

Review

Sphingolipids in Major Depression

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Key Words

Neurogenesis • Ceramide • Acid sphingomyelinase • Major depression

Abstract

Major depression is one of the most common and severe diseases affecting the world's population. However, the pathogenesis of the disease remains inadequately defined. Previously, a lack of monoaminergic neurotransmitters was the focus of pathophysiological concepts; however, recent concepts focus on an alteration of neurogenesis in the hippocampus. This concept suggests that neurogenesis is decreased in major depression with a rarefaction of neuronal networks and a lack of new, immature neurons in the hippocampus, events that may result in the clinical symptoms of major depression. However, molecular targets involved in the pathogenesis of major depression and, in particular, a reduction of neurogenesis, are largely unknown. We have recently discovered that an inhibition of the acid sphingomyelinase/ceramide system mediates the effects of tri- and tetracyclic antidepressants. Moreover, an accumulation of ceramide in the hippocampus results in depression-like symptoms. This suggests the acid sphingomyelinase/ceramide system is very important in the pathogenesis of major depression.

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Pathophysiological models of major depression

With a lifetime prevalence of more than 10% in the overall population and an estimated suicide rate of 10%, major depressive disorder is one of the most severe chronic illnesses [1]. Patients with major depression suffer not only from depressed mood, loss of interest, anhedonia, fear, feelings of worthlessness, weight loss, insomnia, and concentration deficits,

but also from cardiovascular symptoms, osteoporosis, adrenocortical activation and a general pro-inflammatory status [2-9].

Although major depression is a very common disease, the pathogenesis is presently still unknown. Many patients show high plasma concentrations of cortisol and dysfunction of the hypothalamic-pituitary-adrenal axis [1]. However, the molecular mechanisms causing this increase as well as the pathophysiological consequences of these changes are still poorly characterized. A recent concept indicated a lack of neurogenesis in the hippocampus [10-20], a concept that will be discussed in more detail below. Surprisingly, even the molecular targets of antidepressants are as of yet unclear. Most antidepressants regulate the concentrations of monoaminergic transmitters in the synaptic space, and the effect of antidepressants on the monoaminergic transporters in the synaptic membrane has been considered the active mechanism of these drugs [21]. However, this hypothesis has been questioned since (i) some antidepressants, in particular tianeptine, promote serotonin reuptake rather than block it [22, 23], (ii) the regulation of synaptic uptake of monoamines is very rapid after treatment, while the clinical effect of antidepressants is usually delayed by 2 to 4 weeks and (iii) anti-inflammatory therapies also show therapeutic effects in major depression, which is difficult to explain with the monoaminergic neurotransmitter hypothesis of antidepressants [4, 5, 24].

In particular the delay of 2-4 weeks in therapeutic effects of many antidepressants suggests a trophic effect of antidepressants and, therefore, recent concepts of the pathogenesis of major depression have focused on a defect in neurogenesis and the correct function of neuronal networks in the hippocampus to explain the pathophysiology of major depressive disorder. The hypothesis that hippocampal neurogenesis is reduced in major depression is also supported by the finding that at least in some cases major depression also leads to hippocampal atrophy [25]. The atrophy seems to be primarily caused by a reduction in the number of glial cells rather than in the number of neurons [26]. The role of glial cells in the genesis of major depression is largely unknown. However, a concept of an imbalance between neurogenesis and possibly glial cell genesis and apoptotic events in the hippocampus has become very attractive to explain many of the pathophysiological findings. It should be noted that the molecular cause for the defect of neuro- and gliogenesis is also still unknown.

Mammalian brains show two hotspots of neurogenesis- the subventricular zone of the lateral ventricles and the hippocampal dentate gyrus [27-33]. However, newborn neurons are able to migrate, for instance into the olfactory bulb [29-31] or the striatum [34]. In the hippocampus, the newborn, immature neurons migrate to and differentiate in the granular cell layer, a process that requires 3 to 4 weeks. This time frame is very similar to the delayed action of many antidepressants and supports the notion that reconstitution of neurogenesis and correct integration and/or formation of neuronal networks is required to treat major depression [30, 35-38]. The molecular mechanisms mediating proliferation of neuronal and glial stem cells are largely unknown. Thus, it was shown that low doses of reactive oxygen species, a reduction of cellular ceramide by inhibition of the acid sphingomyelinase and an activation of phosphatidylinositol-3-kinase (PI3K), Akt, Erk1/2, Wnt3a, Notch molecules and cAMP response element-binding protein (CREB) trigger neurogenesis [39-47], but the molecular details of these signaling molecules and pathways in neurogenesis warrant further research.

Consistent with the hypothesis that hippocampal neurogenesis is impaired in major depression, it was demonstrated that antidepressants such as fluoxetine, desipramine, imipramine, and amitriptyline induce neurogenesis of cultured neurons *in vitro*, but more importantly also *in vivo* in the hippocampus [10-12, 15, 16, 18, 19]. The latency of antidepressant-induced neurogenesis is consistent with the delayed action of these drugs. Neurogenesis correlated with the improvement of depressive-like behavior in mouse models which was blocked by irradiation of the hippocampus [19], suggesting that neuronal proliferation is not just a simple readout of changes in the brain during major depression, but rather causative in the pathogenesis of the disease.

On the other hand, clinical experience indicates that irradiation of the brain or general chemotherapy blocking proliferation of stem cells does not necessarily result in major depression. Further, treatment of major depression with sleep deprivation or electroconvulsive therapy shows fast effects that are inconsistent with neurogenesis, maturation and integration into or even the formation of neuronal networks of newly formed neurons. Thus while neurogenesis may be required for the effects of antidepressants, it may not be sufficient to overcome major depression. Thus, there are likely unknown mechanisms that are disturbed and must be targeted by antidepressants. However, massive formation of immature neurons, for instance after electroconvulsive therapy [48] may change the excitability of the hippocampus and the limbic system [49]. In addition newborn neurons might negatively regulate the hypothalamic-pituitary-adrenal axis, which seems to be overactive in many patients with major depression [50, 51]. Such a negative feedback system between immature neurons in the hippocampus and the hypothalamic-pituitary-adrenal axis may allow an adequate response to stress and therefore be important for the treatment of major depression [51, 52].

Finally, stress and glucocorticoids reduce, and antidepressants as well as electroconvulsive therapy induce, the production of a variety of growth factors, such as brain-derived neurotrophic factor (BDNF), vascular endothelial growth factor (VEGF), and nerve growth factor (NGF), which act on neurons and on vascular endothelial cells and thereby induce neurogenesis and angiogenesis [26, 53, 54]. An improved coupling of neuronal stem cells and the vascular cells in the stem cell niche might promote neurogenesis and thereby reduce symptoms of major depression.

As discussed below, antidepressants and many growth factors inhibit the acid sphingomyelinase/ceramide system. It might be possible that antidepressants have a relatively weak effect on that system, while growth factors released by electroconvulsive therapy might exhibit a strong inhibition of the acid sphingomyelinase/ceramide system. If inhibition of the hippocampal ceramide concentrations under a certain critical level is required for treatment of major depression, the strong effects of electroconvulsive therapy have a fast therapeutic effect, while the lower potency of antidepressants to inhibit the acid sphingomyelinase only slowly reduces the concentration of hippocampal ceramide explaining the delayed onset of the action of these drugs. If ceramide-levels are too high to be reduced by antidepressants under that critical level, the patient would fail to respond to therapy. This model is certainly speculative, and has to be proven *in vivo*, but it may explain many of the discrepancies between the fast action of electroconvulsive therapy and the slow onset of action of antidepressants.

Role of the acid sphingomyelinase/ceramide system in major depression

We have previously shown an important role of the acid sphingomyelinase (EC 3.1.4.12, sphingomyelin phosphodiesterase, optimum pH 5.0; gene symbol, *Smpd1*) and ceramide system in major depression. Acid sphingomyelinase is a glycoprotein that functions as a lysosomal hydrolase, catalyzing the degradation of sphingomyelin to phosphorylcholine and ceramide at acidic pH [55]. Acid sphingomyelinase is present in lysosomes but also on small acidic domains of the outer leaflet of the plasma membrane [56]. The latter form was shown to have important signaling functions [56]. In addition, acid sphingomyelinase is also present in mitochondria, but the function of the mitochondrial form is unknown [57]. Depending on its glycosylation, acid sphingomyelinase is also secreted into the extracellular space [58].

Ceramide is formed by hydrolysis of sphingomyelin by the activity of acid, neutral, and alkaline sphingomyelinases depending on the pH optimum of the enzyme activity [59], by de novo synthesis [60], by degradation of complex (glyco)sphingolipids [61] and even from sphingosine by a reverse ceramidase activity [62]. Ceramide generated by acid sphingomyelinase has been shown to play a pivotal role in the mediation of stress and apoptotic stimuli including CD95, CD40, DR5/TRAIL, Fc γ RII, CD5, LFA-1, CD28, TNF α , Interleukin-1 receptor,

PAF-receptor, infection with *P. aeruginosa*, *S. aureus*, *N. gonorrhoeae*, Sindbis-Virus, measles virus, Rhinovirus, γ -irradiation, UV-light, Cu^{2+} , cisplatin or gemcitabine [56, 63-84].

Mechanistically, surface acid sphingomyelinase releases ceramide in the outer leaflet of the cell membrane [56]. Ceramide molecules spontaneously form small ceramide-enriched membrane domains that fuse to large ceramide-enriched membrane platforms. These platforms serve to trap and cluster receptor molecules and to initiate stress signals by mechanisms that still require definition. However, ceramide also directly regulates molecules, in particular ceramide released within multilamellar bodies or lysosomes binds to and activates cathepsin D, which translocates into the cytoplasm and causes cell death by activating Bid [85]. Other proteins that bind to ceramide include: kinase suppressor of Ras (KSR), which mediates cell death via Bad [86]; LCIIIB, which mediates autophagy [87]; PLA₂; which mediates the release of arachidonic acid; several protein kinase C (PKC) isoforms [88, 89]; and additional metabolites that may be important not only for apoptosis but also for the generation of inflammatory responses [88]. Finally, ceramide was shown to regulate important cellular ion channels, in particular calcium-release activated calcium channels (CRAC) and the potassium channel Kv1.3 [90, 91].

Although the regulation of acid sphingomyelinase is not well characterized, the enzyme seems to be regulated by redox mechanisms [92], in particular via a redox sensitive cysteine at position 629 (Cys629) [93]. As mentioned above some growth factors, for instance VEGF, inhibit acid sphingomyelinase [94].

The first link between the acid sphingomyelinase and major depression came from the observation that many tricyclic and tetracyclic antidepressant drugs, such as desipramine, imipramine or amitriptyline, functionally inhibit the activity of acid sphingomyelinase [95, 96]. Tri- and tetracyclic antidepressants interfere with the binding of the enzyme to the lysosomal and possibly also outer plasma membrane surface, displace the enzyme from the surface and induce a proteolytic degradation of the enzyme within lysosomes or the release of the enzyme from the surface and thereby mediate a functional inhibition of the acid sphingomyelinase [97-102]. While it was assumed that this effect of antidepressants is a side effect, we have shown that therapeutic concentrations of the antidepressants amitriptyline and fluoxetine also reduce acid sphingomyelinase activity and ceramide concentrations in the hippocampus. This action mediates the therapeutic effects of antidepressants, in particular increased neuronal proliferation, maturation, and survival and improved behavior in models of stress-induced depression [12]. These studies employed genetically-modified animals that either lacked or overexpressed the acid sphingomyelinase to prove that the effects of antidepressants are in fact mediated by targeting the acid sphingomyelinase [12]. Moreover, micro-injection of C16-ceramide (a natural ceramide) into the hippocampus PDMP-induced increased abundance of ceramide, or accumulation of ceramide within the hippocampus by genetic heterozygosity of the acid ceramidase or transgenic overexpression of the acid sphingomyelinase decreased neuronal proliferation, maturation, and survival, and resulted in a depression-like behavior in mice even in the absence of stress [12]. This indicates that increased levels of ceramide are able to trigger symptoms of major depression even without stress, and that antidepressants act, at least partially, via a reduction of ceramide levels in the hippocampus. Chronic unpredictable stress resulted in increased hippocampal ceramide abundance [12]. It is presently unknown whether patients with major depression exhibit increased ceramide levels in the hippocampus. In blood samples, increased acid sphingomyelinase activity and ceramide concentrations have been found in major depressive disorder, depressive syndromes and in posttraumatic stress disorder [94, 103-106]. Endogenous changes in ceramide metabolism may result in at least some forms of major depression. At present, neither the molecular mechanisms of the regulation nor the targets of ceramide in major depression are known.

In summary, studies in recent years provide evidence that neurogenesis, neuronal maturation, and the function of immature neurons in the hippocampus are novel cellular pathophysiological systems that may be altered in major depression, and that these are in-

interesting novel targets for treatment. Mechanistically, the acid sphingomyelinase/ceramide system does not only serve as target for antidepressants but an accumulation of ceramide also directly induces major depression. It will be very exciting to explore how ceramide mediates major depression, whether this is a specific effect restricted to experimental systems or whether such a role of ceramide applies to many forms of major depression, and whether certain levels of ceramide in the hippocampus are involved on the range of major depression from mild to severe cases.

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Disclosure Statement

The authors declare to have no conflict of interest.

References

- 1 Belmaker RH, Agam G: Major depressive disorder. *N Engl J Med* 2008;358:55-68.
- 2 Cizza G, Ravn P, Chrousos GP, Gold PW: Depression: A major, unrecognized risk factor for osteoporosis? *Trends Endocrinol Metab* 2001;12:198-203.
- 3 Forlenza MJ, Miller GE: Increased serum levels of 8-hydroxy-2'-deoxyguanosine in clinical depression. *Psychosom Med* 2006;68:1-7.
- 4 Hibbeln JR, Palmer JW, Davis JM: Are disturbances in lipid-protein interactions by phospholipase-a2 a predisposing factor in affective illness? *Biol Psychiatry* 1989;25:945-961.
- 5 Howren MB, Lamkin DM, Suls J: Associations of depression with c-reactive protein, il-1, and il-6: A meta-analysis. *Psychosom Med* 2009;71:171-186.
- 6 Maes M, Smith R, Christophe A, Vandoolaeghe E, Van Gastel A, Neels H, Demedts P, Wauters A, Meltzer HY: Lower serum high-density lipoprotein cholesterol (hdl-c) in major depression and in depressed men with serious suicidal attempts: Relationship with immune-inflammatory markers. *Acta Psychiatr Scand* 1997;95:212-221.
- 7 Musselman DL, Evans DL, Nemeroff CB: The relationship of depression to cardiovascular disease: Epidemiology, biology, and treatment. *Arch Gen Psychiatry* 1998;55:580-592.
- 8 Pandey GN, Dwivedi Y, Rizavi HS, Ren X, Conley RR: Decreased catalytic activity and expression of protein kinase c isozymes in teenage suicide victims: A postmortem brain study. *Arch Gen Psychiatry* 2004;61:685-693.
- 9 Parker G, Gibson NA, Brotchie H, Heruc G, Rees AM, Hadzi-Pavlovic D: Omega-3 fatty acids and mood disorders. *Am J Psychiatry* 2006;163:969-978.
- 10 David DJ, Samuels BA, Rainer Q, Wang JW, Marsteller D, Mendez I, Drew M, Craig DA, Guillard BP, Guilloux JP, Artymyshyn RP, Gardier AM, Gerald C, Antonijevic IA, Leonardo ED, Hen R: Neurogenesis-dependent and -independent effects of fluoxetine in an animal model of anxiety/depression. *Neuron* 2009;62:479-493.
- 11 D'Sa C, Duman RS: Antidepressants and neuroplasticity. *Bipolar Disord* 2002;4:183-194.
- 12 Gulbins E, Palmada M, Reichel M, Lüth A, Böhmer C, Amato D, Müller CP, Tischbirek CH, Groemer TW, Tabatabai G, Becker KA, Tripal P, Staedtler S, Ackermann TF, van Brederode J, Alzheimer C, Weller M, Lang UE, Kleuser B, Grassmé H, Kornhuber J: Acid sphingomyelinase-ceramide system mediates effects of antidepressant drugs. *Nat Med* 2013;19:934-938.
- 13 Kheirbek MA, Klemenhagen KC, Sahay A, Hen R: Neurogenesis and generalization: A new approach to stratify and treat anxiety disorders. *Nat Neurosci* 2012;15:1613-1620.

- 14 Lee HJ, Kim JW, Yim SV, Kim MJ, Kim SA, Kim YJ, Kim CJ, Chung JH: Fluoxetine enhances cell proliferation and prevents apoptosis in dentate gyrus of maternally separated rats. *Mol Psychiatry* 2001;6:610, 725-618.
- 15 Malberg JE, Eisch AJ, Nestler EJ, Duman RS: Chronic antidepressant treatment increases neurogenesis in adult rat hippocampus. *J Neurosci* 2000;20:9104-9110.
- 16 Manev H, Uz T, Smalheiser NR, Manev R: Antidepressants alter cell proliferation in the adult brain in vivo and in neural cultures in vitro. *Eur J Pharmacol* 2001;411:67-70.
- 17 Manji HK, Drevets WC, Charney DS: The cellular neurobiology of depression. *Nat Med* 2001;7:541-547.
- 18 Sahay A, Hen R: Adult hippocampal neurogenesis in depression. *Nat Neurosci* 2007;10:1110-1115.
- 19 Santarelli L, Saxe M, Gross C, Surget A, Battaglia F, Dulawa S, Weisstaub N, Lee J, Duman R, Arancio O, Belzung C, Hen R: Requirement of hippocampal neurogenesis for the behavioral effects of antidepressants. *Science* 2003;301:805-809.
- 20 Spalding KL, Bergmann O, Alkass K, Bernard S, Salehpour M, Huttner HB, Boström E, Westerlund I, Vial C, Buchholz BA, Possnert G, Mash DC, Druid H, Frisén J: Dynamics of hippocampal neurogenesis in adult humans. *Cell* 2013;153:1219-1227.
- 21 Hirschfeld RM: History and evolution of the monoamine hypothesis of depression. *J Clin Psychiatry* 2000;61 Suppl 6:4-6.
- 22 Brink CB, Harvey BH, Brand L: Tianeptine: A novel atypical antidepressant that may provide new insights into the biomolecular basis of depression. *Recent Pat CNS Drug Discov* 2006;1:29-41.
- 23 Czéh B, Michaelis T, Watanabe T, Frahm J, de Biurrun G, van Kampen M, Bartolomucci A, Fuchs E: Stress-induced changes in cerebral metabolites, hippocampal volume, and cell proliferation are prevented by antidepressant treatment with tianeptine. *Proc Natl Acad Sci U S A* 2001;98:12796-12801.
- 24 Müller N: Cox-2 inhibitors as antidepressants and antipsychotics: Clinical evidence. *Curr Opin Investig Drugs* 2010;11:31-42.
- 25 Videbech P, Ravnkilde B: Hippocampal volume and depression: A meta-analysis of mri studies. *Am J Psychiatry* 2004;161:1957-1966.
- 26 Ekstrand J, Hellsten J, Tingström A: Environmental enrichment, exercise and corticosterone affect endothelial cell proliferation in adult rat hippocampus and prefrontal cortex. *Neurosci Lett* 2008;442:203-207.
- 27 ALTMAN J: Are new neurons formed in the brains of adult mammals? *Science* 1962;135:1127-1128.
- 28 Altman J, Das GD: Post-natal origin of microneurons in the rat brain. *Nature* 1965;207:953-956.
- 29 Alvarez-Buylla A, Lim DA: For the long run: Maintaining germinal niches in the adult brain. *Neuron* 2004;41:683-686.
- 30 Braun SM, Jessberger S: Adult neurogenesis and its role in neuropsychiatric disease, brain repair and normal brain function. *Neuropathol Appl Neurobiol* 2014;40:3-12.
- 31 Doetsch F, Caillé I, Lim DA, García-Verdugo JM, Alvarez-Buylla A: Subventricular zone astrocytes are neural stem cells in the adult mammalian brain. *Cell* 1999;97:703-716.
- 32 Kuhn HG, Dickinson-Anson H, Gage FH: Neurogenesis in the dentate gyrus of the adult rat: Age-related decrease of neuronal progenitor proliferation. *J Neurosci* 1996;16:2027-2033.
- 33 Reynolds BA, Weiss S: Generation of neurons and astrocytes from isolated cells of the adult mammalian central nervous system. *Science* 1992;255:1707-1710.
- 34 Ernst A, Alkass K, Bernard S, Salehpour M, Perl S, Tisdale J, Possnert G, Druid H, Frisén J: Neurogenesis in the striatum of the adult human brain. *Cell* 2014;156:1072-1083.
- 35 Bergmann O, Liebl J, Bernard S, Alkass K, Yeung MS, Steier P, Kutschera W, Johnson L, Landén M, Druid H, Spalding KL, Frisén J: The age of olfactory bulb neurons in humans. *Neuron* 2012;74:634-639.
- 36 Ming GL, Song H: Adult neurogenesis in the mammalian brain: Significant answers and significant questions. *Neuron* 2011;70:687-702.
- 37 Sanai N, Nguyen T, Ihrie RA, Mirzadeh Z, Tsai HH, Wong M, Gupta N, Berger MS, Huang E, Garcia-Verdugo JM, Rowitch DH, Alvarez-Buylla A: Corridors of migrating neurons in the human brain and their decline during infancy. *Nature* 2011;478:382-386.
- 38 Zhao C, Deng W, Gage FH: Mechanisms and functional implications of adult neurogenesis. *Cell* 2008;132:645-660.

- 39 Bruel-Jungerman E, Veyrac A, Dufour F, Horwood J, Laroche S, Davis S: Inhibition of pi3k-akt signaling blocks exercise-mediated enhancement of adult neurogenesis and synaptic plasticity in the dentate gyrus. *PLoS One* 2009;4:e7901.
- 40 Huang W, Zhao Y, Zhu X, Cai Z, Wang S, Yao S, Qi Z, Xie P: Fluoxetine upregulates phosphorylated-akt and phosphorylated-erk1/2 proteins in neural stem cells: Evidence for a crosstalk between akt and erk1/2 pathways. *J Mol Neurosci* 2013;49:244-249.
- 41 Kim JY, Duan X, Liu CY, Jang MH, Guo JU, Pow-anpongkul N, Kang E, Song H, Ming GL: Disc1 regulates new neuron development in the adult brain via modulation of akt-mtor signaling through kiaa1212. *Neuron* 2009;63:761-773.
- 42 Le Belle JE, Orozco NM, Paucar AA, Saxe JP, Mottahedeh J, Pyle AD, Wu H, Kornblum HI: Proliferative neural stem cells have high endogenous ros levels that regulate self-renewal and neurogenesis in a pi3k/akt-dependant manner. *Cell Stem Cell* 2011;8:59-71.
- 43 Kolla N, Wei Z, Richardson JS, Li XM: Amitriptyline and fluoxetine protect pc12 cells from cell death induced by hydrogen peroxide. *J Psychiatry Neurosci* 2005;30:196-201.
- 44 Magnusson JP, Göritz C, Tatarishvili J, Dias DO, Smith EM, Lindvall O, Kokaia Z, Frisén J: A latent neurogenic program in astrocytes regulated by notch signaling in the mouse. *Science* 2014;346:237-241.
- 45 Okuyama N, Takagi N, Kawai T, Miyake-Takagi K, Takeo S: Phosphorylation of extracellular-regulating kinase in nmda receptor antagonist-induced newly generated neurons in the adult rat dentate gyrus. *J Neurochem* 2004;88:717-725.
- 46 Peltier J, O'Neill A, Schaffer DV: Pi3k/akt and creb regulate adult neural hippocampal progenitor proliferation and differentiation. *Dev Neurobiol* 2007;67:1348-1361.
- 47 Yoshinaga Y, Kagawa T, Shimizu T, Inoue T, Takada S, Kuratsu J, Taga T: Wnt3a promotes hippocampal neurogenesis by shortening cell cycle duration of neural progenitor cells. *Cell Mol Neurobiol* 2010;30:1049-1058.
- 48 Madsen TM, Treschow A, Bengzon J, Bolwig TG, Lindvall O, Tingström A: Increased neurogenesis in a model of electroconvulsive therapy. *Biol Psychiatry* 2000;47:1043-1049.
- 49 Ikrar T, Guo N, He K, Besnard A, Levinson S, Hill A, Lee HK, Hen R, Xu X, Sahay A: Adult neurogenesis modifies excitability of the dentate gyrus. *Front Neural Circuits* 2013;7:204.
- 50 de Kloet ER, Derijk RH, Meijer OC: Therapy insight: Is there an imbalanced response of mineralocorticoid and glucocorticoid receptors in depression? *Nat Clin Pract Endocrinol Metab* 2007;3:168-179.
- 51 Snyder JS, Soumier A, Brewer M, Pickel J, Cameron HA: Adult hippocampal neurogenesis buffers stress responses and depressive behaviour. *Nature* 2011;476:458-461.
- 52 Surget A, Tanti A, Leonardo ED, Laugeray A, Rainer Q, Touma C, Palme R, Griebel G, Ibarguen-Vargas Y, Hen R, Belzung C: Antidepressants recruit new neurons to improve stress response regulation. *Mol Psychiatry* 2011;16:1177-1188.
- 53 Bergmann O, Frisén J: Neuroscience. Why adults need new brain cells. *Science* 2013;340:695-696.
- 54 Rotheneichner P, Lange S, O'Sullivan A, Marschallinger J, Zaunmair P, Geretsegger C, Aigner L, Couillard-Despres S: Hippocampal neurogenesis and antidepressive therapy: Shocking relations. *Neural Plast* 2014;2014:723915.
- 55 Schneider PB, Kennedy EP: Sphingomyelinase in normal human spleens and in spleens from subjects with niemann-pick disease. *J Lipid Res* 1967;8:202-209.
- 56 Grassme H, Jekle A, Riehle A, Schwarz H, Berger J, Sandhoff K, Kolesnick R, Gulbins E: Cd95 signaling via ceramide-rich membrane rafts. *J Biol Chem* 2001;276:20589-20596.
- 57 Matsumoto A, Comatas KE, Liu L, Stamler JS: Screening for nitric oxide-dependent protein-protein interactions. *Science* 2003;301:657-661.
- 58 Schissel SL, Schuchman EH, Williams KJ, Tabas I: Zn²⁺-stimulated sphingomyelinase is secreted by many cell types and is a product of the acid sphingomyelinase gene. *J Biol Chem* 1996;271:18431-18436.
- 59 Goñi FM, Alonso A: Sphingomyelinases: Enzymology and membrane activity. *FEBS Lett* 2002;531:38-46.
- 60 Lahiri S, Futerman AH: The metabolism and function of sphingolipids and glycosphingolipids. *Cell Mol Life Sci* 2007;64:2270-2284.

- 61 Ishibashi Y, Nakasone T, Kiyohara M, Horibata Y, Sakaguchi K, Hijikata A, Ichinose S, Omori A, Yasui Y, Imamura A, Ishida H, Kiso M, Okino N, Ito M: A novel endoglycoceramidase hydrolyzes oligogalactosylceramides to produce galactooligosaccharides and ceramides. *J Biol Chem* 2007;282:11386-11396.
- 62 Okino N, He X, Gatt S, Sandhoff K, Ito M, Schuchman EH: The reverse activity of human acid ceramidase. *J Biol Chem* 2003;278:29948-29953.
- 63 Abdel Shakor AB, Kwiatkowska K, Sobota A: Cell surface ceramide generation precedes and controls fcgammarii clustering and phosphorylation in rafts. *J Biol Chem* 2004;279:36778-36787.
- 64 Boucher LM, Wiegmann K, Fütterer A, Pfeffer K, Machleidt T, Schütze S, Mak TW, Krönke M: Cd28 signals through acidic sphingomyelinase. *J Exp Med* 1995;181:2059-2068.
- 65 Dumitru CA, Gulbins E: Trail activates acid sphingomyelinase via a redox mechanism and releases ceramide to trigger apoptosis. *Oncogene* 2006;25:5612-5625.
- 66 Esen M, Schreiner B, Jendrossek V, Lang F, Fassbender K, Grassmé H, Gulbins E: Mechanisms of staphylococcus aureus induced apoptosis of human endothelial cells. *Apoptosis* 2001;6:431-439.
- 67 Gassert E, Avota E, Harms H, Krohne G, Gulbins E, Schneider-Schaulies S: Induction of membrane ceramides: A novel strategy to interfere with t lymphocyte cytoskeletal reorganisation in viral immunosuppression. *PLoS Pathog* 2009;5:e1000623.
- 68 Göggel R, Winoto-Morbach S, Vielhaber G, Imai Y, Lindner K, Brade L, Brade H, Ehlers S, Slutsky AS, Schütze S, Gulbins E, Uhlig S: Paf-mediated pulmonary edema: A new role for acid sphingomyelinase and ceramide. *Nat Med* 2004;10:155-160.
- 69 Grammatikos G, Teichgräber V, Carpinteiro A, Trarbach T, Weller M, Hengge UR, Gulbins E: Overexpression of acid sphingomyelinase sensitizes glioma cells to chemotherapy. *Antioxid Redox Signal* 2007;9:1449-1456.
- 70 Grassmé H, Gulbins E, Brenner B, Ferlinz K, Sandhoff K, Harzer K, Lang F, Meyer TF: Acidic sphingomyelinase mediates entry of *N. gonorrhoeae* into nonphagocytic cells. *Cell* 1997;91:605-615.
- 71 Grassmé H, Jendrossek V, Bock J, Riehle A, Gulbins E: Ceramide-rich membrane rafts mediate cd40 clustering. *J Immunol* 2002;168:298-307.
- 72 Grassmé H, Jendrossek V, Riehle A, von Kürthy G, Berger J, Schwarz H, Weller M, Kolesnick R, Gulbins E: Host defense against *pseudomonas aeruginosa* requires ceramide-rich membrane rafts. *Nat Med* 2003;9:322-330.
- 73 Grassmé H, Riehle A, Wilker B, Gulbins E: Rhinoviruses infect human epithelial cells via ceramide-enriched membrane platforms. *J Biol Chem* 2005;280:26256-26262.
- 74 Hauck CR, Grassmé H, Bock J, Jendrossek V, Ferlinz K, Meyer TF, Gulbins E: Acid sphingomyelinase is involved in ceacam receptor-mediated phagocytosis of *neisseria gonorrhoeae*. *FEBS Lett* 2000;478:260-266.
- 75 Jan JT, Chatterjee S, Griffin DE: Sindbis virus entry into cells triggers apoptosis by activating sphingomyelinase, leading to the release of ceramide. *J Virol* 2000;74:6425-6432.
- 76 Lacour S, Hammann A, Grazide S, Lagadic-Gossmann D, Athias A, Sergeant O, Laurent G, Gambert P, Solary E, Dimanche-Boitrel MT: Cisplatin-induced cd95 redistribution into membrane lipid rafts of ht29 human colon cancer cells. *Cancer Res* 2004;64:3593-3598.
- 77 Lang PA, Schenck M, Nicolay JP, Becker JU, Kempe DS, Lupescu A, Koka S, Eisele K, Klarl BA, Rübber H, Schmid KW, Mann K, Hildenbrand S, Hefter H, Huber SM, Wieder T, Erhardt A, Häussinger D, Gulbins E, Lang F: Liver cell death and anemia in wilson disease involve acid sphingomyelinase and ceramide. *Nat Med* 2007;13:164-170.
- 78 Mathias S, Younes A, Kan CC, Orlow I, Joseph C, Kolesnick RN: Activation of the sphingomyelin signaling pathway in intact e4 cells and in a cell-free system by il-1 beta. *Science* 1993;259:519-522.
- 79 Ni HT, Deeths MJ, Li W, Mueller DL, Mescher MF: Signaling pathways activated by leukocyte function-associated ag-1-dependent costimulation. *J Immunol* 1999;162:5183-5189.
- 80 Rotolo JA, Zhang J, Donepudi M, Lee H, Fuks Z, Kolesnick R: Caspase-dependent and -independent activation of acid sphingomyelinase signaling. *J Biol Chem* 2005;280:26425-26434.
- 81 Santana P, Peña LA, Haimovitz-Friedman A, Martin S, Green D, McLoughlin M, Cordon-Cardo C, Schuchman EH, Fuks Z, Kolesnick R: Acid sphingomyelinase-deficient human lymphoblasts and mice are defective in radiation-induced apoptosis. *Cell* 1996;86:189-199.

- 82 Schütze S, Potthoff K, Machleidt T, Berkovic D, Wiegmann K, Krönke M: Tnf activates nf-kappa b by phosphatidylcholine-specific phospholipase c-induced "Acidic" Sphingomyelin breakdown. *Cell* 1992;71:765-776.
- 83 Schwandner R, Wiegmann K, Bernardo K, Kreder D, Kronke M: Tnf receptor death domain-associated proteins tradd and fadd signal activation of acid sphingomyelinase. *J Biol Chem* 1998;273:5916-5922.
- 84 Simarro M, Calvo J, Vilà JM, Places L, Padilla O, Alberola-Ila J, Vives J, Lozano F: Signaling through cd5 involves acidic sphingomyelinase, protein kinase c-zeta, mitogen-activated protein kinase kinase, and c-jun nh2-terminal kinase. *J Immunol* 1999;162:5149-5155.
- 85 Heinrich M, Neumeyer J, Jakob M, Hallas C, Tchikov V, Winoto-Morbach S, Wickel M, Schneider-Brachert W, Trauzold A, Hethke A, Schütze S: Cathepsin d links tnf-induced acid sphingomyelinase to bid-mediated caspase-9 and -3 activation. *Cell Death Differ* 2004;11:550-563.
- 86 Basu S, Bayoumy S, Zhang Y, Lozano J, Kolesnick R: Bad enables ceramide to signal apoptosis via ras and raf-1. *J Biol Chem* 1998;273:30419-30426.
- 87 Sentelle RD, Senkal CE, Jiang W, Ponnusamy S, Gencer S, Selvam SP, Ramshesh VK, Peterson YK, Lemasters JJ, Szulc ZM, Bielawski J, Ogretmen B: Ceramide targets autophagosomes to mitochondria and induces lethal mitophagy. *Nat Chem Biol* 2012;8:831-838.
- 88 Huwiler A, Johansen B, Skarstad A, Pfeilschifter J: Ceramide binds to the calb domain of cytosolic phospholipase a2 and facilitates its membrane docking and arachidonic acid release. *FASEB J* 2001;15:7-9.
- 89 Müller G, Ayoub M, Storz P, Rennecke J, Fabbro D, Pfizenmaier K: Pkc zeta is a molecular switch in signal transduction of tnf-alpha, bifunctionally regulated by ceramide and arachidonic acid. *EMBO J* 1995;14:1961-1969.
- 90 Gulbins E, Szabo I, Baltzer K, Lang F: Ceramide-induced inhibition of t lymphocyte voltage-gated potassium channel is mediated by tyrosine kinases. *Proc Natl Acad Sci U S A* 1997;94:7661-7666.
- 91 Lepple-Wienhues A, Belka C, Laun T, Jekle A, Walter B, Wieland U, Welz M, Heil L, Kun J, Busch G, Weller M, Bamberg M, Gulbins E, Lang F: Stimulation of cd95 (fas) blocks t lymphocyte calcium channels through sphingomyelinase and sphingolipids. *Proc Natl Acad Sci U S A* 1999;96:13795-13800.
- 92 Zhang Y, Li X, Carpinteiro A, Gulbins E: Acid sphingomyelinase amplifies redox signaling in pseudomonas aeruginosa-induced macrophage apoptosis. *J Immunol* 2008;181:4247-4254.
- 93 Qiu H, Edmunds T, Baker-Malcolm J, Karey KP, Estes S, Schwarz C, Hughes H, Van Patten SM: Activation of human acid sphingomyelinase through modification or deletion of c-terminal cysteine. *J Biol Chem* 2003;278:32744-32752.
- 94 Hammad SM, Truman JP, Al Gadban MM, Smith KJ, Twal WO, Hamner MB: Altered blood sphingolipidomics and elevated plasma inflammatory cytokines in combat veterans with post-traumatic stress disorder. *Neurobiol Lipids* 2012;10:2.
- 95 Albouze S, Hauw JJ, Berwald-Netter Y, Boutry JM, Bourdon R, Baumann N: Tricyclic antidepressants induce sphingomyelinase deficiency in fibroblast and neuroblastoma cell cultures. *Biomedicine* 1981;35:218-220.
- 96 Teichgräber V, Ulrich M, Endlich N, Riethmüller J, Wilker B, De Oliveira-Munding CC, van Heeckeren AM, Barr ML, von Kürthy G, Schmid KW, Weller M, Tümmler B, Lang F, Grassme H, Döring G, Gulbins E: Ceramide accumulation mediates inflammation, cell death and infection susceptibility in cystic fibrosis. *Nat Med* 2008;14:382-391.
- 97 Hurwitz R, Ferlinz K, Vielhaber G, Moczall H, Sandhoff K: Processing of human acid sphingomyelinase in normal and i-cell fibroblasts. *J Biol Chem* 1994;269:5440-5445.
- 98 Hurwitz R, Ferlinz K, Sandhoff K: The tricyclic antidepressant desipramine causes proteolytic degradation of lysosomal sphingomyelinase in human fibroblasts. *Biol Chem Hoppe Seyler* 1994;375:447-450.
- 99 Kölzer M, Werth N, Sandhoff K: Interactions of acid sphingomyelinase and lipid bilayers in the presence of the tricyclic antidepressant desipramine. *FEBS Lett* 2004;559:96-98.
- 100 Kornhuber J, Tripal P, Reichel M, Terflath L, Bleich S, Wiltfang J, Gulbins E: Identification of new functional inhibitors of acid sphingomyelinase using a structure-property-activity relation model. *J Med Chem* 2008;51:219-237.
- 101 Kornhuber J, Tripal P, Reichel M, Mühle C, Rhein C, Muehlbacher M, Groemer TW, Gulbins E: Functional inhibitors of acid sphingomyelinase (fiasmas): A novel pharmacological group of drugs with broad clinical applications. *Cell Physiol Biochem* 2010;26:9-20.

- 102 Kornhuber J, Muehlbacher M, Trapp S, Pechmann S, Friedl A, Reichel M, Mühle C, Terfloth L, Groemer TW, Spitzer GM, Liedl KR, Gulbins E, Tripal P: Identification of novel functional inhibitors of acid sphingomyelinase. *PLoS One* 2011;6:e23852.
- 103 Kornhuber J, Medlin A, Bleich S, Jendrossek V, Henkel AW, Wiltfang J, Gulbins E: High activity of acid sphingomyelinase in major depression. *J Neural Transm (Vienna)* 2005;112:1583-1590.
- 104 Mielke MM, Maetzler W, Haughey NJ, Bandaru VV, Savica R, Deuschle C, Gasser T, Hauser AK, Gräber-Sultan S, Schleicher E, Berg D, Liepelt-Scarfone I: Plasma ceramide and glucosylceramide metabolism is altered in sporadic parkinson's disease and associated with cognitive impairment: A pilot study. *PLoS One* 2013;8:e73094.
- 105 Gracia-Garcia P, Rao V, Haughey NJ, Banduru VV, Smith G, Rosenberg PB, Lobo A, Lyketsos CG, Mielke MM: Elevated plasma ceramides in depression. *J Neuropsychiatry Clin Neurosci* 2011;23:215-218.
- 106 Demirkan A, Isaacs A, Ugozai P, Liebisch G, Struchalin M, Rudan I, Wilson JF, Pramstaller PP, Gyllenstein U, Campbell H, Schmitz G, Oostra BA, van Duijn CM: Plasma phosphatidylcholine and sphingomyelin concentrations are associated with depression and anxiety symptoms in a dutch family-based lipidomics study. *J Psychiatr Res* 2013;47:357-362.