biochem Pharmacol 66:1601–1609

Zanatta LM, Nascimento FC, Barros SV, Silva GR, Zugno AI, Netto CA, Wyse AT (2001) In vivo and in vitro effect of imipramine and fluoxetine on Na^+, K^+-ATPase activity in synaptic plasma membranes from the cerebral cortex of rats. Braz J Med Biol Res 34:1265–1269

Zhang M, Zhao Z, He L, Wan C (2010) A meta-analysis of oxidative stress markers in schizophrenia. Sci China Life Sci 53(1): 112–124

Zugno AI, Valvassori SS, Scherer EB, Mattos C, Matté C, Ferreira CL, Rezin GT, Wyse AT, Quevedo J, Streck EL (2009) Na^+, K^+-ATPase activity in an animal model of mania. J Neural Transm 116(4):431–436