Department of Physiology
Faculty of Medicine
University of Helsinki
Finland

Doctoral program Brain & Mind

SLEEP AND DEPRESSION: DEVELOPMENTAL ASPECTS AND MOLECULAR MECHANISMS

Olena Santangeli

ACADEMIC DISSERTATION

To be publicly discussed with the permission of the Faculty of Medicine,
University of Helsinki,
in Biomedicum Helsinki 1, Lecture Hall 3, Haartmaninkatu 8,
on April 20th 2018, at 12 noon

Supervisors: Adjunct Professor (Docent) Tarja Stenberg (Porkka-Heiskanen), M.D., Ph.D.
Department of Physiology, Faculty of Medicine, University of Helsinki
and
Professor Tiina Paunio, M.D., Ph.D.
Genomics and Biomarkers Unit, National Institute for Health and Welfare, Helsinki, Finland, Department of Psychiatry, University of Helsinki and Helsinki University Hospital, Helsinki

Thesis committee: Professor Erkki Isometsä, M.D., Ph.D.
Department of Psychiatry, University of Helsinki and Helsinki University Hospital, Helsinki
and
Professor Pertti Panula, M.D., Ph.D.
Department of Anatomy, Faculty of Medicine, Neuroscience Center, University of Helsinki

Reviewed by: Docent Tomi Rantamäki, Ph.D.
Neuroscience Center, University of Helsinki, Helsinki
Division of Physiology and Neuroscience, Department of Biosciences, Faculty of Biological and Environmental Sciences, University of Helsinki
and
Docent Soili M Lehto, M.D., Ph.D.
Institute of Clinical Medicine/Psychiatry, University of Eastern Finland, Kuopio
Department of Psychiatry, Kuopio University Hospital
Department of Psychology and Logopedics, Faculty of Medicine, University of Helsinki

Opponent: Associate Professor Janne Grønli, Ph.D.
Bergen Stress and Sleep Group, Department of Biological and Medical Psychology, Faculty of Psychology, University of Bergen, Bergen, Norway

Custos: Professor Antti Pertovaara, M.D., Ph.D.
Department of Physiology, Faculty of Medicine, University of Helsinki

Dissertationes Scholae Doctoralis
Ad Sanitatem Investigandam Universitatis Helsinkiensis
15/2018
ISSN 2342-3161 (print)
ISSN 2342-317X (online)
ISBN 978-951-51-4072-2 (paperback)
ISBN 978-951-51-4073-9 (PDF)
http://ethesis.helsinki.fi/
Helsinki 2018

Contents

Abstract

Early life stress, sleep disturbances and alterations in neuronal plasticity have been associated with depression, yet the relation between these factors and depression remain poorly understood. This project explored the interconnection between depression and sleep disturbances, starting from early stage of development. Possible molecular mechanisms (such as brain derived neurotrophic factor (BDNF) and adenosine) underlying this interaction were assessed using animal models. The investigation of sleep disturbances in early-onset depression in humans was performed in depressed adolescents. The association between *BDNF* gene polymorphism (Val66Met) and the development of sleep was studied using a birth cohort.

First, we investigated the epigenetic mechanisms of *BDNF* gene regulation during spontaneous sleep and after sleep deprivation using methylated DNA immunoprecipitation in rats. We showed that the dynamics of *BDNF* transcription during spontaneous sleep-wake cycle in brain areas crucial for sleep regulation (basal forebrain (BF) and frontal cortex) is partially regulated by brain area specific DNA methylation, and that the degree of sleep deprivation-induced *BDNF* up-regulation is transcript- and brain area-dependent.

To assess the effect of early life stress on sleep and possible mechanisms of this effect, we used cross-fostering in rats. Long-term polysomnographic recordings in undisturbed and sleep deprived rats were performed to assess sleep profile. *In vivo* microdialysis during spontaneous sleep with following high-performance liquid chromatography was performed to assess adenosine level in the BF. *BDNF* gene expression in the involved brain areas was determined by quantitative PCR. Behaviour was assessed using open field and sucrose preference tests. The results indicated that early life stress induced by cross-fostering procedure produce negligible phenotypic alterations during adolescence or in adult rats. Cross-fostered rats demonstrated, however, clear changes in sleep architecture. Rapid eye movement (REM) sleep was affected the most: duration and number of its episodes was significantly increased in cross-fostered rats. Moreover, cross-fostering led to persistent molecular changes in the brain, which was evidenced by decreased adenosine level during spontaneous sleep-wake cycle and by a tendency towards lower *BDNF* gene expression in the BF.

Further, to investigate sleep disturbances in early-onset depression we studied macro- and microarchitecture of sleep in depressed adolescent boys using polysomnography. Slow-wave activity (SWA) dissipation and rise rate were assessed and compared to age-matched controls. The sleep of depressed boys was characterized by lower SWA and its slower rise during the course of first non-REM (NREM) sleep episode compared to healthy boys. The dissipation (decline from the first to the third NREM episode) of SWA through the night in depressed patients had flatter shape compared to healthy subjects. Moreover, a negative correlation between SWA dissipation and depression severity was detected. All the changes were more pronounced in the frontal derivation compared to central derivation that support the idea that sleep regulation and depression might share common neurobiological mechanisms.

To examine whether the *BDNF* Val66Met polymorphism can predispose individuals to depression and sleep disturbances Finnish CHILD-SLEEP birth cohort (n=1678) was used. The association between this polymorphism and early life development in infants of 3 and 8 months of age was assessed. Genotyping was done with Illumina Infinium PsychArray BeadChip. The developmental parameters (e.g. sleep, psychomotor development, temperament) were assessed by using specific questionnaires. We found no robust association between *BDNF* Val66Met polymorphism and studied parameters during early postnatal life of infants.

In summary, early life stress leaves persistent molecular trace in the brain, and sleep appears to be one of the most prominent indicators of early stressful events. Early life onset depression is characterized by changes in sleep macro-and microarchitecture that are associated with depressive symptoms severity. Although BDNF is playing an important role in sleep and mood regulation, its Val66Met polymorphism is not associated with development of these parameters during early stages of life.

Abstract in Finnish

Uni ja depressio: kehityskulkuja ja mekanismeja

Masennukseen liitetään usein varhaisen kehityksen aikainen stressi, univaikeudet ja hermoston plastisuuden muutokset, mutta näiden tekijöiden ja depression suhde tunnetaan kuitenkin huonosti. Tässä työssä tutkittiin masennuksen ja unihäiriöiden välisiä yhteyksiä keskittyen varhaiseen kehitykseen. Molekyylitason mekanismeja (kuten brain derived neurotrophic factor, BDNF, ja adenosiini) tutkittiin eläinmallien avulla, unihäiriöiden esiintymistä varhaisessa depression vaiheessa tutkittiin teini-ikäisillä pojilla, ja unen kehityksen ja BDNF geenin polymorfismi Val66Met:in välistä yhteyttä syntymäkohortin avulla.

Tutkimme aluksi rotilla BDNF geenin epigeneettistä säätelyä normaalin unen ja unen puutteen aikana käyttäen DNA:n immunosaostusta. Osoitimme, että BDNF geenin luentaa eri unta säätelevillä aivoalueilla (etuaivojen pohjaosa, BF ja etuaivokuori) säätelee osittain aivoalueille tyypillinen DNA:n metylaatio, ja että unen puutteen aiheuttama BDNF geenin luennan lisäys riippuu sekä aivoalueesta että BDNF:n geenikopiosta.

Tutkiessamme varhaisen stressin vaikutusta myöhempään unen kehitykseen ja vaikutuksen mekanismeja, käytimme uutta stressimallia: poikasten vaihtoa emojen välillä (cross-fostering). Uniprofiilin selvittämiseksi suoritimme aivosähkökäyrän ja lihasjänteyden määrityksiä varhaisnuorilla ja aikuisilla rotilla. Suoritimme myös *in vivo* mikrodialyysin adenosiinimäärityksiä varten; BDNF geenin ilmentyminen määritettiin eri aivoalueilta. Käyttäytymistä, erityisesti masennuksen merkkejä etsien, tutkittiin avokenttätestillä sekä sokerijuomatestin avulla. Tuloksista ilmeni, että varhaisen kehityksen aikainen, stressi (poikasten vaihto) ei näytä vaikuttavan eläinten masennuksena tulkittavaan käyttäytymiseen varhaisnuoruudessa tai aikuisena. Kuitenkin se aiheutti merkittäviä muutoksia erityisesti REM uneen: REM unijaksojen lukumäärä ja pituus olivat lisääntyneet. Aivoissa havaittiin myös molekyylitason muutoksia: adenosiinitaso oli laskenut ja etuaivojen pohjaosan (basal forebrain) BDNF:n ilmenemisessä oli taipumus mataliin arvoihin.

Selvittääksemme unen häiriöitä varhain alkaneessa masennuksessa mittasimme polysomnografiaa käyttäen unen mikro- ja makrorakennetta

teini-ikäisillä pojilla. Tutkimme hidasaaltoaktiviteetin (SWA) vähenemistä ensimmäisestä viimeiseen NREM unijaksoon (SWA dissipaatio) sekä SWA:n nousua ensimmäisen NREM unijakson aikana ja vertasimme näitä terveiden poikien vastaaviin arvoihin. Totesimme, että masentuneilla pojilla SWA oli kokonaisuudessaan madaltunut ja kasvu ensimmäisen NREM jakson aikana hidastunut. SWA dissipaatio oli latistunut ja madaltuma korreloi masennuksen vakavuuden kanssa. Kaikki muutokset havaittiin voimakkaimpina etuaivolohkon mittauksissa verrattuna muiden aivoalueiden mittauksiin, vahvistaen käsitystä siitä, että unen häiriöillä ja masennuksella voi olla yhteisiä säätelymekanismeja.

Tutkimuksessa BDNF Val66Met polymorfismin merkityksestä altistumisessa masennukselle ja unen häiriöille käytimme suomalaista CHILD_SLEEP syntymäkohorttia (n=1678). Tämän polymorfismin korrelaatiota erilaisiin varhaisen kehityksen mittareihin tutkittiin 3 ja 8 kuukauden iässä. Genotyypitys suoritettiin Illumina Infinium PsychArray BeadChipillä. Kehitystä kuvaavia suureita (esim. uni, psykomotorinen kehitys, temperamentti) selvitettiin kyselylomakkeiden avulla. Työssä ei löytynyt vahvaa yhteyttä BDNF geenin polymorfismien ja kehityssuureiden välille.

Yhteenvetona voidaan todeta, että varhain koettu stressi jättää aivoihin pysyvän molekyylijäljen ja että uni/unen häiriöt näyttävät olevan herkin mittari varhaisen stressin vaikutuksien toteamisessa. Varhain alkavassa masennuksessa nähdään unen sekä mikro- että makrotasolla muutoksia, jotka korreloivat masennuksen vakavuuteen. Vaikka BDNF:llä on tärkeä rooli sekä unen että kehityksen säätelyssä, sen Val66Met polymorfismi ei näytä vaikuttavan unen häiriöiden tai masennuksen syntyyn kehityksen varhaisessa vaiheessa.

List of Original Publications and Manuscripts

I **Ventskovska, O.**, Porkka-Heiskanen, T., & Karpova, N. N. (2015). Spontaneous sleep-wake cycle and sleep deprivation differently induce Bdnf1, Bdnf4 and Bdnf9a DNA methylation and transcripts levels in the basal forebrain and frontal cortex in rats. Journal of sleep research, 24(2), 124-130. doi:10.1111/jsr.12242

II **Santangeli, O.**, Lehtikuja, H., Palomaki, E., Wigren, H. K., Paunio, T., & Porkka-Heiskanen, T. (2016). Sleep and Behavior in Cross-Fostering Rats: Developmental and Sex Aspects. Sleep, 39(12), 2211-2221. doi:10.5665/sleep.6328.

III **Santangeli O.**, Porkka-Heiskanen T., Virkkala J., Castaneda A., Marttunen M., Paunio T., Urrila AS,. (2017). Sleep and slow-wave activity in depressed adolescent boys: a preliminary study. Sleep Medicine, 38, 24-30. doi:10.1016/j.sleep.2017.06.029.

IV **Santangeli O.,** Kylliäinen A., Salo P., Kantojärvi K., Saarenpää-Heikkilä O., Paavonen J., Vierikko E., Satomaa AL., Himanen SL., Porkka-Heiskanen T., Paunio T. BDNF functional variation Val66Met is not associated with early life infants' sleep and neurodevelopment. Manuscript.

None of the articles have been used in other dissertations. The articles have been reprinted with the permissions of their copyright holders. The studies are referred in the text of dissertation as Study I, II, III, and IV, respectively. Unpublished material is also presented this thesis.

Author's contribution to the original publications and the manuscript:

I Planning and performing experiments, data analyses, writing the manuscript.

II Participated in the generation of the original idea. Planning and performing experiments, data analyses, writing the manuscript.

III Designing the study, data analyses, writing the manuscript.

IV Designing the study, data analyses, writing the manuscript.

Abbreviations

5-mC	5-methylcytosine
aCSF	artificial cerebrospinal fluid
ACTB	beta actin (gene)
ACTH	adrenocorticotropic hormone
ADP	adenosine diphosphate
AIS	Athens insomnia scale
AMP	adenosine monophosphate
ATP	adenosine triphosphate
BDI-21	Beck depression inventory
BDNF	brain-derived neurotrophic factor
BISQ	Brief infant sleep questionnaire
BL	baseline
BMI	body mass index
CF	cross-fostering
CTRL	control
DNAMT	DNA methyltransferase
EEG	electroencephalography
EOG	electrooculography
FFT	fast Fourier transform
HAT	histone acetyltransferase
HDAC	histone deacetylase
HDRS	Hamilton depression rating scale
HDMT	histone demethylase

HMT	histone methyltransferase
HPA	hypothalamic–pituitary–adrenal (axis)
HPLC	high-performance liquid chromatography
IBQ-R	revised infant behaviour questionnaire
IL	interleukin
IP	immunoprecipitation
LTD	long-term depression
LTP	long-term potentiation
MeDIP	methylated DNA immunoprecipitation
MDD	major depressive disorder
NREM	non-rapid eye movement (sleep)
OF	open field
PCR	polymerase chain reaction
PND	postnatal day
REM	rapid eye movement (sleep)
SEI	sleep efficiency index
SHY	synaptic homeostasis hypothesis
SPT	sucrose preference test
SSRI	selective serotonin reuptake inhibitor
SWS	slow-wave sleep
SWA	slow-wave activity
TNF	tumor necrosis factor
TST	total sleep time
WASO	wakefulness time after sleep onset
ZT	*Zeitgeber*

1. Introduction

Major depressive disorder (MDD) affects about 15% of the population at some point in their lives and by the year 2020, MDD is estimated to be the second leading cause of world disability (Bromet et al., 2011; Kessler, 2007). One of the key symptoms of depressive disorder is sleep disturbance (Nutt et al., 2008). Up to 90% of patients who suffer from an acute depressive episode report changes in their sleep. Moreover, people with persistent sleep problems have elevated risk of developing psychiatric disorders, including depression (Breslau et al., 1996). Among the whole range of psychiatric disorders, MDD has the strongest lifetime association with sleep disturbances (Breslau et al., 1996). Thus, depression and sleep disorders have complex and reciprocal relationships.

Early life stress is one of the important predisposing factors for depression (Arnow, 2004). The incidence of depressive disorder increases promptly from childhood to adolescence (Newman et al., 1996; Oldehinkel et al., 1999) and then frequently recurs or persists into adulthood (Newman et al., 1996; Pine et al., 1999; Sagatun et al., 2016). Adverse experiences during early stage of life, such as physical, or emotional abuse, neglect, or loss of a parent and poor child-parent relationships, are important susceptibility factors for development of MDD in adulthood, as evidenced by both epidemiological studies and experimental studies using animal models (Heim & Nemeroff, 2001; McCormick et al., 2008). Notably, sleep disturbances associated with early life negative experiences can persist for years after the stressful events (Chapman et al., 2011; Greenfield et al., 2011; Koskenvuo et al., 2010).

Emerging evidence links deficits or impairment in synaptic and structural plasticity to mood disorders (Castren et al., 2007; Duman, 2002; Dwivedi, 2009). The abnormal neural plasticity may be related to alterations in the levels of neurotrophic factors; especially brain-derived neurotrophic factor (BDNF), which plays a central role in neuronal and synaptic plasticity. Changes in the BDNF levels have been implicated both in the aetiology of depression and in antidepressant drug action (Castren & Rantamaki, 2010). Importantly, this molecule has also been shown to play an important role in sleep regulation and the level of *BDNF* expression is fluctuating depending on the vigilance sate (Cirelli & Tononi, 2000a;

Hairston et al., 2004). So, BDNF might be an important "bridge" connecting the topics of stress, depression and sleep.

The mechanisms of propagation of depressive disorder and related sleep disturbances remain unclear. Studying sleep in depressed adolescents can provide insight into the sleep structure changes common for early-onset depression. Genetic mechanisms that can predispose to depression and/or sleep problems can be investigated using large-scale birth cohort studies, which start during pregnancy and follow children through adolescence and beyond. In-depth mechanistic studies require, however, the use of experimental animals. Among animal models of early life stress, models based on the manipulation of mother-infant interaction, namely cross-fostering models (Barbazanges et al., 1996; Meaney & Szyf, 2005), are the less studied in the context of depressive-like behaviour and sleep alterations. Yet, they can be an invaluable tool in understanding the molecular mechanisms underlying the effect of early life stress and relationship between depression and sleep disturbances.

2. Review of the literature

2.1. Interconnection between depression and sleep disorders

2.1.1 Sleep: physiology, regulation and development

Sleep. Sleep is a reversible behavioural state of reduced responsiveness, which fulfils some universal, but yet unknown, vital function (Cirelli & Tononi, 2008). Sleep occurs in repeating periods, during which the body alternates between two different types of sleep: rapid eye movement (REM) and non-rapid eye movement (NREM) sleep (Hobson, 2005). NREM sleep is characterized by slow frequency and high amplitude waves in the electroencephalogram (EEG) (Figure 1). In humans, the NREM sleep can be further divided into three stages (N1, N2 and N3) distinguished by the amplitude of slow wave oscillations. N1 is the lightest stage of sleep, which is characterized by lowering EEG frequency and muscle tone, measured by electromyography (EMG). N2 stage is characterized by slow brain waves with occasional bursts of rapid waves (sleep spindles, 11-16 Hz). N3 stage is the deepest sleep stage, characterized by slow-wave activity (0.5-4.5 Hz) of high amplitude (>75μV). Usually sleep stages alternate during the night within cycles of approximately 1.5-2 h. Sleep is entered through N1 and N2 stages and progresses to the deepest stage N3, which is also referred as slow-wave sleep (SWS). The cycle usually ends with REM sleep episode. REM sleep is characterized by high frequency and low amplitude waves in EEG, abolished muscle tone, and rapid eye movements, which can be detected by electrooculography (EOG). The deepest NREM stages tend to predominate during the first sleep cycles, when the sleep pressure is high, and decrease towards the morning, while REM sleep and light stages of NREM mostly predominate during the second part of the night. The total length of sleeping bout is about 6-8h, although the total duration of sleep per day can vary considerably between individuals.

Although human subjects are widely used in studies on sleep and sleep disorders, animal models provide an invaluable tool for research aimed at understanding the physiology of sleep and the underlying mechanisms of sleep

disorders. While laboratory rodents (rats and mice) are the most commonly used animal models in sleep research, the differences in their sleep as compared to humans must be considered. Human sleep is consolidated usually to one main sleep phase, taking place during the dark period of the day. In contrast, rats are polyphasic animals, and their NREM–REM sleep cycles are relatively short (about 10-14 mins) and occur periodically throughout the day, although predominantly during the light phase of the day (Toth & Bhargava, 2013). In spite of these differences, the EEG pattern during sleep (Figure 1) and general mechanisms of sleep regulation are remarkably similar between rat and human (Borbely, 1982; Vyazovskiy & Tobler, 2005).

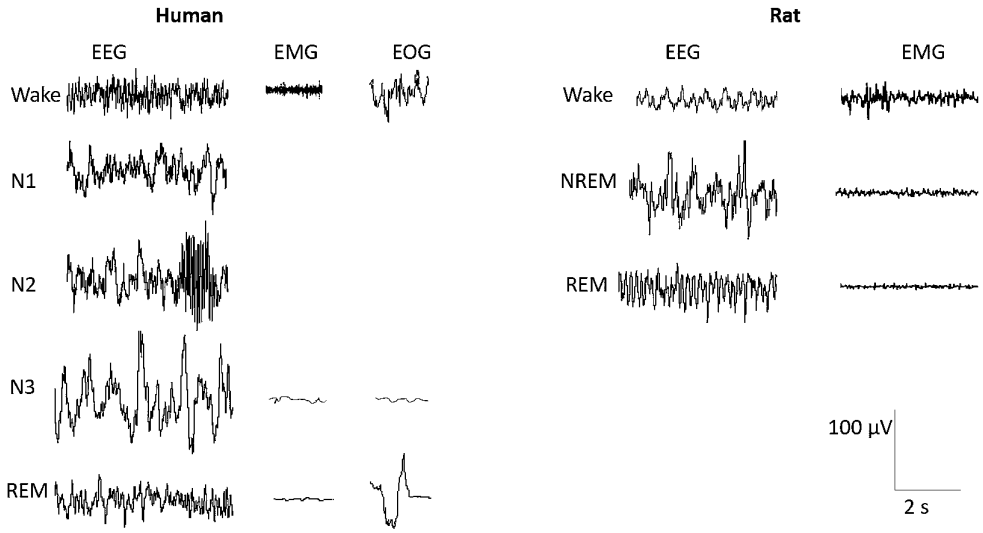

Figure 1. EEG, EMG and EOG patterns in humans and rats during wakefulness and sleep. EEG: electroencephalogram, EMG: electromyogram, EOG: electrooculogram, REM: rapid eye movement sleep, NREM: non-rapid eye movement sleep, N1-3: NREM sleep stages 1-3.

Sleep regulation. In spite of sleep being a behavioural state of quietness, it is an actively regulated process, not simply the passive result of diminished waking. According to the two-process model of sleep regulation, sleep is regulated by two interacting components: the homeostatic (Process S) and the circadian (Process C) (Borbely, 1982). The original version of this sleep regulation model was established to account for sleep regulation in humans (Borbely, 1982) and was later verified in rodents (Tobler et al., 1992).

The C process regulates the daily rhythms of the body and brain. Circadian patterns of activity are found in cells throughout the body, but the main circadian pacemaker is located in the suprachiasmatic nucleus of the hypothalamus (Moore, 2007). This nucleus provides an oscillatory pattern of activity which controls the timing of the sleep-wake cycle and coordinates it with circadian rhythms in other brain areas and tissues.

The homeostatic component (Process S) of sleep regulation is dependent on the duration of preceding wake period. The homeostatic sleep pressure rises during waking and results in sleep. Recently it has been shown that not only the duration of preceding wakefulness is affecting the homeostatic component of sleep regulation, but also the type and amount of activity during waking (Hanlon et al., 2009; Huber et al., 2007).

One of the principal physiological marker of sleep homeostasis (Process S) is the NREM sleep slow-wave activity (SWA, defined as spectral power in the 0.5-4.5 Hz band) (Borbely & Achermann, 1999). SWA power increases progressively in the course of wakefulness (Borbely et al., 1981), and gradually reduces during sleep, including naps (Borbely & Achermann, 1999). In both humans and rats, the highest SWA is observed at the beginning of the resting phase and it declines towards its end (Dijk, 2009; Franken et al., 1991b). Thus, SWA is considered as a reliable indicator of sleep pressure and may be linked to the restorative function of sleep (Dijk, 2009).

Sleep across development. The amount of sleep and sleep characteristics change throughout the life span. The average sleep duration in adult humans varies between 6 to 9 h, while sleep length in infants is up to 16-17 h per day. Major changes occur in the sleep-wake pattern across the first year of life when sleep changes from multiphasic to consolidated nocturnal uninterrupted sleep (Henderson et al., 2011). Sleep structure undergoes significant changes as well. The proportion of active sleep (analogous of REM sleep in adults) is high after birth and gradually declines with age (reaching an approximate adult value of 25% at the age of 6 months) and proportion of quiet sleep (analogous of NREM sleep in adults) increases (de Weerd & van den Bossche, 2003; Mirmiran et al., 2003). Hence, REM sleep is believed to be important for the brain maturation during early postnatal life (Marks et al., 1995).

Another significant developmental change in sleep occurs during adolescence, when sleep and its regulation is undergoing significant maturational changes (Brand & Kirov, 2011; Colrain & Baker, 2011; Hagenauer et al., 2009). The most remarkable change is the reduction in the amount of SWS (by up to 40%) and SWA (by up to 60%) (Baker et al., 2011; Buchmann et al., 2011; Campbell & Feinberg, 2009; Colrain & Baker, 2011; Feinberg et al., 2006; Jenni & Carskadon, 2004). Beside changes in sleep, the major maturational changes occur in brain neuronal networks, so called synaptic pruning (Hagenauer et al., 2009). Sleep, and particularly SWA, are closely interconnected with these changes and the observed decline in SWA during adolescence can represent a reduction in synaptic density occurring at this period of life (Feinberg & Campbell, 2010; Huttenlocher, 1979).

As in humans, the EEG in new-born rats is undifferentiated and EEG power gradually increases during early postnatal life. Sleep staging is usually based on behavioural criteria, defining active and quiet sleep (Frank & Heller, 1997). The adult features for each sleep stage are established around 3-4 weeks after birth. EEG maturation during adolescence in rats, as well as in humans, involves a decline in the SWA range (Mendelson & Bergmann, 1999).

2.1.2 Major depressive disorder

The incidence of depression is relatively low before puberty, but rises dramatically from the early teens with a preponderance among girls (Costello et al., 2003). According to epidemiological research, the incidence of MDD increases rapidly across adolescence (Burke et al., 1990; Newman et al., 1996; Oldehinkel et al., 1999) and its prevalence ranges from 5 to 12% (Haarasilta et al., 2001; Hankin et al., 1998; Lewinsohn et al., 1994; Oldehinkel et al., 1999). In a recent meta-analysis the prevalence of MDD was estimated as 5.7% among 13-18- year old teenagers with a female to male ratio of 1.3:1 (Costello et al., 2006). Moreover, depression during adolescence frequently recurs or persists into adulthood, being a major contributing factor to health-related disability of young adults (Newman et al., 1996; Pape et al., 2012; Pine et al., 1999; Sagatun et al., 2016). After adolescence and throughout adulthood, the gender divergence in depressed mood continues with adult women being twice as likely as men to experience depression (Nolen-Hoeksema, 2001).

MDD in adulthood is characterized by depressed mood and loss of interest or pleasure in daily activities that is present across most of the situations for more than two weeks, and it is usually accompanied by five or more of the following symptoms: depressed mood, markedly diminished interest or pleasure in activities previously found pleasurable, significant weight change (5%) or change in appetite, insomnia or hypersomnia, psychomotor agitation or retardation, fatigue or loss of energy, feelings of guilt or worthlessness, reduced concentration, and thoughts of death or suicide (*American Psychiatric Association. Diagnostic and Statistical Manual of Mental Disorders*, 2013). The above-mentioned symptoms cause clinically significant distress and impairment in social, occupational, or other important areas of life.

MDD in adolescence is diagnosed by the same set of symptoms as in adults, besides two exceptions (*American Psychiatric Association. Diagnostic and Statistical Manual of Mental Disorders*, 2013). First, depressed mood in children and adolescents can be substituted by irritability, and second, the duration for dysthymic disorder is one instead of two years. The clinical syndrome of depression is remarkably similar among children, adolescents, and adults. However, there are some minor developmental differences in the phenomenology of depression (Rao & Chen, 2009; Weiss & Garber, 2003).

Understanding the pathophysiology of MDD is a challenging scientific problem and despite numerous studies, it still remains unclear (Brigitta, 2002; Hasler, 2010; Krishnan & Nestler, 2008). A few key factors in development of depressive disorder have given the foundation for basic theories of MDD pathophysiology.

Genetic theory. From early adolescence through adulthood women have a twofold higher risk of MDD as compared to men (Nolen-Hoeksema, 2001; Van de Velde et al., 2010). The neurobiological basis for this increased risk is unknown, and could be related to gender differences in hormonal or stress response systems, or to sexual dimorphism in brain areas involved in depression pathophysiology (Nestler et al., 2002; Nolen-Hoeksema, 2001). In a large twin study of children and adolescents it has been shown that postmenarcheal adolescent girls had elevated heritability for depressive disorders compared with boys or premenarcheal adolescent girls (Silberg et al., 1999). This finding made

the authors to conclude that in adolescent girls, increased risk for depressive disorder is explained by an emerging genetic liability for depression combined with an increase in stressful life events. Family, twin, and adoption studies provide very solid and consistent evidence that MDD is a familial disorder and that this familiarity is mostly due to genetic factors (Sullivan et al., 2000). The influence of genetic factors is accounted for 30-40%, the remaining 60-70% of the variance in MDD vulnerability is explained by non-genetic factors, which usually are individual-specific environmental effects, such as adverse childhood events or other lifetime trauma and ongoing or recent stress. Several candidate genes have been extensively studied regarding their association with MDD, including serotonin transporter (*5HTT/SLC6A4*), serotonin receptor 2A (*HTR2A*), brain-derived neurotrophic factor (*BDNF)* and tryptophan hydroxylase (*TPH2*) (Lohoff, 2010). The results suggest that, as in other complex psychiatric disorders, there is no universal susceptibility gene for MDD, but rather multiple genes with small effect sizes contribute to the emergence of depression.

Stress hormone theory. There is convergent evidence that the hypothalamic-pituitary-adrenal (HPA) stress system plays a major role in the pathogenesis of certain types of depression (Pariante & Lightman, 2008). The HPA axis is one of the major neuroendocrine systems that is responsible for adaptation to changing demands of the environment and thereby contributes to the maintenance of different bodily functions (McEwen, 2004). Hyperactivity of HPA system during MDD contributes to a range of physical long-term consequences of depression, such as coronary heart disease, type II diabetes, and osteoporosis (Gold & Chrousos, 1999). The role of altered stress hormone secretion (cortisol; a glucocorticoid) in depression vulnerability is particularly important in the mechanisms of childhood adverse experience (Heim et al., 2008). Moreover, epigenetic regulation of the glucocorticoid receptors is associated with early life adverse experience, as evidenced by both human and animal research (McGowan et al., 2009; Weaver et al., 2004). Environmental programming of gene expression through epigenetic regulation represents one possible mechanism that links early life stress to abnormal HPA axis function and increased risk of psychiatric diseases in adulthood.

Monoamine-deficiency theory. According to this theory, the pathophysiological basis of depression is a consequence of insufficient amount of the neurotransmitters serotonin, norepinephrine or dopamine in the central nervous system. The monoaminergic neurons are located in the midbrain and brainstem nuclei and send projections to large areas of the entire brain, particularly to the cortex. Monoamine theory of depression derives from the clinical observations that enhancing of serotonin and noradrenaline transmission have antidepressant effect (Prins et al., 2011). Experimental depletion of the serotonin leads to development of depressive symptoms in subjects at increased risk of depression (Neumeister et al., 2004). Almost all established antidepressants target the monoamine systems: they amplify serotonin or norepinephrine signalling by inhibiting their catabolism or reuptake at the synaptic cleft (Mann, 2005). The several classes of antidepressant drugs include: selective serotonin reuptake inhibitors (SSRIs), norepinephrine-reuptake inhibitors, and dual-action agents that inhibit uptake of serotonin and norepinephrine. For long time the monoamine-deficiency theory was considered to be the most clinically relevant neurobiological theory of depression. However, there are several limitations in this theory. There are cases of full or partial resistance to the monoamine-based drugs, which suggests that dysfunctions of monoaminergic neurotransmitter systems found in MDD can be secondary to effects of other, more primary abnormalities (Massart et al., 2012). Moreover, therapeutic responses with antidepressants can only be achieved after at least 2–3 weeks of treatment, whereas antidepressants alter synaptic monoamine levels within hours (Frazer & Benmansour, 2002) .

Inflammation theory. Recently, the role of inflammation in depression development has been extensively investigated (Miller & Raison, 2016). Sickness behaviour shares many symptoms with depressive disorder, including fatigue, anhedonia, psychomotor retardation, and cognitive impairment. Patients with MDD exhibit all principal features of an inflammatory response, such as increased expression of pro-inflammatory cytokines (interleukin (IL) - 1β, 2, 6, 8, 12, interferon γ, tumor necrosis factor (TNF)-α) and their receptors and increased levels of acute-phase reactants, chemokines and soluble adhesion molecules in peripheral blood and cerebrospinal fluid (Miller et al., 2009; Zorrilla et al., 2001). In

rodents, administration of cytokines induces depressive-like symptoms, including anhedonia and anxiety (Anisman & Merali, 2002).

Circadian disruption theory. In depression, multiple circadian rhythms are disturbed, including the rhythmic secretion of many hormones, body temperature circadian profile, as well as sleep-wake cycle (Germain & Kupfer, 2008; Hasler & Northoff, 2011). Among the most common circadian disruptions that MDD patients experience are: shortened latency of REM sleep, early morning awakenings, early morning rise of adrenocorticotropic hormone (ACTH; stimulates cortisol synthesis in the adrenal glands) and night peaks of prolactin and growth hormone. All these abnormalities are the symptoms of a phase advance circadian disorder. The fact that in depression a variety of circadian functions are disturbed led to a speculation that the association with depression is due to a disruption in central circadian clock function and not to an alteration in one specific rhythm (Turek, 2007).

Neurotrophin deficiency theory. Neuronal atrophy in the hippocampus and other forebrain regions, as well as reduced neurotrophic factor expression observed in depressed patients, have supported the neurotrophin deficiency hypothesis of depression (Castren et al., 2007; Duman & Monteggia, 2006). Several neurotrophic factors are associated with depression or antidepressant action, and the most studied of them is BDNF (Castren & Rantamaki, 2010). More detailed description of this theory will be provided in the section *1.2.2 Role of BDNF in depression* after giving basic information about BDNF.

Additional MDD theories include altered glutamatergic and reduced GABAergic neurotransmission, impaired endogenous opioid function (Belmaker & Agam, 2008), glial-based synaptic dysfunction (Rial et al., 2016) and vitamin D deficiency (Milaneschi et al., 2014).

Integrated model of current MDD hypotheses. The existence of so many theories of depression, which are supported by solid evidences from epidemiological, clinical and experimental studies clearly argue against a "unified hypothesis of depression" and suggest that depression is clinically and etiologically heterogeneous disorder. Integration between these numerous hypotheses of depression pathophysiology may better explain the biology of depression and its heterogeneous clinical presentations.

Hyperactivation of HPA axis, which is a common finding in patients with MDD results in high level of the stress hormone cortisol, which may cause impairment in neuroplasticity and cellular resistance (Manji et al., 2003). Indeed, glucocorticoids are well established to affect synaptic plasticity and to contribute for synaptic atrophy in several brain regions (Sousa et al., 2008). Moreover, an imbalance between glucocorticoid and mineral corticoid receptors in depression along with high-density glucocorticoid receptors may also contribute to the hippocampus' susceptibility to neuronal damage (de Kloet et al., 2007). Furthermore, HPA axis overactivity, in conjunction with amygdala activation in depression, leads to increased sympathetic tone, which promotes the release of pro-inflammatory cytokines (TNF-α, IL-1, 6) from macrophages (Maletic et al., 2007) (Figure 2). On the one hand, this increase in pro-inflammatory cytokines has been associated with loss of insulin and glucocorticoid receptors sensitivity, which further aggravate metabolic and neuroendocrine abnormalities. On the other hand, pro-inflammatory cytokines may also diminish neurotrophic support and monoamine neurotransmission (including serotonin and dopamine) that can lead to further neuronal apoptosis and glial damage. Stress, depression and immune dysregulation lead to activation of microglia that then contribute to the existing immune disruption by additional release of pro-inflammatory cytokines (Maletic et al., 2007). Pro-inflammatory cytokines, such as TNF-α and IL-1β has been shown to decrease neurogenesis in selected brain areas, possibly by affecting *BDNF* expression in neurons (Audet & Anisman, 2013; Maes et al., 2009). In addition to cytokines, glucocorticoids can also affect BDNF transcription by interfering with its transcriptional mechanisms (e.g. by decreasing cyclic-AMP-response-element-binding protein (CREB) activity) (Krishnan & Nestler, 2008).

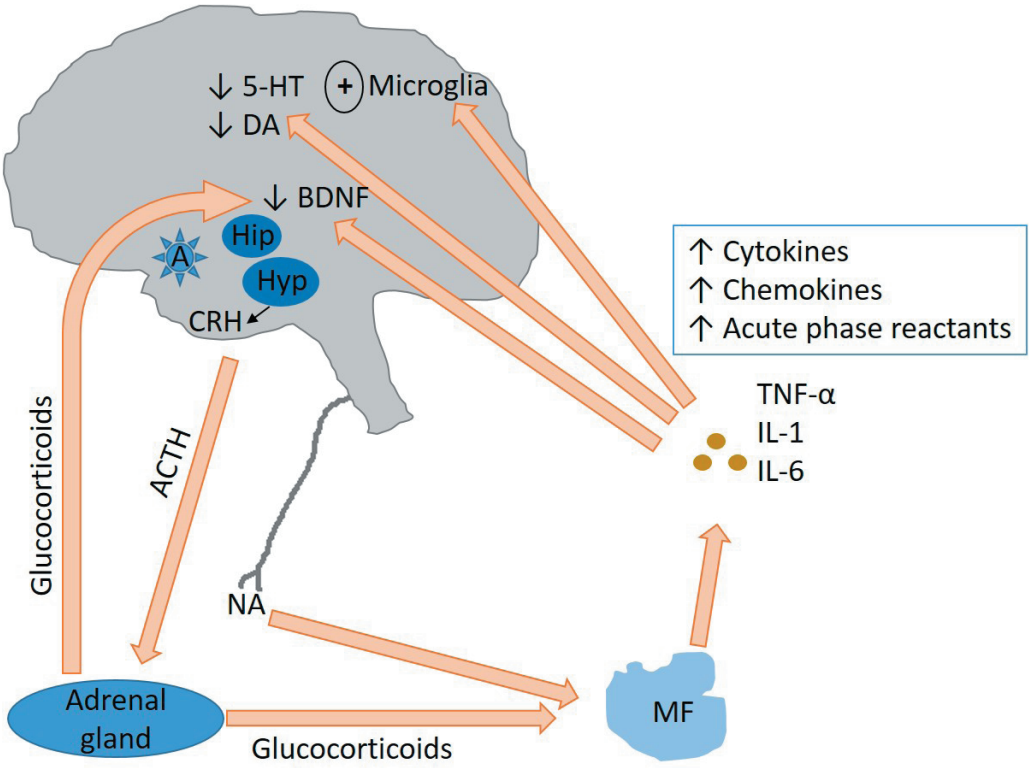

Figure 2. Simplified scheme of interaction between different hypotheses of depression. Hyperactivation of HPA axis leads to an increased release of CRH and pro-inflammatory cytokines (TNF, IL-1, 6), and, in conjunction with amygdala activation, causes increased sympathetic tone. Sympathetic overactivity contributes to immune activation and release of inflammatory cytokines. Inflammatory cytokines further interfere with monoaminergic and neurotrophic signalling and activate microglia. HPA: hypothalamic–pituitary–adrenal axis, A: amygdala, Hip: hippocampus, Hyp: hypothalamus, ACTH: adrenocorticotropic hormone, CRH: corticotrophin releasing hormone, BDNF: brain-derived neurotrophic factor, 5-HT: serotonin; DA: dopamine, NA: noradrenaline, MF: macrophage, TNF: tumor necrosis factor, IL: interleukin.

2.1.3 Sleep in major depressive disorder

Depressive disorder is usually characterized by subjective sleep disturbances (Nutt et al., 2008; Thase, 2006), which is one of the diagnostic symptoms of depression. Multiple disturbances in sleep structure are evident in about 45% of

depressed outpatients and 80% of more severely depressed inpatients (Tsuno et al., 2005). Moreover, people with persistent sleep problems have elevated risk of developing MDD within one year (Breslau et al., 1996; Paunio et al., 2015). The relationship between sleep and depression is bidirectional, complex and has remained poorly understood.

The whole range of sleep problems observed in depressed adults (Armitage, 2007) can be divided in three groups (Steiger & Kimura, 2010): impaired sleep continuity (prolonged sleep latency, increased number of night awakenings); rapid eye movement (REM) sleep disinhibition (shortened REM sleep latency, elevated REM sleep frequency and duration); and NREM sleep changes (decreased NREM sleep duration and reduced SWA). The majority of depressed patients complain about insomnia (~80%), while hypersomnia is observed in some cases (15-35%) (Steiger & Kimura, 2010).

Different REM and NREM sleep parameters have been previously shown to correlate with the severity of depressive symptoms in adults (Perlis et al., 1997). The sleep parameters include SWA, increased relative amount of N1 stage, increased REM activity and efficiency, increased sleep latency, and depressive symptoms included loss of appetite/weight, decreased libido, sleep problems, feelings of dissatisfaction and failure.

As well as in adulthood, sleep disturbance in adolescence is one of the key symptoms of depressive disorder. Moreover, the prevalence of insomnia is rising from childhood to adolescence reaching about 11-13% (Hysing et al., 2013; Johnson et al., 2006). Depressed adolescents often complain about their sleep (Liu et al., 2007; Urrila et al., 2012), but the results from objective polysomnographic studies show different findings. Some studies in depressed adolescents have demonstrated sleep changes, which are common for depressed adults, such as increased sleep onset latency, REM sleep amount and density, decreased REM sleep latency and sleep efficiency (Augustinavicius et al., 2014; Kutcher et al., 1992; Lopez et al., 2010; Lopez et al., 2012; Rao et al., 2009; Robert et al., 2006). Few studies have also reported lower SWA in the first NREM sleep episode and its irregular dissipation through the night in depressed adolescents compared to healthy ones (Armitage et al., 2001; Lopez et al., 2012). Recently, it has also been shown that the topographical pattern of SWA distribution in depressed adolescents is characterized by increased

SWA over the frontal cortex compared to healthy controls (Tesler et al., 2016). Other studies have failed to demonstrate abnormalities in sleep architecture (Dahl et al., 1990; Goetz et al., 1987; Khan & Todd, 1990). The difference in findings can be explained by e.g. the heterogeneity of studied samples in terms of gender, age/pubertal status, and the variable clinical features/symptoms of depression.

Sleep abnormalities observed in depression point to a disruption in both homeostatic and circadian components of sleep regulation. Decreased REM latency, increase in waking time and N1 stage, and waking up early could be associated to a circadian disturbance resulting in earlier onset of sleep rhythms. Whether the circadian rhythm disruption is a cause or a consequence of depression is a subject of many studies focusing on genetic control of the mammalian clock (Germain & Kupfer, 2008).

The homeostatic process deficiency hypothesis suggests that depression is accompanied by an attenuated build-up of sleep pressure, and S process does not rise to its usual level in the course of waking (Borbely, 1987; Borbely & Wirz-Justice, 1982). This leads to subsequent reduction in SWA power during sleep. This theory is supported by studies that demonstrate reduced SWA in both adult (Armitage, 2007; Borbely et al., 1984; Hoffmann et al., 2000) and adolescent (Armitage et al., 2001; Lopez et al., 2012) depressed patients. Diminished SWA in sleep was also observed in animal models of depression (Savelyev et al., 2012; Touma et al., 2009). Moreover, SWA distribution across the first two NREM sleep episodes (known as delta sleep ratio) has been previously linked to clinical outcome and responsiveness to depression treatment in adult patients with MDD (Ehlers et al., 1996; Kupfer et al., 1990): a better response to therapy was observed in subjects with a higher delta sleep ratio.

Suggestion of participation of both S and C processes in depression rises from observations that total or partial (in the second half of the night) sleep deprivation (SD) in MDD patients improves mood during the next day (Giedke & Schwarzler, 2002). SD rapidly improves mood in depressed patients, but a subsequent sleep/nap commonly reverses this improvement. Mood in many MDD patients is also worse in the morning and gradually improves during the day, and then reverses back to depression in the course of sleep. One of the explanations for the therapeutic effect of SD is that it changes the phase-angle between advanced

circadian pacemaker and sleep need (Wirz-Justice & Van den Hoofdakker, 1999). According to the homeostatic S process deficiency theory, SD is helpful because the level of sleep pressure is transiently increased to normal, and relapse occurs after recovery sleep due to the return to low levels of sleep pressure. Moreover, according to recent research, the mechanism of antidepressant effect of SD can be associated with rapid increase in BDNF serum levels on the day following the partial or total SD (Gorgulu & Caliyurt, 2009; Schmitt et al., 2016). However, the above mentioned possible mechanisms are not mutually exclusive.

Thus, depression and sleep disturbances are tightly interconnected. The existence of common subtracts for sleep and mood regulation could potentially explain the bidirectional association between depression and sleep disorders.

2.2. BDNF: a possible bridge between major depression and sleep disorders

2.2.1 Characteristics of BDNF

BDNF is one of the most widely distributed neurotrophins of the central nervous system, which is abundantly expressed in the developing and adult mammalian brain. In immature neurons, BDNF is involved in growth, differentiation, maturation and survival while in mature neurons it plays an important role in synaptic plasticity, augmentation of neurotransmission and regulation of receptor sensitivity (Calabrese et al., 2009; Gonzalez et al., 2016; Numakawa et al., 2010). BDNF mediates its biological effects via two types of receptors: TrkB and p75. Recent studies indicate that binding of mature BDNF protein with TrkB receptors promotes neuronal survival, enables long-term potentiation and enhances synaptic plasticity, whereas binding of precursor (pro-BDNF) with p75 receptors has biological effects oppose to those of mature BDNF (Lu et al., 2005). Thus, the balance between pro- and mature BDNF on the one hand, and between the p75 and TrkB receptors on the other, is of critical importance in determining the functional characteristics of the BDNF signal.

Gene structure and regulation. The BDNF protein is coded by the *BDNF* gene. Rodent and human *BDNF* genes have multiple untranslated 5′-exons (I–IX). Every

single 5′-exon is normally spliced to one coding exon with two different 3′-UTRs (untranslated region), resulting in numerous possible *BDNF* transcripts variants. Each exon has its own promoter, which is located upstream of corresponding 5′ -noncoding exon allowing time- and brain area-specific control of *BDNF* expression in an activity-dependent manner (Aid et al., 2007; Chen & Chen, 2017; Karpova, 2014). This provides adequate adaptive responses to different environmental factors, such as physical exercise, learning, environmental enrichment or stress (Calabrese et al., 2009). Interestingly, all transcripts are eventually translated into an identical protein (Aid et al., 2007; Pruunsild et al., 2007).

The regulation of *BDNF* expression, depending on environmental conditions, is mediated through epigenetic mechanisms. Epigenetic regulation is a recently emerged frontier in genetic science, which has been shown to be able to alter DNA accessibility and chromatin structure, thereby regulating patterns of gene expression without changing the DNA sequence (Bird, 2007). Epigenetic mechanisms of regulation include methylation, acetylation, phosphorylation, ubiquitylation, sumoylation, and microRNA control (Weinhold, 2006). In activity dependent *BDNF* regulation, the most frequently studied mechanisms have been DNA methylation and translational histone modifications.

DNA methylation is an essential epigenetic mechanism for gene silencing (Figure 3). DNA methyltransferases (DNMT) catalyze the transfer of a methyl group from a donor S-adenosylmethionine to a cytosine at position C5, resulting in a new nucleotide 5-methylcytosine (5-mC), generally in the context of CpG dinucleotides (Karpova, 2014). CpG dinucleotides tend to cluster in CpG islands (CG-rich DNA regions). Methylated CpG islands are typically associated with silent DNA whereas unmethylated CpG islands are targets for transcription factors. Evidence for existence of a specific DNA demethylase that may directly remove the methyl group from 5-mC is controversial, but studies suggest that it can be done with the help of DNA glycosylases through a base excision-repair pathway (Bhattacharya et al., 1999; Zhu, 2009).

In addition to changes in DNA-methylation, post-translational modifications of histones are involved in modulation of *BDNF* gene expression (Chen & Chen, 2017; Karpova, 2014; Mitchelmore & Gede, 2014). One of these modifications is histone acetylation, which influences chromatin structure. Histone acetylation is

regulated by histone acetyltransferases (HATs) and histone deacetylases (HDACs) (Figure 3). It should be mentioned that DNA is wrapped around nucleosomes, which each consist of 8 histone protein cores that are highly basic. HATs catalyze the lysine acetylation that neutralizes the lysine's positive charge and weakens the interactions between the negative DNA backbone and the histone, making the DNA more accessible for the transcription machinery. In contrast, HDACs mediate the removal of the acetylation mark, resulting in the formation of DNA that is less accessible to transcription factors. Another mechanism affecting gene transcription is histone methylation/demethylation, which is regulated by histone methyltransferases (HMTs) and demethylases (HDMTs), respectively. For example, methylation at lysine (K) 27 on histone H3 (H3K27) is usually associated with transcriptional repression (Tsankova et al., 2007). Adverse environmental conditions have been shown to reduce *BDNF* expression through repressive histone H3 methylation at the *BDNF* promoters in rodent brains (Karpova et al., 2010; Onishchenko et al., 2008), whereas positive experience has been shown to increase *BDNF* expression through histone modifications (Kuzumaki et al., 2011).

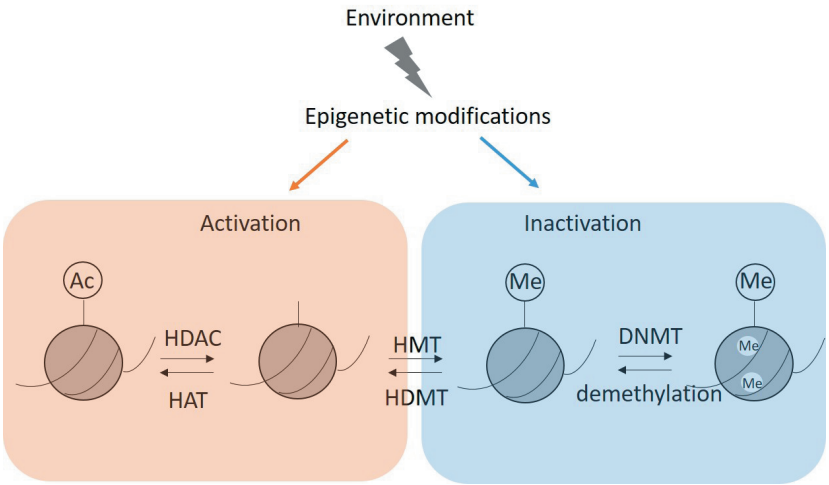

Figure 3. A scheme of reversible epigenetic modifications involved in the regulation of *BDNF* gene transcription. Acetylation (Ac) of histones leads to activation, whereas methylation (Me) leads to inactivation of gene expression. Methylation (Me) of cytosines at CpG dinucleotides in DNA also leads to inactivation of gene expression. DNMT: DNA methyltransferase; HAT: histone acetyltransferase; HDAC: histone deacetylases; HMT: histone methyltransferase; HDMT: histone demethylase. Modified from (Mitchelmore & Gede, 2014).

BDNF gene polymorphisms. Among all *BDNF* gene polymorphisms, the most widely studied is Val66Met polymorphism, a naturally occurring variation in the human *BDNF* gene, resulting in a valine (Val) to methionine (Met) substitution at codon 66. This substitution does not affect mature BDNF protein function, but it has been shown to alter the intracellular trafficking and packaging of pro-BDNF (Egan et al., 2003). This in turn leads to the reduction of activity-dependent release of the mature peptide (Egan et al., 2003). It is still unclear what the consequences of the Val66Met polymorphism are on brain function, as both alleles have been associated with different disease processes including substance-related disorders, schizophrenia, and eating disorders (Gratacos et al., 2007). It is also known that *BDNF* allele frequencies differ by ethnicity with higher prevalence of Met allele in Asian population (40-50%) compared to Caucasians (25-32%), where Val/Val genotype is the most prevalent (Pivac et al., 2009).

2.2.2 Role of BDNF in depression

BDNF hypothesis of MDD originates from the observations that acute and chronic stress in humans decrease endogenous neurotrophin levels and can lead to atrophy and cell loss in limbic brain structures (especially hippocampus), the structures involved in mood regulation (Duman, 2004; Duman & Monteggia, 2006). In contrast to the effects of stress, antidepressant treatment increases the expression of neurotrophic factors and increases neurogenesis (Castren & Rantamaki, 2010; Lee & Kim, 2010). Thus, depression is considered to be a result of inability of the brain to make appropriate adaptive responses to environmental stimuli as a consequence of impaired synaptic plasticity and structural plasticity, mediated by neurotrophins.

BDNF in depression. In experimental studies using animal models of depression, it has been shown that different types of stressors (immobilization stress, foot shocks, social defeat, and early maternal deprivation), as well as exogenous corticosterone administration, reduce hippocampal *BDNF* expression (Roceri et al., 2002; Schaaf et al., 1998; Smith et al., 1995; Ueyama et al., 1997), whereas adrenalectomy increases the level of BDNF in the hippocampus, indicating that the regulation of hippocampal BDNF occurs partially through HPA axis (Chao et al., 1998). Similar observations were made in the frontal cortex, where

corticosterone administration decreased level of *BDNF* expression, while treatment with antidepressants returned its level to normal (Dwivedi et al., 2006). It should be noted that sole central reduction in BDNF level, observed in heterozygous *BDNF* knockout mice, is not sufficient to produce depressive-like behaviour, but rather, the reduction appears to increase susceptibility to stress: exposure of *BDNF* heterozygous knockout mice to stress induces depressive-like behaviour, as does the blockade of BDNF-TrkB signalling following stress (Duman et al., 2007).

The relevance of findings obtained in basic animal research studies has been confirmed by findings in clinical studies on depressed patients. Post-mortem studies demonstrate that levels of BDNF in the hippocampus or prefrontal cortex are decreased in depressed suicide subjects as compared to matched, non-depressed controls (Dwivedi et al., 2003). Moreover, depressed individuals, who were treated with antidepressant drugs, had increased *BDNF* expression compared to medicine-free patients (Chen et al., 2001). Recent clinical studies on serum or plasma BDNF levels in patients with MDD demonstrated that medicine-free MDD patients have lower BDNF levels than healthy individuals (Gonul et al., 2005; Lee et al., 2007; Shimizu et al., 2003). Although blood platelets represent the main source of BDNF in serum (Chacon-Fernandez et al., 2016), BDNF can also, to some extent, cross the blood-brain barrier in both directions, indicating that circulating BDNF could partially originate from neurons of the brain (Pan et al., 1998; Poduslo & Curran, 1996). In this regard, differences in BDNF levels in plasma may represent the variable BDNF secretion in the human brain.

BDNF and antidepressant drug action. Delayed onset of action of conventionally used antidepressant drugs prompted the idea that other mechanisms, than just increase in monoamine levels, e.g. BDNF-mediated reorganization of neuronal networks, could be responsible for the antidepressant response.

Indeed, it has been shown in many studies that commonly used antidepressants increase BDNF mRNA and protein expression in the rodent brain, particularly in the hippocampus and cortical regions in a timeframe similar to the onset of the antidepressant-like response (Duman & Monteggia, 2006; Hajszan et al., 2005; Nibuya et al., 1995; Warner-Schmidt & Duman, 2006). Moreover, the antidepressant effects of BDNF have been shown to be mediated by TrkB activation (Saarelainen et al., 2003). Direct infusion of BDNF protein into the midbrain or

hippocampus also led to an antidepressant-like effect in rodents (Shirayama et al., 2002; Siuciak et al., 1997).

Recently it has been suggested that one of the mechanism of antidepressant effect of BDNF is the ability to reactivate developmental-like neuronal plasticity in the adult brain (Castren & Rantamaki, 2010). Previous work, using the monocular deprivation during early development, resulting in impaired vision of the deprived eye in adulthood, had shown that the impairment could be recovered during adulthood by chronic administration of fluoxetine (an antidepressant of the SSRI class) combined with simultaneous patching of the dominating eye (Maya-Vetencourt et al., 2008). It is known that activity-dependent reorganization of neuronal networks is efficient only during a critical period of postnatal development and it is greatly reduced after the end of this period. Thus, if the vision of deprived eye is not corrected during the critical period of development, it remains permanently impaired during adulthood. However, fluoxetine combined with rehabilitation was able to activate neuronal plasticity, leading to functional reorganization of neuronal networks even after the end of the critical developmental period. Moreover, during the treatment, BDNF levels increased in the visual cortex, and direct infusion of BDNF into the visual cortex mimicked the effects of fluoxetine (Maya-Vetencourt et al., 2008)), indicating that BDNF may be a critical mediator of the antidepressant drug effect.

Thus, according to this theory conventionally used antidepressants might work similarly to fluoxetine and "open" sensitive developmental-like window of BDNF-mediated plasticity in the brain, leading to the reorganization of neuronal networks and their better adjustment to environmental challenges.

BDNF Val66Met polymorphism and depression. Number of studies have associated *BDNF* Val66Met polymorphism with increased susceptibility to neuropsychiatric disorders, including MDD and anxiety disorder (Autry & Monteggia, 2012; Neves-Pereira et al., 2002; Sen et al., 2003; Sklar et al., 2002). However, the risk allele for depression and depression-related traits has not been clearly defined. It has been suggested that early life stress coupled with presence of Met allele may play a crucial role in increasing the susceptibility for MDD (Gatt et al., 2009). Met allele has been also associated with alterations in brain anatomy and memory, leading to impaired episodic memory and reduced hippocampal volume

(Hariri et al., 2003; Pezawas et al., 2004), findings often observed in subjects with MDD. The association between *BDNF* Val66Met polymorphism and neuroticism, as a major risk factor for psychiatric disorders, including depression (Jylha et al., 2009), has been studied, but the results are controversial (Huennerkopf et al., 2007; Itoh et al., 2004). Some studies indicate higher levels of neuroticism in Val allele homozygous individuals than in Met carriers, while others failed to show such association (Tochigi et al., 2006; Wray et al., 2008); one study found the association in females, but not in males (Itoh et al., 2004). Moreover, a link between *BDNF* allelic variation and another temperament trait, extraversion, has been identified (Terracciano et al., 2010). Val allele has also been found associated with higher anxiety trait (Lang et al., 2005), which is a potent risk factor for the development of depression. Also, a family-based study showed that Val allele is associated with childhood-onset mood disorders (Strauss et al., 2005)

In an animal model of the Val66Met polymorphism, Met allele was associated with neuronal development defects that lead to reduced hippocampal volume. However, this structural change did not contribute to depression-like behaviour in rodents (Frielingsdorf et al., 2010). In another rodent study, it has been shown that Met/Met mice had increased anxiety-related behaviour, which could not be reversed by fluoxetine treatment (Chen et al., 2006).

2.2.3 Role of BDNF in sleep regulation

BDNF has a particular role in sleep regulation, which has been shown in both experimental animal studies and clinical research (Cirelli, 2009; Faraguna et al., 2008; Giese et al., 2014b; Martinowich et al., 2011; Nishichi et al., 2013; Savelyev et al., 2012). In rats, the expression of *BDNF* mRNA and protein levels are higher during spontaneous or prolonged wakefulness than during sleep (Cirelli & Tononi, 2000a; Hairston et al., 2004). Moreover, there is a causal link between cortical BDNF expression and SWA in the following sleep episode: the higher the cortical expression of *BDNF* is during wakefulness, the larger is the increase in SWA during the subsequent sleep period (Faraguna et al., 2008). On the other hand, impaired secretion of BDNF protein or blocking its receptors leads to decrease in sleep duration and efficiency (Faraguna et al., 2008; Martinowich et al., 2011). Short-term SD (up to 12h) significantly increases *BDNF* expression in the cortex and

hippocampus (Cirelli & Tononi, 2000b; Fujihara et al., 2003), whereas prolonged sleep loss (for up to 7 days) led to smaller elevation in *BDNF* expression (Cirelli et al., 2006). One study analysed the effect of SD on different *BDNF* transcripts and found that activity-dependent *BDNF* transcripts *BDNF*1-4 and *BDNF* 9a are involved in sleep homeostasis (Martinowich et al., 2011). Moreover, disruption in activity-dependent *BDNF4* transcription led to impaired sleep homeostasis and short sleep duration (Martinowich et al., 2011). These findings suggest that *BDNF* transcription during spontaneous sleep-wake cycle can be exon-specific and epigenetic mechanisms might play an important role in BDNF regulation of sleep homeostasis.

In clinical studies, patients with symptoms of insomnia demonstrated decreased serum BDNF levels, and its level correlated with severity of insomnia (Giese et al., 2013; Giese et al., 2014b), while after therapeutic SD in MDD patients, fast increase in serum BDNF level was observed (Giese et al., 2014a). These contradictory findings can be explained by biphasic model of stress, and therefore, different response in BDNF expression to chronic vs. acute stress (Schmitt et al., 2016).

Studies on the role of *BDNF* polymorphisms, particularly Val66Met, in sleep regulation are scarce. Val66Met polymorphism has been associated with sleep efficiency, thus Val allele homozygotes individuals have been shown to have higher SWA in baseline sleep and post-sleep deprivation recovery sleep, than carriers of Met allele (Bachmann et al., 2012).Moreover, some changes in microarchitecture of sleep were observed in Met carriers (Guindalini et al., 2014).

2.3. Sleep and homeostatic plasticity

A large number of studies in humans and animals suggest that sleep is required for memory consolidation and learning, which are based on processes of long-term synaptic potentiation (LTP) and depression (LTD) (Rasch & Born, 2013). In classical Hebbian plasticity, the process of presynaptically emerged action potential activating the postsynaptic neuron, strengthens the synapse and forms the neurophysiological mechanism for memory (Fox & Stryker, 2017). New synapses are formed and strengthened in the course of the learning process. However,

this cannot continue endlessly, a restriction mechanism is needed. Homeostatic plasticity offers such mechanism(s).

The theory which links sleep with synaptic homeostasis (synaptic homeostasis hypothesis or SHY) was proposed by Tononi and Cirelli (Tononi & Cirelli, 2003, 2006) in order to explain the fundamental function of sleep as restoration of synaptic homeostasis. According to SHY, wakefulness is accompanied by synaptic potentiation in a large fraction of cortical circuits, resulting in a LTP and a net increase in synaptic weight. Rodent studies show a diffuse increase in synaptic density in animals exposed to enriched environments (Klintsova & Greenough, 1999) and activation of LTP-related genes, including the *BDNF,* during spontaneous wakefulness (Cirelli & Tononi, 2000a). The amount of SWA during sleep is tied to the amount of synaptic potentiation that has occurred during previous wakefulness (Cirelli & Tononi, 2000b; Huber et al., 2004; Kelly & Deadwyler, 2003). This relation between synaptic potentiation and following SWA is proportional: the higher the amount of synaptic potentiation in cortical circuits during wakefulness, the higher the increase in SWA during subsequent sleep. Moreover, it has been shown that SWA increases locally in the brain areas which were involved in synaptic potentiation during wakefulness, and this increase correlates with post-sleep improvements in performance (Huber et al., 2004). Evidence for a relationship between synaptic density and SWA also comes from the developmental studies: both synaptic density and SWA reach their peak in adolescence, after which they gradually decline (Feinberg & Campbell, 2010; Feinberg et al., 2006; Feinberg et al., 1990).

Moreover, according to SHY, SWA is associated with generalized depression or downscaling of synaptic strength, i.e. proportional reduction in the strength of all synapses converging onto the same neuron (de Vivo et al., 2017; Tononi & Cirelli, 2003, 2014). In other words, sleep in needed for weakening synapses through a process of downscaling or synaptic renormalization. Indirect evidences supporting this idea come from the experiments demonstrating a downregulation of LTP-related genes (Cirelli & Tononi, 2000a) and upregulation of molecules implicated in depotentiation/depression during spontaneous sleep (Cirelli et al., 2004) indicating that sleep may be an unfavourable condition to synaptic potentiation. Two recent imaging studies of cortical dendrite spine morphology support the idea

that sleep may play an important role in downscaling the strength and number of synaptic connections (Maret et al., 2011; Yang & Gan, 2012). These studies showed that the ratio of spines eliminated versus those formed was greater after a period of sleep than a period of wakefulness. Authors of SHY also speculate that a process of generalized downscaling may not be compatible with wakefulness, while it would be ideally compatible with sleep, a state during which the brain is both spontaneously active and virtually disconnected from the environment; and the reduced activity of the noradrenergic system during sleep would ensure that only downscaling occurs, and not potentiation (Tononi & Cirelli, 2003).

SHY is consistent with other theories that propose a similar synaptic weakening effect of sleep (Crick & Mitchison, 1983; Giuditta et al., 1995). A number of studies in insects, rodents and humans provide evidences that are in line with SHY (Gilestro et al., 2009; Huber et al., 2004; Liu et al., 2010; Vyazovskiy et al., 2008). However, this hypothesis also meets criticism since very little is known about the mechanisms that underlie the process of synapses weakening during sleep (Frank, 2012, 2013).

According to another view, homeostatic plasticity stabilizes neuronal excitability and maintains the so called neuronal firing rate homeostasis through mechanisms of intrinsic and synaptic plasticity (Turrigiano, 2011). Moreover, neuronal activity in neuronal circuits returns to baseline individual set point after periods of quietness and activity. It has been shown that the firing rate homeostasis is gated by sleep/wake states, and that sleep inhibits rather than promotes, firing rate homeostasis. This idea was prompted by the research studying the effects of visual deprivation on neuronal firing rates in the primary visual cortex using monocular deprivation model (Hengen et al., 2016). It has been shown that in spite the fact that monocular deprivation depressed at first the firing rate of individual neurons in visual cortex, firing rates returned precisely to the neuron's individual baseline later during periods of active wake, but not during the sleep. Thus, this finding suggests that the relationships between sleep and homeostatic plasticity are opposite to those that has been proposed in SHY. According to this model wakefulness but not sleep state enables the expression of homeostatic plasticity.

Recently, another model was introduced, which suggests that sleep is needed for homogenization of the firing rate distribution through cortical neurons

(Levenstein et al., 2017; Watson et al., 2016). This model is based on a study using large-scale recordings to examine the activity of neurons in the frontal cortex of freely behaving rats (Watson et al., 2016). It has been shown that the distribution of pyramidal cell firing rates was wide and strongly skewed toward high firing rates. Furthermore, neurons from different parts of this distribution were differentially modulated by sleep stages: NREM sleep reduced the activity of high firing rate neurons and tended to upregulate firing of slow-firing neurons, whereas REM sleep reduced firing rates across the entire rate spectrum.

All above mentioned theories have strong experimental support and whether they are describing the same phenomenon will be studied in future.

2.4. Adenosine: role in sleep regulation and depression

The idea that adenosine plays a role in sleep control came from the studies on systemic and central administrations of adenosine agonists and antagonists showing the sleep-inducing effects of first ones and the opposite effect of the last ones (Dunwiddie & Worth, 1982; Feldberg & Sherwood, 1954; Fredholm et al., 1999; Ticho & Radulovacki, 1991). The first direct evidence of spontaneous adenosine fluctuations during sleep-wake cycle came from *in vivo* microdialysis measurements of adenosine in freely behaving animals (Porkka-Heiskanen et al., 1997). During spontaneous sleep adenosine levels decrease in the cortex, basal forebrain, hypothalamus, and brainstem (Porkka-Heiskanen et al., 2000; Porkka-Heiskanen et al., 1997). However, during prolonged wakefulness (sleep deprivation) adenosine levels continue to rise or are stable only in one of these brain regions, the basal forebrain. Thus, a relatively specific area, the basal forebrain, which appears to be central in the regulation of sleep-wake cycle and recovery sleep, has been identified.

One of the best functional theories for adenosine's role in sleep–wake regulation derives from the fact that adenosine, a by-product of energy metabolism, may serve as a homeostatic regulator of energy in brain during sleep (Porkka-Heiskanen & Kalinchuk, 2011). Adenosine is an end product of ATP (adenosine triphosphate) utilization (Chikahisa & Sei, 2011). Intracellularly, ATP is metabolized to ADP (adenosine diphosphate) and then subsequently metabolized to AMP (adenosine monophosphate), which in turns is further metabolized to adenosine

by cytosolic 5′-nucleotidase enzyme. Extracellularly, ATP, ADP, and AMP are converted to adenosine by ecto-5′-nucleotidases. When ATP is utilized in the cell, excess adenosine is transported to the extracellular space. Extracellular adenosine concentrations have been shown to increase with increased metabolism and increased neural activity (Minor et al., 2001; Pull & Mcilwain, 1972). Under increased energy demand during wake periods, when metabolic rate is approximately 30% higher than during NREM sleep, adenosine levels increase and promote delta activity in NREM sleep (Kalinchuk et al., 2008).

Currently, there are known four different adenosine receptors, A_1, A_{2a}, A_{2b} and A_3 (Fredholm, 2010). For sleep-wake homeostasis the A_1 and A_{2a} receptors have received the most attention and have been shown to mediate adenosine's sleep inducing effects (Satoh et al., 1998; Strecker et al., 2000). The effects of adenosine in the basal forebrain on recovery sleep are mostly mediated by A_1 receptors. Perfusion of an A_1 receptor antagonist into the basal forebrain reduced recovery sleep after sleep deprivation, and no effect was observed by perfusion of an A_{2a} receptor antagonist (Gass et al., 2009).

Regarding the role of adenosine in mood regulation, there are some indirect evidences pointing at the association between adenosine and depression. From epidemiological studies, it is known that consumption of caffeine, non-selective A_1/A_{2a} adenosine receptor antagonist, modifies the mood of healthy people and psychiatric patients (Greden et al., 1978; Smith, 2009). Consumption of caffeine by healthy subjects was associated with a reduced risk of depression compared to the incidence of depression in those with no caffeine intake (Smith, 2009). In animal studies, the nonselective activation of adenosine receptors in the brain induced depressive-like symptoms, whereas administration of caffeine effectively reduced depressive-like behaviour in mice (El Yacoubi et al., 2003; Kulkarni & Mehta, 1985). Selective A_{2a} receptor antagonists also produced antidepressant activity (Yamada et al., 2014). One of the possible mechanism which could explain these effects of adenosine can be the ability of adenosine to modulate the release of neuromediators directly involved in mood regulation, such as acetylcholine, glutamate, dopamine, GABA and serotonin (Dunwiddie & Masino, 2001). Moreover, adenosine has also neuroprotective properties mediated through A_1 and A_{2a} receptors, which may be

involved during inflammatory process in depression and can normalize glutamate release (Cunha, 2005)

Apart from the neurochemical mechanisms, adenosine may affect depression through affecting sleep. Depression is characterized by decreased SWA during NREM sleep, which reflects decreased sleep intensity (Borbely, 1987). This may occur due to insufficiency of homeostatic regulation of sleep. Adenosine being one of the most important homeostatic sleep molecule in the brain (Porkka-Heiskanen et al., 1997), is known to be decreased in pharmacological animal model of depression (Savelyev et al., 2012). Thus, it might be concluded that adenosine deficiency in depression can contribute to sleep problems found often in depression, and on the other side it can aggravate the depressive state by itself.

2.5. Early life stress as a predisposing factor for depression development

2.5.1 Early life stress as a risk factor for depression

Early childhood and adolescence are extremely sensitive periods in human development, during which the brain, especially the systems controlling emotions, attention, self-control and stress, are shaped by the interaction with environment. Multiple epidemiological studies have indicated a tight association between adverse childhood experience and an increased risk to develop depression later in life (Edwards et al., 2003; Heim et al., 2008; Kessler, 1997; Kessler et al., 2010). The strong dose–response relationship has been found between different types of early life stressors, such as physical or sexual abuse, neglect or loss of the parent, and general mental health problems in adulthood. The global prevalence of child sexual abuse is quite high, and is estimated to be 11.8% overall, with higher prevalence for girls (18.0%) compared to boys (7.6%) (Stoltenborgh et al., 2011). Recent meta-analysis studies also provided statistics on the global prevalence of child physical abuse, physical neglect and emotional neglect being 22.6%, 16.3%, and 18.4%, respectively, with no apparent gender differences (Stoltenborgh et al., 2013a; Stoltenborgh et al., 2013b). An important role of childhood traumatic events

in the development of MDD has been also demonstrated in twin studies (Kendler et al., 2000; Nelson et al., 2002).

The most severe childhood adversities have significant effects on early-onset depression (defined as an onset before age 20) but not on later-onset depression (Kessler, 1997). This indicates that there is a developmental window, where childhood adverse events have the strongest impact on depression vulnerability. Furthermore, individuals with early life adverse experience are more vulnerable to stress and have lower threshold for developing a depressive disorder in response to stressors (Dougherty et al., 2004; Hammen et al., 2000; Kendler et al., 2004). Interaction between environment and genes contributes to likelihood of depression development in response to stressful events in people who experienced maltreatment during their childhood (Bradley et al., 2008; Caspi et al., 2003; Kaufman et al., 2004). There are a few genes highlighted in these studies, including the *BDNF* (Buchmann et al., 2013; Liu, 2010).

The underlying mechanism through which early life stress increase depression vulnerability has been a subject for many studies. There are alterations in brain development in children who experienced early life adversities, including reduced volume of limbic, cortical brain areas, and interhemispheric connections (Pechtel & Pizzagalli, 2011; Teicher et al., 2003; Teicher et al., 2004). It was suggested that early life stress interferes with the critical waves of neurogenesis, synaptic overproduction, and pruning of synapses and receptors, resulting in deficits of cognitive functions (Teicher et al., 2006).

Studies in rodents and non-human primates have focused on epigenetic modifications of stress reactivity as a result of early life adverse experience. These studies suggest that early maltreatment (e.g. maternal separation or low maternal care) induces epigenetic, structural, and functional changes in neural system affecting cognitive and emotional development (Meaney & Szyf, 2005; Sanchez et al., 2001).

Early life environment can shape offspring epigenome and affect stress reactivity in future life. In rodent studies, early life stress has been shown to cause adverse alterations of different genes expression profiles, including arginine vasopressin, glucocorticoid receptor, and *BDNF* gene (Schroeder et al., 2010). In rats, difference in maternal nursing led to different methylation level of hippocampal glucocorticoid receptor gene promoter (Weaver et al., 2004), which

in turn affected their stress responsiveness in adulthood. Early life separation of new-born mice from their mothers resulted in a persistent upregulation of arginine vasopressin due to hypomethylation of the arginine vasopressin gene enhancer region and led to constant release of corticotrophin-releasing hormone (CRH), inducing a sustained hyperactivity of the HPA axis (Murgatroyd et al., 2009). Early maltreatment has been also shown to affect *BDNF* gene silencing in the prefrontal cortex due to DNA methylation mechanism that lasted into adulthood and was accompanied by abusive behaviour (Roth et al., 2009).

All these changes converge in increased endocrine and autonomic reactivity to stress, anxiety-like behaviour, anhedonia, and impairment of cognitive functions. In fact, many of the neurobiological and behavioural effects of early life stress in animal models are related to similar signs and symptoms of depressive disorder in humans, including sensitization of central stress response systems and changes in epigenome (Heim & Nemeroff, 2001, 2002; Heim et al., 2008; McGowan et al., 2009).

2.5.2 Animal models of early life stress

Animal models are invaluable tools in studies on mechanisms and pathophysiology of different mental disorders, including depression (Frazer & Morilak, 2005; Fuchs & Fliugge, 2006; Kalueff et al., 2007). However, it should be noted that complex human psychiatric diseases, as depression, are impossible to model fully in rodents, especially such symptoms as e.g. low self-esteem, feelings of guilt, or suicidality. But a number of core symptoms of human depression do have an equivalent in animals (Table 1), and one of them is altered sleep structure.

Table 1. Correlates of core depression symptoms in humans with the possible analogous parameters in rodents.

Core symptoms in humans	Analogous parameter in rodents
Anhedonia	Reduced preference for sweetened solutions
Loss of motivation	Passive behaviour, low locomotor activity
Sleep disturbances, altered sleep structure	Altered sleep structure
Anxiety	Anxiety-like behaviour
Cognitive deficits	Cognitive deficits
Hypercortisolism, hyperactivity of the HPA axis	Hyperactivity of the HPA axis

Modified from (Schmidt et al., 2011).

Currently, there is no established adolescent animal model of clinical depression. It is important to understand that the developmental progression of puberty and adolescence differs between humans and traditional laboratory animals. In humans, puberty is initiated by an increased secretion of hormones from the gonads, such as oestrogens and androgens, and androgens from the adrenals (Hagenauer & Lee, 2013). Whereas puberty in humans is defined by reproductive development, adolescence is defined as the period of social, emotional and cognitive transition between childhood and adulthood. In animal studies, the term "adolescence" is traditionally used specifically to refer to research focusing on the neural and behavioural changes accompanying the transition from juvenile dependence into the relative independence of adulthood and lasting from artificial weaning until the time of attaining reproductive competence and social interaction skills.

Of the many animal models of depression in adults, the models based on early life stress are probably the most relevant to model early-onset depression in humans. Techniques based on manipulation of the mother-infant interaction in rodents during the first weeks of postnatal life, which corresponds to the third trimester of fetal development and early postnatal development in humans, are widely used for this purpose (Roque et al., 2014; Vetulani, 2013; Wilber & Wellman, 2009). Maternal separation represents the model of serious childhood incidences in humans. It reduces the amount of maternal care for the pups, thereby modelling emotional as well as physical child neglect. In this model, newborn pups are separated from their mothers during the so-called stress hyporesponsive period in rodents, which lasts during postnatal days (PNDs) 2–14 in the rat and PNDs 1–12 in the mouse and is characterized by low basal ACTH and corticosterone secretion and low corticosterone response to stressors (Levine, 1970; Pryce et al., 2005; Schmidt et al., 2003). Maintenance of this critical stress hyporesponsive period is dependent on maternal care: isolation of the pups from the dam for a single period of at least 8 h leads to an increase in ACTH and corticosterone secretion and also leads to increased HPA responsiveness to discrete stressors (Pryce et al., 2005).

Maternal separation in the models of early life stress can be carried out for different intervals of time, ranging from single episode of 24 h deprivation to repeated shorter periods of maternal deprivation of 3, 6, 8, or 12 h. The

behavioural and biological consequences of early life maternal separation in adult animals resemble those of human depression (Roque et al., 2014). This model inflicts considerable stress for both the pups and their mothers associated with alterations in behavioural, immunological, and endocrine parameters, including hyperactivation of HPA axis and increased corticosterone levels (Liu et al., 1997; Plotsky & Meaney, 1993; Roque et al., 2014).

Regarding the effect of maternal separation on sleep, several studies have studied spontaneous and stress-induced sleep in adulthood (Mrdalj et al., 2013; Reyes Prieto et al., 2012; Sampath et al., 2014; Tiba et al., 2003; Tiba et al., 2004, 2008). The core of the reported is following: the majority of the changes manifest as increases in different parameters of REM sleep; one study reported decrease of REM sleep (Reyes Prieto et al., 2012). Some studies also reported changes in NREM sleep (Mrdalj et al., 2013; Reyes Prieto et al., 2012; Sampath et al., 2014).

Taking into account that maternal separation is associated with a profound stress for both infants and mothers, it may not be an ideal representation of events in human real life. In this regard other modifications of the mother-infant interaction, which may better simulate the early life stress in childhood, can be of interest. One such approach is the model based on changing pups between mothers, namely cross-fostering (CF) (Barbazanges et al., 1996; Meaney & Szyf, 2005). The pup litters are changed between mothers during the critical postnatal period, which is known to be sensitive for environmental stimulation (at the age of PND 2-12) (Barbazanges et al., 1996). The age at which the CF procedure is performed has a great effect on the outcome (Barbazanges et al., 1996; Darnaudery et al., 2004). Thus, an early adoption, when pups are changed at PND 1 increases maternal behaviour and as a result improves cognitive performance and prevents prenatal stress-induced impairments in glucocorticoid feedback in offspring (Barros et al., 2004; Maccari et al., 1995). Pups that are raised by foster mothers, which adopted them later (from PND 5 on), have higher stress reactivity and impaired cognitive functioning (Barbazanges et al., 1996). Therefore, the latter procedure can be used as an alternative to maternal separation, but there is little or no data regarding its link to depression-like phenotype.

3. Aims and hypotheses of the study

3.1. General aim and hypothesis

Our main hypothesis was that early life stress leaves persistent molecular changes in the brain, predisposing to depression and sleep problems development in future.

The aim of this thesis was to follow the development of depression and concomitant sleep disturbances starting from early life, and study their molecular mechanisms.

3.2. Specific aims and hypotheses

The specific hypotheses of the study were:

1) *BDNF* gene expression has an exon-specific transcription pattern during spontaneous sleep and is affected by sleep deprivation through the transcription-silencing 5-methylcytosine DNA modification.

2) Cross-fostering induces depressive-like behaviour accompanied by sleep changes and decreased *BDNF* gene expression in involved brain regions.

3) Adolescent depression is characterized by sleep disturbances and slow-wave activity abnormalities.

4) Genetically determined variation in *BDNF* gene (Val66Met) modulates sleep, psychomotor development and temperament that can be assessed already at early stages of infancy.

The specific aims of the study were:

1) To study the expression dynamics of three activity-dependent *BDNF* transcripts (*BDNF*1, *BDNF* 4 and *BDNF* 9a) and methylation levels of their promoters (p1, p4 and p9) in spontaneous sleep and after sleep deprivation.

2) To validate cross-fostering animal model as a model of early life stress reflecting traumatic early life events happening in real human life.

3) To explore sleep structure and slow-wave activity of early-onset depression in adolescents.

4) To find out whether functional *BDNF* gene polymorphism (Val66Met), which leads to reduced BDNF secretion, is associated with sleep consolidation, psychomotor development and temperament at early stages of human infancy.

4. Materials and methods

The study outline is presented in the table 2. Animal models were used to study the propagation of depression from early life period to adulthood together with concomitant sleep problems and possible underlying molecular mechanisms (I, II). Sleep disturbances in early-onset depression were studied in the clinical sample of depressed adolescent boys (III). The association between *BDNF* gene polymorphism (Val66Met) and early life development, including sleep was studied in the birth cohort (IV).

Table 2. The study outline showing the original studies by Roman numerals.

Period	Early life		Adolescence		Adulthood	
Studied parameters	Animal	Human (IV)	Animal (II)	Human (III)	Animal (I,II)	Human
Sleep		x		x	x	
BDNF		x	x		x	
Adenosine					x	
Behaviour		x	x	x	x	

The topics addressed in the original studies (indicated by the roman numerals) are marked with the crosses. Animal studies I and II are focused on possible molecular mechanisms of interaction between depression and sleep disorders. The focus of clinical study III is sleep abnormalities in adolescent depression. Study IV is aimed at the investigation of association between *BDNF* Val66Met polymorphism and early life development.

4.1. Animal studies

4.1.1 Role of exon-specific *BDNF* expression and methylation at different promoters in sleep regulation

4.1.1.1. Rats and housing procedures

Young male Wistar rats (12 weeks old, 300-400g, received from formerly Harlan Laboratories B.V., now Envigo, Venray, Netherlands) were housed at

constant temperature (23±1°C) and humidity (50% - 60%) under 12h light/12h dark cycle (light on at 08:00 h) with *ad libitum* access to food and water. Animals were handled starting at least 5 days before the experiment. All animal procedures were performed according to the guidelines of the National Institute of Health Guide for the Care and Use of Laboratory Animals, and were approved by the experimental Animal Ethics Committee of Southern Finland (animal license ESAVI/5028/04.10.07/2014).

4.1.1.2. Sleep deprivation protocol

Thirty six rats were divided into 6 groups: three controls and three SD (Figure 4). SD was performed from the beginning of the light period (starting at 8 am, or *zeitgeber* time [ZT0]) for 3 h (finished at 11 am, or ZT3), 6 h (finished at 1 pm, or ZT6) or 12 h (finished at 8 pm, or ZT12). Animals were sleep deprived using the gentle handling method (Franken et al., 1991a). Briefly, SD was enforced by exposing the rats to novel objects to engage them in exploratory activity. In case of 12 h SD, closer to the end of sleep deprivation period animals were subjected to external stimulation, such as mild noises, tapping the cage, or direct contact with the animal through a soft brush. Control animals were left undisturbed in a separate room.

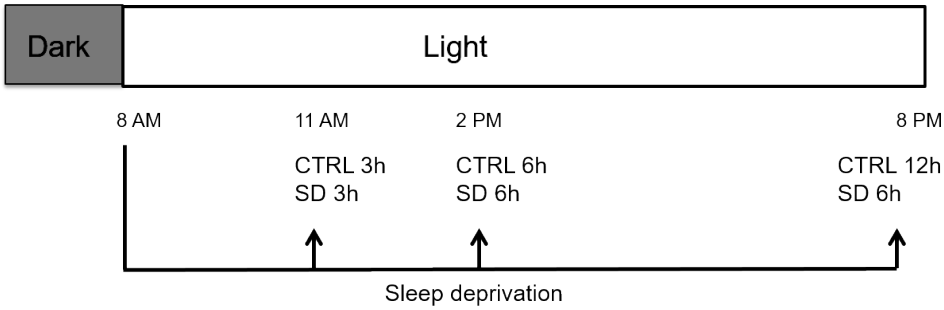

Figure 4. Experimental protocol: non-deprived groups (CTRL) 3, 6 or 12 h and sleep-deprived (SD) groups 3, 6 or 12 h (N=6 animals per each group). Modified from Study I.

4.1.1.3. Brain tissue collection

Immediately after SD, rats from both groups, SD and CTRL, were killed by carbon dioxide followed by decapitation and the brains were quickly removed. Two

mm coronal slices of the target areas were prepared. The basal forebrain and frontal cortex were dissected from the slices with a scalpel. The tissues were immediately placed into RNA-stabilizing reagent RNAlater (Sigma-Aldrich, Helsinki, Finland), frozen on dry ice and stored at -80 °C.

4.1.1.4. BDNF messenger RNA analysis

After brains homogenization total RNA was immediately extracted using the RNeasy Plus Mini Kit (Qiagen, Helsinki, Finland), according to the manufacturer's instructions. One µg of total RNA was reverse-transcribed using a Maxima first standard cDNA synthesis kit (Thermo Scientific, Helsinki, Finland). The cDNA regions marked on figure 5 were amplified with a Maxima SYBR Green qPCR Master Mix (Thermo Scientific) and selected primers (details in the Study I). The amplification was done using a LightCycler 480 real-time polymerase chain reaction (PCR) system (Roche, Espoo, Finland). Relative quantification of template was performed using the ΔΔCt method (Livak & Schmittgen, 2001), with experimental cDNA data normalized to a housekeeping gene level (glyceraldehyde-3-phosphate dehydrogenase, *GAPDH*).

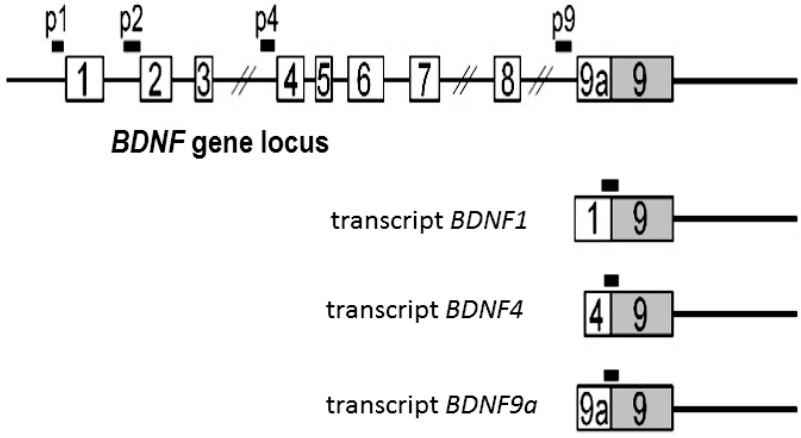

Figure 5. Each 5' exon is controlled by its own promoter and alternatively spliced to the protein coding exon 9 (marked with grey box), resulting in different *BDNF* transcripts (here shown *BDNF1*, *BDNF4* and *BDNF9a*) coding for the same protein. Genomic regions p1, p2, p4, p9 (marked with black rectangles) were amplified in the methylated DNA immunoprecipitation (MeDIP) experiment; 1–9, 4–9 and 9a–9 exon junction regions of *BDNF* transcripts were amplified in the mRNA expression experiment. Modified from Study I.

4.1.1.5. Methylated DNA immunoprecipitation

Methylated DNA immunoprecipitation (MeDIP) is an efficient technique of detection of methylated DNA using the anti-5-methylcytosine (5mC) antibodies (Thu et al., 2009). In brief, genomic DNA is randomly sheared by sonication and immunoprecipiteted (IP) with a monoclonal antibody that specifically recognizes 5-mC. The DNA amount in the 5mC IP fraction reflects the amount of methylated CpG dinucleotides on genomic DNA. The resulting enrichment of methylated DNA in the IP fraction can be determined by PCR to assess the methylation state of individual regions.

GeneJET Genomic DNA Purification Kit (Thermo Scientific) was used for purification of genomic DNA from the same rat's brain tissues that were used for RNA purification. Genomic DNA (1.2 µg) in 100 uL Tris/ethylenediamine tetraacetic acid (EDTA; TE; 10 mM Tris pH 8.0, 1 mM EDTA) was sheared randomly by sonication to the 200–500 base pairs size using a Vibra-Cell Ultrasonic Liquid Processor (Sonics & Materials Inc., Newtown, CT, USA), and then heat-denatured. One part (0.2 µg) of sheared DNA (input) was stored at +4 °C during the IP reaction, while the remaining five parts (1 µg, IP sample) was used for the IP reaction. The IP sample was incubated with 1 µg of the anti-5mC monoclonal antibody (clone 33D3; Diagenode, Liege, Belgium) overnight at +4 °C and precipitated with the protein G Dynabeads (Invitrogen, Helsinki, Finland) for 2 h at +4°C. Proteinase K was used for digestion of immunoprecipitated DNA–antibody complex for 3 h at +56 °C. After this the released DNA was column-purified using the GeneJET DNA Cleanup Micro Kit (Thermo Scientific).

Final input and IP DNA samples were diluted in 100 uL TE and kept at -20 °C. The resulting enrichment of methylated DNA in the IP versus the input fraction was quantified by PCR using a LightCycler 480 real-time PCR system (Roche) with a Maxima SYBR Green qPCR Master Mix (Thermo Scientific) and the primers for *BDNF* promoters p1, p4 and p9, and the promoter p2, less regulated by neuronal activity or SD (Figure 5). Relative quantification of each IP template was normalized to its corresponding input level and multiplied by 5 to equalize the original amount of the IP and input genomic DNA samples.

Data followed a normal distribution and the main effects of sleep deprivation or circadian time (SD or ZT) factors were analysed by using two-way analysis of

variance (ANOVA). Effect of ZT within the same treatment group (CTRL or SD) was analysed by one-way ANOVA followed by Fisher's post-hoc test (protected least significant difference, PLSD). The CTRL and respective SD groups on the same ZT-point (3, 6 or 12 h) were compared using a Student's unpaired two-tailed t-test. Cohen's d was used to estimate the effect sizes as follows: small ≥ 0.2, medium ≥ 0.5, large ≥ 0.8. The group differences showed medium effect sizes. The linear regression ANOVA analyses were performed using a StatView statistical package (Study I).

4.1.2 Role of early life stress in depression development and sleep disturbances

4.1.2.1. Rats, cross fostering model and housing procedures

Modification of the mother–infant interaction during early postnatal period, namely CF, has specific effects on the development of offspring characterized by higher stress reactivity and impaired cognitive functioning (Barbazanges et al., 1996). Different approaches have been used to model CF but they all based on the assumption that the offspring development is programmed by the prenatal and/or postnatal maternal environment (McCarty, 2017).

In our study, pup litters of Hannover Wistar rats (average size 10 pups) were changed between mothers at the end of stress hyporesponsive period, on PND12 (Barbazanges et al., 1996). The mothers were carefully removed from their home cage and temporarily placed in empty cages containing some nesting material and food pellets. Meanwhile, the litters were placed in artificial nests made from soft nesting material placed on top of two rubber gloves filled with warm water. The bedding in home cages was quickly changed. The whole procedure took no longer than 5 min. The litters were placed in the cage of their foster mothers, and the mothers were returned to their home cages. After some reorganization of the pups and the nest, the dams calmed down and began to lactate. The group of mothers with litters, which served as a control (CTRL) group was left undisturbed. Since it has been previously shown (Barbazanges et al., 1996) that the short separation by itself does not lead to behavioural or hormonal changes, we did not have additional control group for checking any possible effects of a single brief separation.

Eight CTRL and 9 CF nests were included in this study. In total, 43 male and 41 female rats were used for this study, which were kept at the same conditions as were described above in the section 3.1.1.1. In adulthood, one male and one female sibling per litter was used for each measurement. The timeline of the experimental procedures is represented in figure 6.

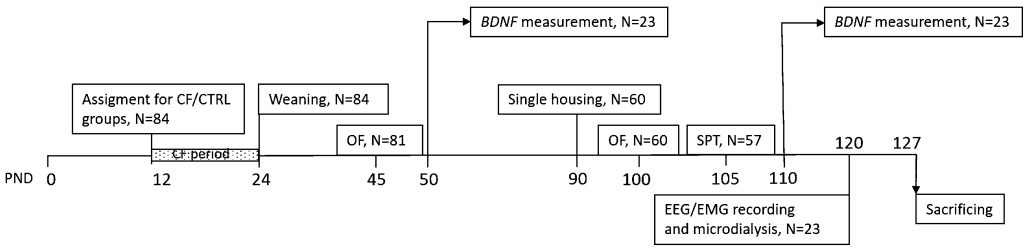

Figure 6. The timeline of the experimental procedures. PND: postnatal day, OF: open field test, SPT: sucrose preference test, CTRL: control rats, CF: cross-fostering rats. Modified from Study II.

4.1.2.2. Behavioural tests

Sucrose preference test. Sucrose preference test (SPT), is widely used for assessing the anhedonia-like phenotype (lack of interest in rewarding stimuli) in laboratory rodents. It has been shown, that the chronic mild stress procedures induce a substantial reduction in consumption of sucrose solutions, which can be interpreted as a decreased sensitivity to reward in stressed animals (Katz, 1982). Preference is measured by volume and/or weight of sweet liquid consumed during testing period, which is then converted to a percent preference compared to a water only.

SPT was performed in individually housed rats once during their adulthood (PND 105). The test was performed in two consecutive sessions. During the training session, which lasted for 48 h, animals were provided simultaneously with 2 bottles in the home cage, one containing a 1% sucrose solution, the other containing tap water. The position of the bottles was switched twice a day to avoid place preference. The volume of tap and sucrose water intake was measured every 24 h. After the training session, animals were deprived of food and water for 18 h with following test session, which lasted for 2 h. The amount of water remaining in each bottle was measured at the end of the testing period. Food and water deprivation

before test session was performed based on previous research showing that it is a necessary component of successful SPT in chronically stressed animals (Katz, 1982; Remus et al., 2015). Overnight food and water deprivation motivates rodents to consume liquid during a limited amount of time during the light cycle where their liquid intake is typically low. On the other hand, food and water deprivation prior to SPT may by itself act as a stressor. This facilitates the anhedonic response in chronically stressed rats, which is associated with such physiological changes as general hypoglycaemia, increase in hippocampal pro-inflammatory cytokine level, and elevated release of cortical dopamine and norepinephrine compared to control rats (Remus et al., 2015). Thus, rodents with depressive-like behaviour may have differential responses to food and water deprivation that results in changes in sucrose intake compared to control animals.

The sucrose preference score was expressed as percent of total fluid intake (sucrose preference = sucrose water consumed/total liquid consumed × 100%) (Bhagya et al., 2008). Factorial ANOVA (factors: sex, treatment) was used to evaluate the effect of these factors on sucrose preference (Study II).

Open field test. The symptoms of anxiety are highly prevalent in depression disorder. In animal models anxiety-like behaviour can be assessed by the open field (OF) test. This test provides an opportunity to assess novel environment exploration, general locomotor activity, and initial screen for anxiety-related behaviour (Prut & Belzung, 2003). Although OF test is commonly used for the assessment of anxiety-like behaviour, it is known that the activity in OF is also affected by depression and chronic stress (Kalueff & Tuohimaa, 2004). Animals with depressive-like behaviour demonstrate reduced basal activity in OF and avoidance of open area, representing a "refractory loss of interest" (Katz et al., 1981).

The OF test apparatus consisted of a white circular arena (diameter 80 cm), enclosed by white walls. The arena was divided into 19 equal sectors by lines drawn on the bottom. The apparatus was placed in a quiet room. Rats were first weighed and then placed in the center of the OF arena. The activity of each rat during the test session (5 min) was video recorded. Between tests the apparatus was cleaned with tap water followed by ethanol and then wiped until dry.

The videos were analyzed as follows. Horizontal (number of squares crossed in the inner and outer areas, latency to leave centre) and vertical (rearings [standing on

hind legs]) activity, grooming (latency to start self-grooming, duration and number of self-grooming episodes), and excretory activity (defecations and urinations number) were scored. Factorial ANOVA (factors: sex, age, and treatment) was used to analyze the effect of these factors and their interaction on weight of the animals and their activity in OF test (Study II).

Rats from CF and CTRL groups were subjected to OF test during 2 time points: adolescence (PND 45) and adulthood (PND 100). Half of the adolescent animals were sacrificed a few days after the test for brain tissue collection (for *BDNF* expression measurements) and another half were kept alive until they reached adulthood.

4.1.2.3. Brain tissue collection

Rats were sacrificed by decapitation using a guillotine, and the brains were quickly removed. Two mm coronal slices of the target areas were prepared. The basal forebrain, frontal cortex, and hypothalamus were dissected from the slices with a scalpel. The tissues were placed into RNA-stabilizing reagent RNA later (Sigma-Aldrich, Helsinki, Finland), frozen on dry ice, and stored at –80°C.

4.1.2.4. BDNF messenger RNA analysis

The brain tissues were homogenized and RNA was immediately extracted using RNA Later Lipid Mini Tissue Kit according to the manufacturer's protocol (Qiagen, Helsinki, Finland). DNase treatment of the RNA fraction was carried out using the RNAse-Free DNase Set (Qiagen, Helsinki, Finland) as a precaution against genomic DNA contamination. The measurement of RNA fraction for purity and concentration was carried out with Nanodrop ND-1000 spectrophotometer (NanoDrop Technologies, Rockland, DE, USA).

TaqMan RT-qPCR was performed using LightCycler 480 Real-Time PCR System (Roche, Espoo, Finland) and TaqMan gene expression assays (Applied Biosystems, Foster City, CA, USA). One µg of total RNA was reverse transcribed using a Maxima first standard cDNA synthesis kit (Thermo Scientific, Helsinki, Finland). cDNA samples were amplified using a TaqMan Universal PCR MasterMix and the commercial gene expression assays with probes for *BDNF* (Rn02531967_s1) and *ACTB* (Rn00667869_m1) from Applied Biosystems.

The PCR program used for the experiment was as follows: 50°C 2 min - 95°C 1 min and 50 cycles of repeating 95°C for 15 seconds and 60°C for 1 min. All samples were run in triplicates. Relative quantification of template was performed using the ΔΔCt method. Experimental cDNA data was normalized to a housekeeping gene (actin beta, *ACTB)* level. Factorial ANOVA (factors: sex, age, and treatment) was used to analyze the effect of these factors and their interaction on *BDNF* gene expression in studied brain regions (Study II).

4.1.2.5. Surgery, *in vivo* microdialysis and polysomnographic recordings and analysis

Under general anesthesia, 14 male and 14 female adult rats from CTRL and CF groups, were implanted with electrodes for the recording of the EEG/EMG and with a unilateral guide cannula for the microdialysis probe.

The rats were anesthetized with isoflurane (IsoFlo Vet 100%, Abbott Laboratories Ltd, England) (5% induction; 2% maintenance) and placed in a stereotaxic device. Fifteen minutes before starting the surgery, rats were injected with buprenorphine (Temgesic, Indivior UK Limited, Slough, UK, 0.05 mg/kg, s.c.). After exposing, cleaning and disinfecting the skull bone, two bipolar screw EEG electrodes were placed fronto-parietally into the skull: 2 mm rostral, 2 mm lateral from bregma, and 4 mm rostral, 1 mm lateral from lambda. While two silver wire electrodes were inserted into the neck muscles for EMG recording. A unilateral guide cannula (CMA 11 Guide, CMA/ Microdialysis, Stockholm, Sweden) was implanted aiming at the basal forebrain (BF) cholinergic area (horizontal diagonal band of Broca, the substantia innominata and the magnocellular preoptic area; the coordinates respective to bregma: anterior = –0.3 mm; lateral = 2.0 mm; vertical = –5.5 mm). The schematic representation of coronal brain section with targeted area is represented in figure 7. Electrodes and guide cannula were fixed to the skull with acrylic dental cement.

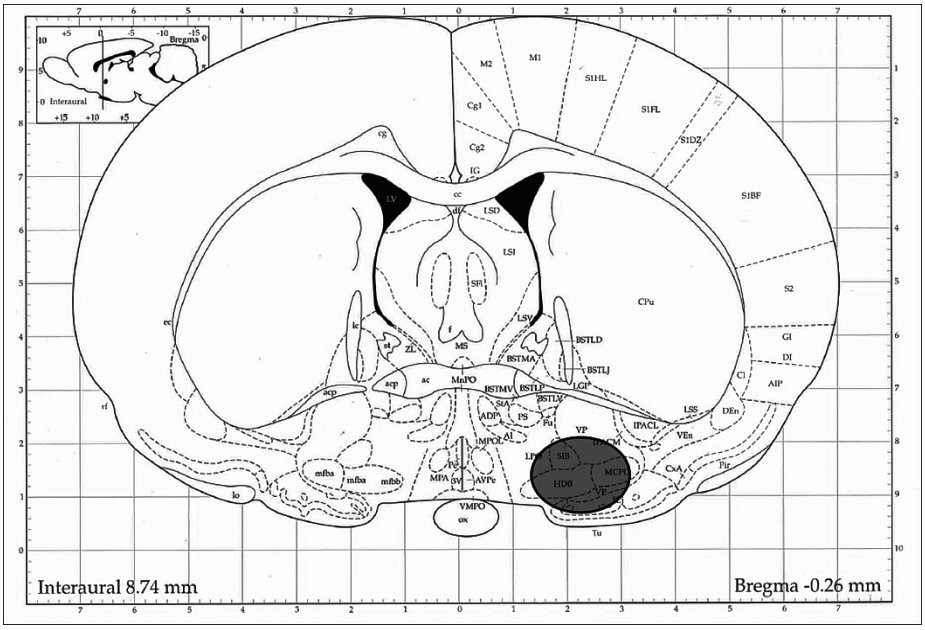

Figure 7. The schematic representation of coronal brain section with targeted BF area marked with grey circle. Modified from (Paxinos & Watson, 1998).

After surgery, rats were single-housed in Plexiglas boxes; the day after the surgery they were injected with antibiotic/analgesic (1.6 g/kg A-Pen, Orion Pharma Oy, Espoo, Finland + 6 mg/kg lidocaine, Orion Pharma Oy, Espoo, Finland, i.p.). Then they were connected to recording cables through swivels, and allowed to recover up to 1 week; a 2 days recording was taken from each rat to estimate their sleep-wake cycle. The experiments were not started until their behaviour and sleep–wake cycle had normalized.

A microdialysis probe (CMA11 Microdialysis probe; 14 mm shaft length, 2 mm dialyzing membrane length, 0.24 mm diameter; CMA/Microdialysis) was inserted through the guide cannula into the basal forebrain and inlet and outlet tubing (1.5 m pieces of fluorinated ethylene propylene tubing; CMA/Microdialysis) connected to the probe the day before the beginning of the experiment.

Half an hour prior to the onset of sample collection, at 09:30 AM, microdialysis tubing was connected to the pump (model R99-EMZ, Razel Scientific Instruments, Vermont, USA) and the fraction collector (Eicom EFC-82 Fraction Collector, San Diego, CA, USA). After that continuous perfusion of the microdialysis probe with

artificial cerebrospinal fluid (aCSF) (147 mM NaCl, 3 mM KCl, 1.2 mM $CaCl_2$, 1 mM $MgCl_2$) was performed at a rate of 1 µL/min throughout the light period of baseline (BL) day (9:30 AM-8:30 PM). The samples (30 µL each) were collected at 30-min intervals and then stored at –80 °C until assayed.

After finishing the experiments, the animals were sacrificed by administrating a lethal dose of pentobarbital (100 mg/kg Mebunat, i.p., Orion Pharma Oy, Espoo, Finland). The verification of the probe location was done by an ink injection through a modified microdialysis probe inserted into the guide cannula. After that the brains were removed, frozen on dry ice, and stored at –80°C. Twenty micrometer coronal sections were cut on a freezing microtome, stained with Toluidine Blue (Sigma-Aldrich), dehydrated with ethanol, and dried. The probe locations were visually inspected under the light microscope. Only animals with probe tips located in the BF cholinergic area were included in the analysis (n = 7 CTRL and 6 CF males; n = 5 CTRL and 5 CF females).

EEG/ EMG recordings were performed during the microdialysis experiments. EEG/ EMG signals were sampled at 271 Hz, amplified (gain 5000) and analogically filtered (high pass: 0.3 Hz; low pass 100 Hz) using the Spike2 program (version 5.19; Cambridge Electronic Devices Ltd., Cambridge, UK). The recordings were semi-automatically scored off-line at 4-s epochs for NREM, REM sleep, and wakefulness using Autoscore-1.7 script and then manually checked using Sleepscore v1.01 script (Cambridge Electronic Design) according to the common criteria (Zant et al., 2012).

The EEG power spectra (in square microvolts) were calculated with Spike2 separately for consecutive 4-s NREM epochs (fast Fourier transform (FFT) routine 512; Hanning window, 0.54 Hz resolution) within a frequency range of 1.08-29.84 Hz and averaged over the time period that was investigated. The power spectra in NREM sleep for delta range (1.08-3.79 Hz) in recovery sleep during one hour of post-sleep deprivation period (4:00 PM – 5:00 PM) and respective period of BL day were normalized to the total NREM power of SD and BL days, respectively. Factorial repeated measures ANOVA (between-subject factors: sex and treatment, within-subject factor: SD) was used to analyze the effect of these factors and their interaction on NREM sleep delta power (Study II).

4.1.2.6. Experimental days

The baseline day, when animals were left undisturbed, served as a reference to which subsequent EEG recordings obtained during the SD day were compared to. During the BL day rats were connected to microdialysis tubings during 30 min after lights-on and perfusion with aCSF was started at 09:30 AM. Perfusion was stopped 30 min before lights-off (i.e., 08:30 PM).

Sleep deprivation day was following the BL day. SD lasting for 3h was performed by gentle handling as was described in the section 3.1.1.2. (Franken et al., 1991a). Briefly, SD was enforced by exposing the rats to novel objects. Every new object was introduced into the cages just following the first signs of slow waves in the EEG recordings. SD was started 4 h after lights-on (at 01:00 PM) and lasted until 4:00 PM. After finishing SD the animals were left undisturbed. The post-SD recovery sleep during remaining light period (4:00 PM to 09:00 PM) was compared to the corresponding hours of BL day.

Analysis of vigilance state-related parameters (wake, NREM, REM sleep duration, and number of REM sleep onsets) was performed by using factorial ANOVA (factors: sex, treatment) for spontaneous sleep (separately for the light and the dark periods of the day). Recovery sleep was analyzed using factorial repeated measures ANOVA (between-subject factors: sex and treatment, within-subject factor: SD) during the remaining light period after ending of SD (04:00 PM to 09:00 PM) separately for NREM and REM sleep.

4.1.2.7. Microdialysis adenosine analysis

All samples collected during the microdialysis experiments were used for measuring adenosine using a high-performance liquid chromatography (HPLC) assay as described in (Savelyev et al., 2012).

The mobile phase, consisting of ammonium acetate 50 mM, tetrabutyl-ammonium hydrogen sulphate 0.2 mM, EDTA 1 mM in 15% of MeOH, pH 5.6, was passed through the column at a flow rate of 1 ml/min. Ten microliters of the first 30 min sample of every hour from BL and SD days were injected through a Kinetex ® C18 column (100A, 150 × 4.60 mm, particle size: 5 micron, Phenomenex, USA), coupled with a pre-column (KJO-4282, Phenomenex, USA). Adenosine was detected at a wavelength of 460 nm and its concentrations were determined by

comparing sample peak areas with those of standards (AMP, adenosine, cAMP, cdAMP, Sigma-Aldrich) using LabSolutions software (Shimadzu). Detection limit: 0.1 nM; linear range of the assay: 0.1 nM - 50 nM; coefficient of variation < 10%.

The basal adenosine level was determined as the average adenosine concentration from the samples collected during the baseline day (09:30 AM–08:30 PM). Analysis of adenosine level in BF was performed for spontaneous sleep during the light period. Factorial ANOVA (factors: sex, treatment) was used to reveal the effect of sex and CF treatment on adenosine level in BF (Study II).

4.2. Human studies

4.2.1 Role of *BDNF* Val66Met polymorphism in early life sleep, temperament and psychomotor development

4.2.1.1. Study cohort

The Finnish CHILD-SLEEP birth cohort was used in this study. Data were collected at Tampere University Hospital (TAYS) during the period from April 2011 to January 2013 (for details, see (Juulia Paavonen et al., 2017)). Altogether 1678 families participated in the study. The umbilical cord blood samples for genetic analyses were collected from 1501 newborns (from 89.5% of the eligible families), with the genotype data available from 1406 individuals. We excluded infants with missing phenotype data (232 infants). Since it is known that compromised birth weight and low 5-minute Apgar scores are predictors for developmental abnormalities throughout childhood and later in life (Gampel & Nomura, 2014) the infants with low or high birth weight (2500 g < weight < 4500 g; 51 infants) and with low Apgar score at 5 min (<7; 4 infants) were also excluded from the analysis. The cohort used for the analysis consisted of 1119 infants at the age of 3 months (79.6% of the genotyped sample) and 994 infants at the age of 8 months (70.7%).

The ethics approval for CHILD-SLEEP was obtained from the Ethics Committee of Pirkanmaa Hospital District (R11032/9.3.2011).

4.2.1.2. Sleep assessment

Sleep quality of infants at both ages (3 and 8 months) was assessed by the Brief Infant Sleep Questionnaire (BISQ) completed by the parents. The questionnaire variables that were chosen for further analysis included: 1) night time sleep duration (between 7:00 PM and 7:00 AM); 2) daytime sleep duration (between 7:00 AM and 7:00 PM); 3) total sleep time (TST; calculated as sum of night and daytime duration); 4) number of night awakenings. To assess the development of sleep consolidation, a proportion of night sleep in relation to TST (night sleep/TST) was calculated. All parameters were correlated to the *BDNF* Val66Met polymorphism.

In order to collect additional information about the sleep quality of infants, a subsample (N=88) of infants underwent ambulatory home polysomnographies at the age of 1, 8 and 24 months as described in (Satomaa et al., 2016). Briefly, the ambulatory polysomnographic recordings were obtained using the Embla Titanium system. EEG (6 channels: F4-A1, C4-A1, O2-A1, F3-A2, C3-A2, O1-A2), EOG (right and left), and chin EMG were recorded. The polysomnographic recordings were scored into the sleep stages in 30-s epochs using Somnologica Studio 5.0 software by two independent, experienced clinical neurophysiologists. The polysomnographic data for 57 (25 boys and 32 girls) infants was available at the age of 8 months and was used in this study. Three polysomnographic parameters were taken into analysis for this study: ratio between REM and NREM sleep (REM/NREM ratio, as a criteria of sleep pattern maturation), sleep efficiency index (SEI; calculated as sum of all NREM stages and REM sleep, divided by the total time in bed and multiplied by 100) and wakefulness time after sleep onset (WASO; reflecting sleep fragmentation).

4.2.1.3. Psychomotor development assessment

Vocalization, socioemotional, fine and gross motor development were assessed at the age of 3 and 8 month by using the modified version of the brief parent questionnaire, which was developed for children aged from 1 to 12 months (Lyytinen, 2000; Nieminen, 2004). The questionnaire consisted of 4 scales: i) vocalization (10 items); ii) socioemotional development (13 items); iii) fine motor development (15 items); iv) gross motor development (12 items). The scales were scored on a three point scale (0 = not observed, 1 = observed once, 2 = observed

many times) about the frequency of certain behaviour. The sum of each scale was used for the analysis.

4.2.1.4. Temperament measurements

Temperament, defined as the constitutional traits that drive individual differences in reactivity and self-regulation, is an important predictor of social, intellectual, and behavioural development (Gartstein & Rothbart, 2003). The temperament was assessed at the age of 8 months by using the short version of revised Infant Behaviour Questionnaire (IBQ-R 91 items; 14 scales). The IBQ-R measures infant temperament scored on a seven point Likert scale (1 = never, 7 = always), about the frequency of certain behaviours in specific situations (playing, bathing, etc.) of the previous week. The means were used for the analysis. The IBQ-R consists of 14 scales, which in turn compose three overarching factors: positive and negative affectivity, and orienting/regulatory capacity. Positive affectivity indicates an infant's tendency for energetic activity, positive affect, and high intensity pleasure, and includes the following scales: Smiling and Laughter, High Intensity Pleasure, Activity Level, Approach, Perceptual Sensitivity, and Vocal Reactivity. Negative affectivity reflects an infant's constitutional tendency towards negative emotions and distress, and includes such scales as Fear, Distress to Limitations, Sadness, and Falling Reactivity (negatively loading). Orienting/ regulatory capacity dimension relates to an infant style of affective regulation including greater impulsivity, fearfulness, and orienting, and it includes Duration of Orienting, Soothability, Cuddliness/Affiliation, and Low Intensity Pleasure scales (Gartstein & Rothbart, 2003). These three dimensions of IBQ-R are known to be analogous to the adults' personality traits of extraversion, neuroticism and conscientiousness, respectively (Putnam et al., 2014). Adequate reliability and validity of this parent-report instrument have been previously documented (Gartstein & Rothbart, 2003; Parade & Leerkes, 2008). In our study these three main dimensions were investigated in relation to the *BDNF* Val66Met polymorphism.

4.2.1.5. Genotyping

Umbilical cord blood samples for genetic analyses were collected from newborns at birth. DNA extraction were done using standard methods at the

National Institute for Health and Welfare. Genotyping of informative SNPs including functional *BDNF* SNP (rs6265, or Val66Met) was done with Illumina Infinium PsychArray BeadChip.

4.2.1.6. Statistical analysis

All statistical analyses were performed with SPSS v.22 (IBM Corp., Armonk, NY, USA). The data came from a normal distribution. First, the effect of gender on studied traits was revealed by one-way ANOVA. Then testing of association between *BDNF* Val66Met variation and different characteristics of infant development was done by one-way ANOVA in case when gender did not have a significant effect, or two-way ANOVA in case when the previous analyses had shown a differences between boys and girls at $P<0.05$ (not corrected for multiple testing). Analyses were performed with the *BDNF* SNP allele dosage (under additive model, three levels: Val66Val, Val66Met and Met66Met) and gender as the independent factors and traits of interest as the dependent variables. The analyses were performed separately at the age of 3 and 8 months.

Both P-values, not corrected for multiple testing (P) as well as corrected values (by multiplying observed P- value on number of comparisons (P_{Bonf}), 9 comparisons at the age of 3 months and 16 at the age of 8 months), are shown in the results' tables. A probability level of $P_{Bonf}<0.05$ was considered to indicate statistical significance for the genetic analyses.

Power calculations were performed with the Genetic Power Calculator (http://pngu.mgh.harvard.edu/~purcell/gpc/) (Purcell et al., 2003) assuming an additive model, 1 % quantitative trait locus (QTL) variance, 5% MAF and perfect linkage disequilibrium between QTL and the marker.

4.2.2 Sleep disturbances in depressed adolescents

4.2.2.1. Participants

A total of 20 non-medicated adolescent boys (age: 15.9±0.8 y) participated in the study. Ten of them were patients suffering from depressive and/or sleep disorder symptoms recruited from the Helsinki University Central Hospital Department of Adolescent Psychiatry outpatient units, and ten of them were healthy controls

recruited via advertisements in a newspaper for hospital staff. Exclusion criteria for all participants consisted of mental retardation, insufficient knowledge of Finnish language, current use of medication, chronic somatic illness, substance abuse/dependence, principal Diagnostic and Statistical Manual of Mental Disorders (DSM-IV) diagnosis other than depressive/sleep disorder.

All subjects underwent detailed clinical and psychiatric evaluation. All adolescents were medicine-free during the study, and the presence of somatic conditions and structural brain pathologies were ruled out based on brain MRI and blood samples. One patient was excluded from the analyses because he was diagnosed with a circadian rhythm sleep disorder only. Nine patients were diagnosed with depressive disorder according to the DSM-IV.

Diagnostic assessment was performed with the Schedule for Affective Disorders and Schizophrenia for School-Age Children—Present and Lifetime version (K-SADS-PL) (Kaufman et al., 1997). Depression symptom severity was further evaluated with two different scales: the self-administered, 21-item Beck Depression Inventory (BDI-21) (Beck et al., 1961), and the Hamilton Depression Rating Scale (HDRS) administered by a clinician (Hamilton, 1960). Insomnia symptoms were assessed by Athens Insomnia Scale (AIS) (Soldatos et al., 2000). Polysomnographic data were not available for one patient due to drop out, leaving 8 patients and 10 control subjects in the analyses of the current study.

Written informed consent for study participation was received both from the participants and their parents or legal guardians. The study protocol conformed to the Declaration of Helsinki and was approved by the ethics committee of the Helsinki University Central Hospital.

4.2.2.2. Blood sampling and testosterone measurement

Serum testosterone levels were measured from peripheral venous blood samples taken in the morning according to standard laboratory procedure at the Helsinki University Central Hospital Laboratory (HUSLAB). Testosterone was measured according to accredited laboratory methods (Liquid chromatography-tandem mass spectrometry). The blood samples were taken after the clinical interview with varying interval (average time between interview and blood sampling for patients 11 days, for controls - 39 days).

4.2.2.3. Polysomnographic recordings and sleep scoring

Polysomnographic recordings were conducted in subjects' home environment with ambulatory recording devices (Embla, Flaga Hf. Medical devices; EEG positions according to the International 10-20 system; derivations F4-M1, C4-M1, O2-M1 and backup derivations F3-M2, C3-M2, and O1-M2; sampling rate 200Hz) for two consecutive nights. EEG, EMG and EOG were recorded according to the standard criteria and the whole recording period was manually scored for sleep stages in 30 s epochs by a certified sleep technician blinded to the patient/control status of the subjects. First recording night served as an adaptation night and second night has been used for the sleep and power spectral analyses.

The following sleep parameters were calculated from the scored data: TST, sleep efficiency (time asleep relative to sleep period, which was calculated as time from sleep onset through last epoch of sleep), SWS and REM sleep latencies (time from sleep onset until first SWS or REM sleep episode, correspondingly), and number of awakening episodes. Data were normally distributed and the difference between depressed and control groups was analysed by one-way ANOVA. Cohen's d was used to estimate the effect sizes as follows: small ≥ 0.2, medium ≥ 0.5, large ≥ 0.8. The group differences were within the range of medium effect size.

NREM periods used for power spectral analysis were determined as a succession of sleep stages 1, 2, and 3 with a duration of 15 mins or more and terminated by REM sleep or wakefulness of least 5 mins. No minimum REM sleep duration was required for the first REM sleep episode. Only the first three episodes of NREM sleep were included in the power spectral analysis, since all the subjects had at least three NREM sleep episodes during the night.

4.2.2.4. Power spectrum analysis

The EEG (sampling rate 200 Hz) was subjected to power spectrum analysis off-line with the help of Spike 2 software (version 8.07 CED, Cambridge). Power spectrum (Hanning window) from central C4-M1 and frontal F4-M1 channels were computed, using FFT size of 512 Hz giving a resolution of 0.39 Hz. The spectral power was averaged in 30 sec epochs to be of identical length with the stage-score epoch length. Power spectra of SWA range (delta frequencies 0.39–3.91Hz) were calculated separately for NREM episodes 1, 2, and 3 from each derivation.

Spectral powers were normalized in each recording to the mean SWA power across two derivations during the first three NREM episodes to enable comparisons of the recordings between persons. To evaluate SWA dissipation, the difference in SWA power between the first and the third NREM sleep episodes was calculated. Artifacts were excluded by visual inspection and only artifact free epochs were included in the power spectral analysis (artifacts comprised on average 1,6 % of the total recording time). Repeated measures ANOVA was used to analyze difference between control and depressed group as well as compare SWA power in different NREM sleep episodes and SWA dissipation. To assess relationship between SWA dissipation and severity of depression (evaluated by HDRS and BDI-21) Pearson correlation coefficients were calculated.

5. Results and Discussion

5.1. Exon-specific *BDNF* expression and methylation at different promoters during spontaneous sleep and after SD

Previous studies have established a role for BDNF as a regulator of sleep homeostasis. However, not much is known about epigenetic mechanisms of *BDNF* gene regulation during sleep. In this thesis, first I investigated the epigenetic mechanisms of *BDNF* gene expression during spontaneous sleep and SD in a rat model. By these means, I obtained additional information about the role of BDNF in sleep regulation.

Increased number of *BDNF* transcripts 1-4 and *BDNF* 9a has been shown after 12 h SD in genetically modified mice (Martinowich et al., 2011). This suggests that *BDNF* transcription during sleep can be exon-specific and epigenetic mechanisms might play an important role in BDNF regulation of sleep homeostasis.

Three *BDNF* promoters, p1, p4 and p9, share similar features. All of them contain a Clock-controlled enhancer element E-box and are the most responsive to neuronal activity in vitro (Pruunsild et al., 2011). We studied the dynamic changes in these three *BDNF* transcripts' expression and their promoters' methylation levels in two brain regions involved in sleep regulation, BF and cortex, during the spontaneous sleep–wake cycle and after SD. We performed *BDNF*1, *BDNF*4 and *BDNF*9a expression and transcription-silencing 5-methylcytosine (5mC) DNA modification analyses in rats that were sleep-deprived for different periods of time (3, 6 and 12 h) and in non-deprived rats.

5.1.1 *BDNF1*, *BDNF4* and *BDNF9a* expression and DNA methylation in corresponding promoter regions during spontaneous sleep and after sleep deprivation in the basal forebrain

BDNF transcripts' expression. We did not find any significant changes in daily oscillation of *BDNF* transcripts in non-deprived rats, instead SD upregulated both *BDNF*1 and *BDNF*4 expression (main effect of SD on *BDNF*1 $F_{1,23} = 8.38$, $P = 0.008$ and *BDNF*4 $F_{1,26} = 13.22$, $P = 0.001$, Figure 8a). It should be noticed that

BNDF4 expression was affected more robustly by SD compared to other analyzed transcripts; its level was increased after 3 h SD and then returned to the level of the control non-deprived rats after 12 h SD (main effect of time $F_{2,26} = 7.53$, $P = 0.003$; interaction SD by time $F_{2,26} = 6.54$, $P = 0.005$). Similar activation of *BDNF* transcription after short-term SD was found in different brain regions in mice exposed to an acute restraint stress (Calabrese et al., 2009).

DNA methylation. Taking into account the total number of CpG dinucleotides that could be potentially detected as methylated in our MeDIP experiment (up to 25 CpG at p1, 18 CpG at p4 and 13 CpG at p9), the percentage of methylated cytosines at *BDNF* p1 was lower than at the other two promoters (Figure 8b). The 5mC levels at *BDNF* promoters were not affected significantly by time or SD factors (Figure 8b), although the amount of methylated cytosines at *BDNF* p1 was slightly increased after SD ($F_{1,27} = 3.70$, $P = 0.065$).

Association between BDNF transcripts' expression and DNA methylation. To further study whether *BDNF* DNA methylation was associated with the levels of transcription driven from the corresponding promoters, a regression analysis was performed. We found a significant negative correlation between the *BDNF1* transcript expression and DNA methylation level at the corresponding promoter in non-deprived rats ($R = -0.713$, $F_{1,13} = 12.40$, $P = 0.004$). After SD this correlation was absent ($R = +0.15$, $F_{1,14} = 0.30$, $P = 0.59$; Figure 8c). No correlation for *BDNF* p4- or p9-driven transcription was found.

Thus, we suggest that during spontaneous sleep–wake cycle, the changes in DNA methylation might contribute to *BDNF1* transcription in rats, but they are not associated with *BDNF4* and *BNDF9a* transcription in the BF.

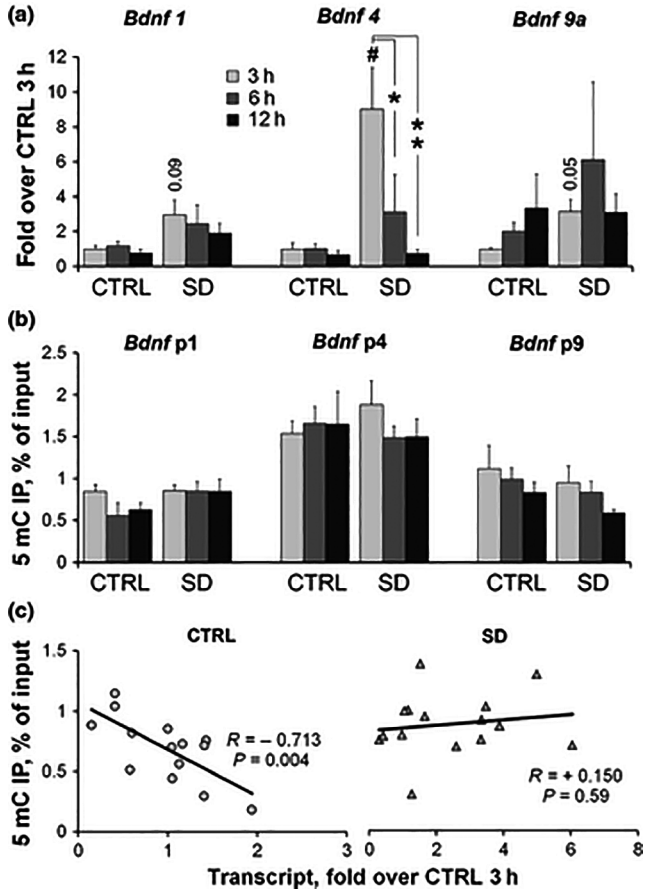

Figure 8. *BDNF* transcripts (a) and 5mC levels at the promoters *BDNF* p1, p4 and p9 (b) in the basal forebrain in the CTRL and SD groups. n = 4–6 rats per group. #P < 0.05 and 0.09, P = 0.09 and 0.05, P = 0.05 in comparison with SD and the respective non-deprived CTRL group; *P < 0.05, **P < 0.01 in comparison with different time-points within the same treatment (CTRL or SD) groups. (c) Regression analysis of the *BDNF*1 transcript and p1 DNA methylation levels; n = 14–15 rats per treatment. Reproduced from (Ventskovska et al., 2015) with permission from the publisher.

5.1.2 *BDNF1*, *BDNF4* and *BDNF9a* expression and DNA methylation in corresponding promoter regions during spontaneous sleep and after sleep deprivation in the cortex

BDNF transcripts' expression. *BDNF* transcripts in the cortex were induced by SD. The most prominent changes were observed in *BDNF*1 expression (main effect

of SD $F_{1,28}$ = 26.56, P < 0.0001). *BDNF*1 transcripts were significantly upregulated after 3 h SD and remained elevated throughout the SD, reaching a 10-fold difference from the controls after 12 h SD. This finding goes in line with previously reported data on the cortical exon-specific *BDNF* expression (Martinowich et al., 2011) and suggests that the *BDNF* promoters' structure allows high flexibility and specificity to respond to different environmental stimuli, including loss of sleep.

The expression of other transcripts was also elevated after SD but in smaller extent than *BDNF*1: for *BDNF*4 $F_{1,28}$ = 10.47, P = 0.003 and for *BDNF*9a $F_{1,28}$ = 3.29, P = 0.081 (Figure 9a). *BDNF*4 expression was upregulated throughout the SD. On the one hand, we observed a decrease in *BDNF* expression with increasing of SD duration that can be regarded as a down-regulation of *BDNF* in response to prolonged stress (Roceri et al., 2004). However, on the other hand, there was a significant effect of the time factor on the decrease in *BDNF*4 levels ($F_{2,28}$ = 4.11, P = 0.027), which can represent a diurnal variation in its expression.

DNA methylation. The percentage of methylated cytosines at the promoter *BDNF* p1 demonstrated daytime regulation: methylation level at this promoter decreased significantly by the end of the light phase (ZT12, CTRL group 12 h) in non-deprived rats, whereas such regulation was absent in SD rats (Figure 9b). The daytime 5mC changes were not observed at the promoter *BDNF* p4 in non-deprived CTRL rats, but there was a trend for the main effect of SD on decrease in *BDNF* p4 methylation ($F_{1,26}$ = 3.46, P = 0.074) with a significant decrease after 12 h SD. The 5mC levels at *BDNF* p9 promoter were decreased at ZT12 time independently of SD treatment (main effect of time $F_{2,27}$ = 7.03, P = 0.0035).

Association between BDNF transcripts' expression and DNA methylation. The regression analysis revealed a significant negative correlation between the *BDNF*9a expression and 5mC levels at BDNF p9 in non-deprived rats (R = 0.558, $F_{1,16}$ = 6.79, P = 0.02). But this correlation was not observed in SD rats (R = +0.042, $F_{1,14}$ = 0.02, P = 0.88; Figure 9c).

Thus, in the cortex, DNA methylation was not associated with the SD-induced *BDNF*1 expression but was associated with the *BDNF*9a levels in CTRL non-deprived rats and, possibly, with the *BDNF*4 levels at the 12-h time-point.

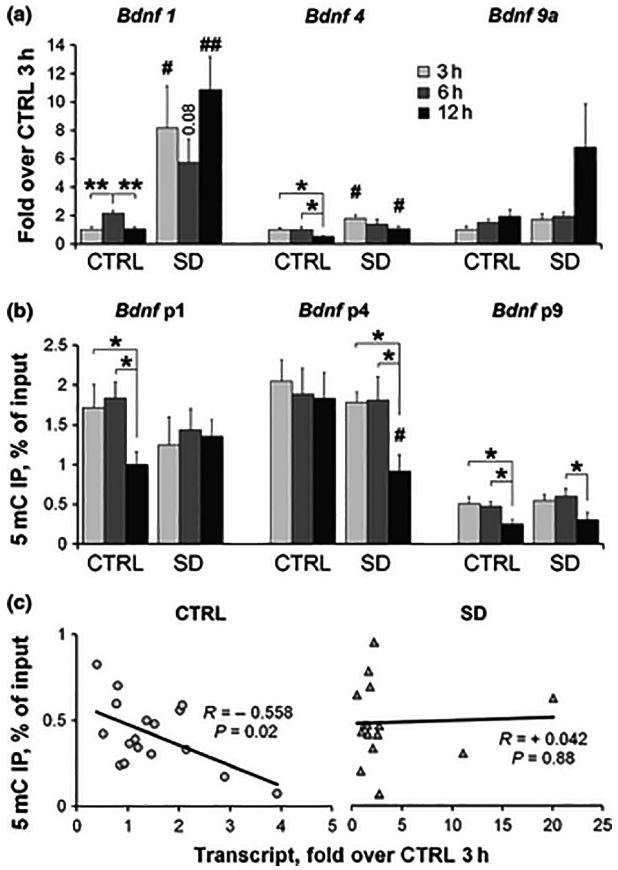

Figure 9. *BDNF* transcripts (a) and 5mC levels at the promoters *BDNF* p1, p4 and p9 (b) in the cortex in the CTRL and SD groups. n = 4–6 rats per group. #P < 0.05, ##P < 0.01 and 0.08, P = 0.08 in comparison with the SD and respective CTRL non-deprived groups; *P < 0.05, **P < 0.01 in comparison with different time-points within the same treatment (CTRL or SD) groups. (c) Regression analysis of the *BDNF*9a transcript and p9 DNA methylation levels; n = 15–17 rats per treatment. Reproduced from (Ventskovska et al., 2015) with permission from the publisher.

SD effects. We demonstrated for the first time that in two interconnected brain areas crucial for sleep regulation (BF and frontal cortex) the magnitude and dynamics of SD-induced *BDNF* up-regulation were transcript- and brain area-dependent. SD led to the robust but transient *BDNF*4 up-regulation in the BF and persistently strong *BDNF*1 up-regulation in the cortex. However, regression analysis between different *BDNF* transcripts' expression and DNA methylation in corresponding promoters did not reveal a significant correlation in SD rats.

Interestingly, short-term SD rapidly induced *BDNF* transcription; the highest *BDNF1* levels in the BF and *BDNF4* levels in both brain areas were observed already after 3 h SD. Similar to this finding, fast and transient increase of *BDNF* mRNA levels was observed in different brain regions (e.g. hippocampus, prefrontal and cingulate cortex) in rats exposed to an acute restraint stress (Calabrese et al., 2009). Although the effect of stress in our experiments was minimized by handling the animals before the experiments and using gentle technique of SD, *BDNF* up-regulation may indicate the necessity of activation of different neuronal plasticity-related genes in response to acute stress (Calabrese et al., 2009). With increasing of SD duration we observed decrease in *BDNF* expression, which is known as down-regulation of *BDNF* in response to prolonged stress and was shown in repeated maternal deprivation experiments (Roceri et al., 2004).

Diurnal variations. We found a daily variation in *BDNF* levels in non-deprived rats: the highest *BDNF1* expression after 6 h of rest phase and the lowest *BDNF4* levels after 12 h of rest phase in the cortex. Diurnal *BDNF* variation was also found previously in the hippocampus, where the lowest *BDNF1* and *BDNF4* levels were seen after 6 h of rest phase (Berchtold et al., 1999). We think that transient increase in *BDNF1* expression may reflect the ongoing, local plasticity processes during sleep, whereas decrease in *BDNF4* expression after 12 h of rest period may reflect the involvement of BDNF in regulation of sleep homeostasis (Porkka-Heiskanen, 2013).

Regression analysis between different *BDNF* transcripts' expression and DNA methylation in corresponding promoters revealed a negative correlation only for the control non-deprived rats: in the BF for *BDNF*1, and in the cortex for *BDNF*9a. It is known that DNA methylation is implicated in circadian regulation of mammalian Clock-related gene expression (Satou et al., 2013). Moreover, recently it has been shown in mice that changing of light conditions alters gene expression profiles and promoter DNA methylation in the SCN, thus it has been suggested that DNA methylation might be a mechanism driving circadian clock plasticity in mammals (Azzi et al., 2014). In the present study we showed for the first time that DNA methylation is also involved in diurnal regulation of non-Clock genes, such as BDNF.

Thus, we showed that the modulation of *BDNF* transcription during spontaneous sleep-wake cycle is at least partially regulated by brain site specific

DNA methylation. Further research is needed to understand the complexity of epigenetic regulation involved in diurnal regulation of *BDNF* expression.

The fact that no associations were found between the 5mC and mRNA levels in sleep deprived rats suggests that SD could disturb the normal epigenetic control of *BDNF* expression, implicating other regulatory mechanisms of BDNF transcription (e.g. 5-hydroxymethylcytosine DNA modification, histone modifications).

5.2. Effect of cross-fostering on sleep, *BDNF* gene expression and behaviour in rats

Adversities experienced during early periods of life, such as childhood and adolescence, have been associated with early-onset depression (Arnow, 2004). Both of these age periods are extremely important for future mental wellbeing and are vulnerable to environmental conditions. The exact mechanisms by which environmental stimuli affect future development have been the subject of many studies but still remain poorly understood. BDNF appears to be a good candidate for mediating short- and long-term effects of early life stress experience on brain function later in adulthood.

So, we studied the molecular level mechanisms by which the early life stress modifies sleep by using animal model (cross-fostering) (Study II). This model is based on changing the pups between mothers at early age. CF may satisfyingly model stressful life events in human childhood and it demonstrates the critical role of the postnatal maternal environment in programming the offspring's development.

5.2.1 Spontaneous sleep, sleep homeostasis and adenosine in adult cross-fostered rats

Cross-fostering procedure led to increased total time spent in NREM sleep ($F_{2,273}$ = 4.14, P = 0.04) and REM sleep ($F_{2,273}$ = 7.17, P = 0.008) and decreased time in waking ($F_{2,273}$ = 4.77, P = 0.03) compared to CTRL rats during light period of the day (Figure 10A).

In addition, a difference in REM sleep parameters between sexes was observed (for REM duration: $F_{2,273}$ = 35.28, P < 0.001; for REM episodes onsets: $F_{2,273}$ = 16.67, P < 0.001). Individual one-way ANOVA within the same-sex group revealed that REM sleep duration in CF females was significantly longer than in CTRL females

($F_{1,118}$ = 4.1, P = 0.04), while in CF males there was no statistically significant difference, although there was observed the same tendency ($F_{1,153}$ = 3.35, P = 0.07).

Both CF male and female rats also had higher number of REM sleep onsets than CTRL rats ($F_{2,273}$ = 18.5, P < 0.001) during the light period (Figure 10B).

During the dark period, CF rats tended to have longer NREM sleep duration ($F_{2,273}$ = 3.46, P = 0.06) (Figure 10A) and had significantly higher number of REM sleep onsets ($F_{2,273}$ = 5.32, P = 0.032) compared to CTRL rats (Figure 10B). It should be noted that individual analysis of main effect of CF within the same-sex group revealed that the number of REM sleep onsets was significantly higher in CF females compared to CTRL females ($F_{1,118}$ = 5.56, P = 0.02), but was similar between CF and CTRL males ($F_{1,154}$ = 0.8, P = 0.37) (Figure 10B).

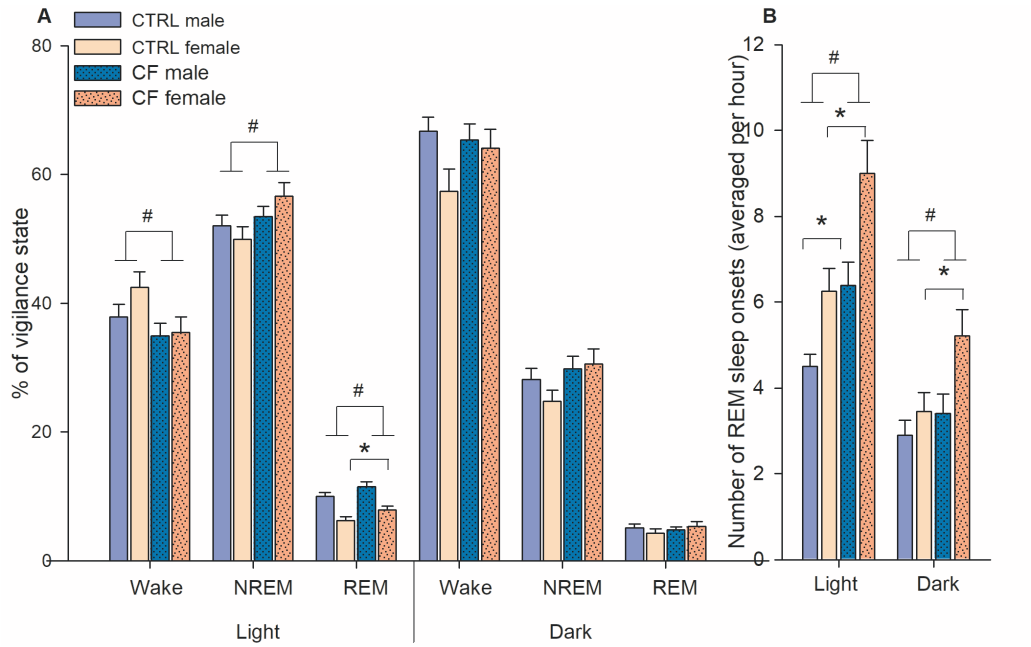

Figure 10. (A) Distribution of the vigilance states through light-dark period and (B) number of REM sleep episode onsets in CTRL and CF male and female rats during spontaneous sleep (BL day). Males (n=7/control group, n=6/CF group), females (n=5/CTRL group, n=5/CF group). # – P < 0.05, difference is significant between CTRL and CF rats (factorial ANOVA (factors: sex and treatment), * – P < 0.05, difference is significant between CTRL and CF rats within same-sex group (one-way ANOVA), CTRL: control rats, CF: cross-fostering rats, BL: baseline day. Modified from Study II.

Previously, it has been shown that early life stress, particularly maternal separation, has an effect on spontaneous sleep in adulthood: increases in different parameters of REM sleep (Sampath et al., 2014; Tiba et al., 2004); some studies have reported also decrease in REM sleep (Perez-Morales et al., 2014; Reyes Prieto et al., 2012). Changes in NREM sleep have also been reported (Mrdalj et al., 2013; Perez-Morales et al., 2014; Reyes Prieto et al., 2012; Sampath et al., 2014). Our results, indicating more time spent in NREM and REM sleep by CF rats are in line with many previous studies, although the CF model deviates from the maternal separation.

Higher number of REM sleep onsets during spontaneous sleep in CF treated rats was the most prominent change in sleep pattern observed in our study. Sleep disturbances and particularly disinhibition of REM sleep is one of the core symptoms of depression in humans (Adrien, 2002; Fleming, 1994), which has its equivalent in rats (Schmidt et al., 2011), indicating that the CF model may be a useful animal model of depression.

Adenosine, produced in BF, is an important regulator of sleep homeostasis (Basheer et al., 2004; Porkka-Heiskanen et al., 1997). We measured its amount during inactive phase of BL day and found that its level was significantly lower in both male ($F_{1,128} = 6.47$, $P = 0.012$) and female ($F_{1,66} = 7.44$, $P = 0.008$) CF rats compared to CTRLs (Figure 11). We suggest that the low adenosine level in BF could contribute to the sleep changes observed in the adult CF rats. Decreased adenosine level in this area has been observed in other animal models of depression (Savelyev et al., 2012), and a role for low adenosine level has been previously suggested in development of depressive disorder (Gass et al., 2010; Hanson, 2009; Kaster et al., 2004).

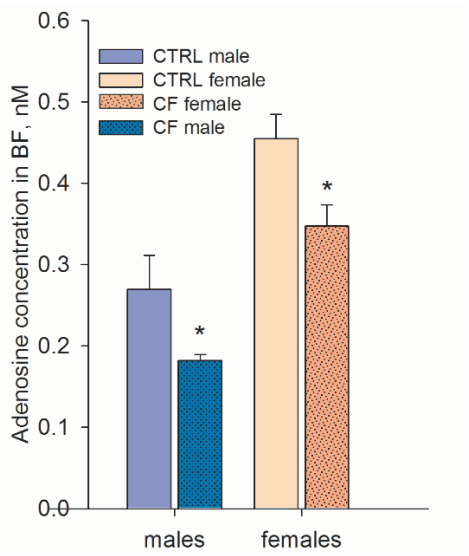

Figure 11. Adenosine concentration (nM) during BL day (09:30 AM–08:30 PM) in CTRL and CF male and female rats in the basal forebrain. Males: n=7/CTRL group, n=6/CF group, females: n=5/CTRL group, n=5/CF group. * – $P < 0.05$, difference is significant between CTRL and CF rats within same-sex group (one-way ANOVA). CTRL: control rats, CF: cross-fostering rats, BF: basal forebrain, BL: baseline. Modified from Study II.

To study whether sleep homeostasis is affected in CF rats, we performed 3h SD during the light period of the day (1 PM – 4 PM) and analysed recovery sleep (4 PM – 9 PM). Factorial repeated measures ANOVA showed that SD led to normal homeostatic response in all rats. Total NREM sleep duration was increased after SD compared to respective baseline period ($F_{1,20} = 35.50$, $P < 0.001$) in both CTRL and CF rats irrespective of sex (Figure 12A).

For REM sleep, factorial repeated measures ANOVA revealed a significant effect of sex and treatment, with significant interactions between SD and sex, and between SD and treatment. The effect of SD did not reach level of significance ($F_{1,20} = 3.93$, $P = 0.06$). Males had longer REM sleep duration than females during spontaneous sleep, but shorter during recovery period ($F_{1,20} = 10.96$, $P = 0.003$, the interaction $F_{1,20} = 28.88$, $P < 0.001$). CF rats had longer REM sleep duration than CTRL rats during spontaneous sleep ($F_{1,20} = 6.72$, $P = 0.02$) but its duration was equal during the recovery period (the interaction $F_{1,20} = 4.96$, $P = 0.04$).

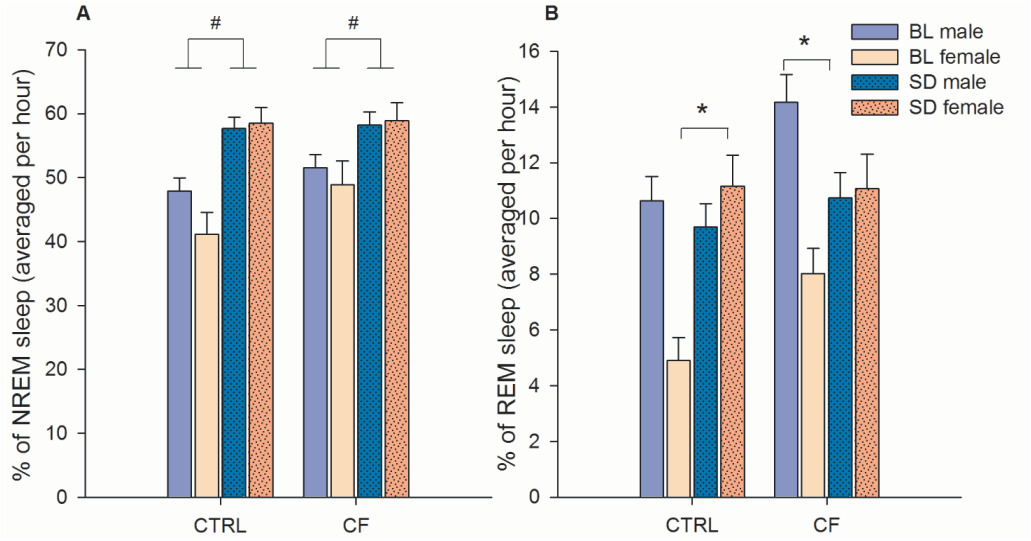

Figure 12. (A) NREM and (B) REM sleep duration in CTRL and CF male and female rats during recovery period (4 PM - 9 PM) after SD. Males (n=7/control group, n=6/CF group), females (n=5/CTRL group, n=5/CF group). # – $P < 0.05$, difference is significant between BL and SD days, when the post-SD recovery sleep was compared to the corresponding hours of BL day (factorial repeated measures ANOVA (between subject factors: sex and treatment, within-subject factor: SD). * – $P < 0.05$, difference is significant between BL and SD days within same–sex, same-treatment group (one-way ANOVA). CTRL: control rats. CF: cross-fostering rats, BL: baseline day, SD: sleep deprivation. Modified from Study II.

Individual one-way ANOVA within same-sex and same-treatment group revealed that REM sleep duration was decreased in CF males after SD ($F_{1,5} = 20.17$, $P = 0.006$) compared to respective hours of BL day; in CTRL males, no changes of REM sleep duration were found. CTRL females demonstrated higher REM sleep duration after SD ($F_{1,4} = 11.10$, $P = 0.03$), but not CF females (Figure 12B).

One of the most powerful sleep homeostasis marker is EEG delta power in NREM sleep (Borbely & Achermann, 1999). We calculated NREM sleep delta power (1.08-3.79 Hz) during one hour of post-SD recovery period (4 PM - 5 PM) and found that all rats demonstrated significant increase in delta power during the recovery sleep ($F_{1,20} = 88.392$, $P < 0.001$) irrespective of treatment (CF vs. CTRL) (Figure 13). This indicates that homeostatic sleep regulation was not significantly impaired in CF rats. Sleep deprivation for 3 h is considered a minimum time to

induce increased NREM sleep duration and intensity, as well as in elevated BF adenosine level (Kalinchuk et al., 2006; Savelyev et al., 2012).

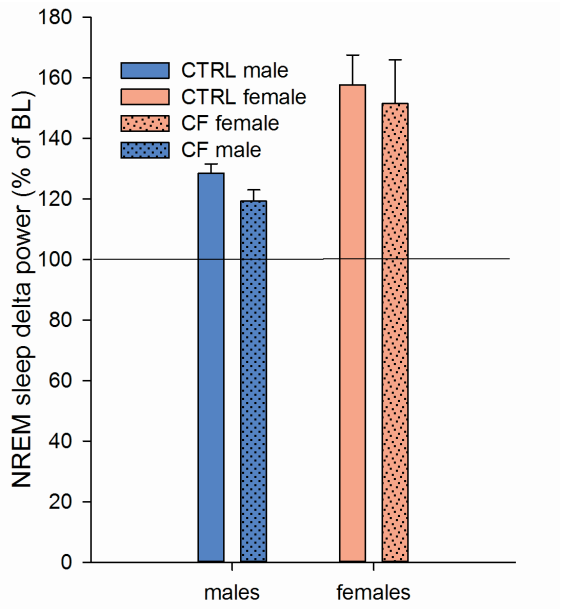

Figure 13. NREM sleep delta (1.08-3.79 Hz) power in CTRL and CF male and female rats during one-hour recovery sleep (4 PM -5 PM) after SD. Delta power during corresponding time (4 PM -5 PM) during BL day was used as a reference value of 100%. Males (n=7/control group, n=6/CF group), females (n=5/CTRL group, n=5/CF group). While there is a significant increase ($P < 0.05$) in NREM sleep delta power after SD (factorial repeated measures ANOVA (between subject factors: sex and treatment, within-subject factor: SD) for both females and males (about 50% and 20%, respectively), there is no difference between CTRL and CF rats. CTRL: control rats. CF: cross-fostering rats, BL: baseline day, SD: sleep deprivation. Modified from Study II.

5.2.2 Effect of cross-fostering treatment on *BDNF* gene expression in adolescence and adulthood

BDNF is one of the main neuroplasticity molecules, which is known to be involved in both sleep regulation and depression development (Castren & Rantamaki, 2010; Dwivedi, 2009; Faraguna et al., 2008). We measured the *BDNF* expression levels in males and females at two time points: during adolescence

(PND 50) and adulthood (PND 110). The measurements were done in three brain regions: basal forebrain, frontal cortex, and hypothalamus.

Factorial ANOVA yielded significant main effects for age and treatment factors in BF, but not for sex factor. No differences were found in other brain areas.

Rats demonstrated higher *BDNF* gene expression during adolescence compared to adulthood ($F_{3,41}$ = 7.75, P = 0.009), and CF rats tended to have lower *BDNF* gene expression compared to CTRLs ($F_{3,41}$ = 3.17, P = 0.08) (Figure 14).

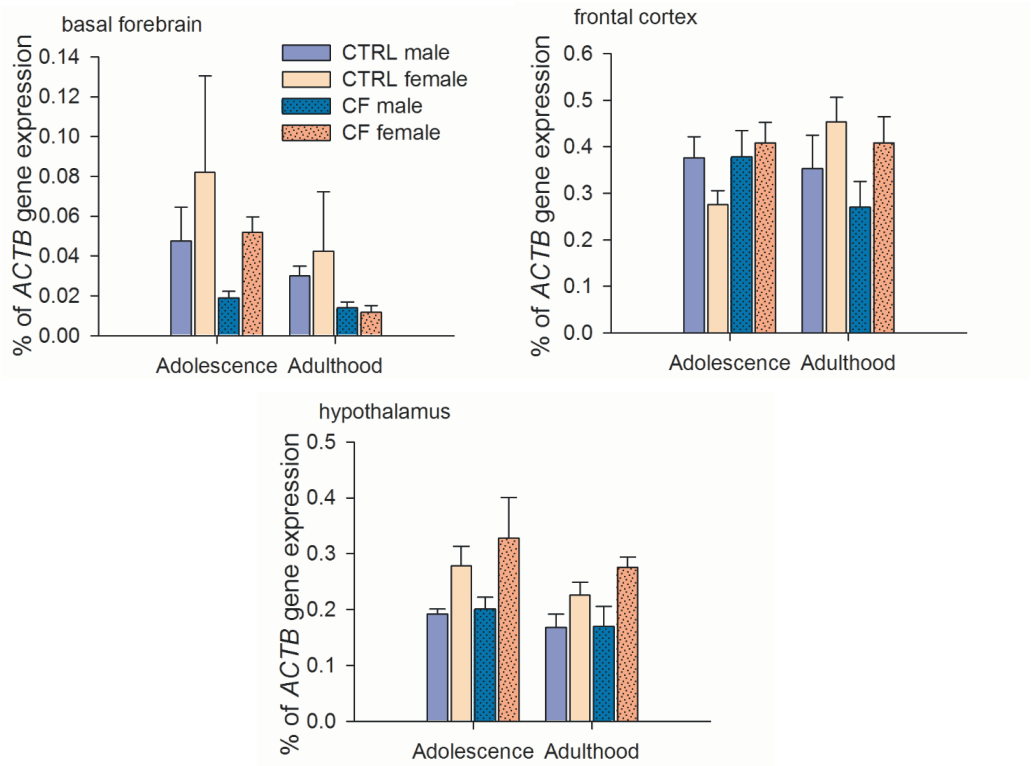

Figure 14. *BDNF* gene expression in the basal forebrain, frontal cortex and hypothalamus in male and female rats during adolescence and adulthood. Males: n=5/CTRL group, n=6/CF group, females: n=6/CTRL group, n=6/CF group. In the basal forebrain, *BDNF* gene expression level was higher in adolescence than in adulthood (factorial ANOVA (factors: sex, age and treatment), P < 0.05) and tended to be lower in CF rats than in CTRLs (factorial ANOVA (factors: sex, age and treatment), P = 0.08). The differences were not observed in other brain areas. CTRL: control rats, CF: cross-fostering rats, ACTB: actin beta. Modified from Study II.

Age-dependent changes in *BDNF* gene expression have been detected before in hippocampus (decreasing with age) and medial prefrontal cortex (increasing with age) (Wang et al., 2015). To our knowledge, this study is the first to investigate changes in *BDNF* gene expression in the BF with age. Previously (Wang et al., 2015), it has been shown that maternal separation differently affects *BDNF* gene expression in individual brain regions: increases in hippocampus and decreases in medial prefrontal cortex. In another study, the down-regulation of *BDNF* expression was shown only in prefrontal cortex, but not in hippocampus (Roceri et al., 2004). We did not find any significant difference between CTRLs and CF rats in any of the studied brain regions (frontal cortex or hypothalamus) beside tendency to lower gene expression in CF rats compared to CTRLs in BF. We hypothesize that this trend in *BDNF* expression observed in the BF, may reflect dysfunction of the cholinergic neurotransmission that may account for the development of cognitive impairment often associated with psychopathologies (Dagyte et al., 2011) and usually observed in cognitive tests.

5.2.3 Effect of cross-fostering treatment on behaviour and body weight in adolescence and adulthood

To assess anxiety and depression-like behaviour in CF rats, we performed two behavioural tests: OF test (in adolescence, at PND 45, and in adulthood, at PND 100) and SPT in adulthood only, at PND 105.

Open field test is widely used to assess general locomotor activity, emotionality, and exploratory behaviour in rodents. Horizontal (number of squares crossed in inner and outer areas) and vertical activities (rearings) are used as measures of exploration behaviour. At the same time, activity in the central vs. peripheral sectors reflects the proportion between exploratory behaviour and harm avoidance, respectively (Archer, 1973; Redina et al., 2009). The number of defecations and urinations is used as indicator of sympathetic nervous activity (Archer, 1973). All these parameters taken together give an estimate of the anxiety level of the animal.

In the OF test, factorial ANOVA yielded a significant effect of sex on 3 parameters, and significant effect of age on 5 parameters, but no significant effect of CF treatment was found in any studied parameter of the OF test.

The number of defecations was higher in males ($F_{3,137}$ = 24.93, P < 0.001) compared to females (Table 3, 4). Females had higher number of squares crossed in outer area ($F_{3,137}$ = 10.51, P = 0.001) and total number of squares crossed ($F_{3,137}$ = 9.02, P = 0.002) compared to males. Moreover, the number of squares crossed in outer area ($F_{3,137}$ = 14.07, P < 0.001), the total number of squares crossed ($F_{3,137}$ = 14.34, P < 0.001), grooming number ($F_{3,137}$ = 24.17, P < 0.001) and duration ($F_{3,137}$ = 17.46, P < 0.001), and defecation number ($F_{3,137}$ = 6.3, P = 0.013) decreased with age (Table 3, 4). The interaction between sex and age was observed for defecation number ($F_{6,134}$ = 9.32, P = 0.003).

Table 3. Effect of CF on behaviour in the OF test in adolescent and adult male rats

	Adolescence		Adulthood	
Behavioural parameters	CTRL	CF	CTRL	CF
Horizontal activity				
Latency to leave the central area, s	3.9±0.4	4.7±0.8	7.2±2.6	5.2±1.2
Number of squares crossed in the inner area	5.4±1.2	2.5±0.8	1.4±0.3	1.8±0.5
Number of squares crossed in the outer area	68.1±6.0	62.4±6.3	40.1±5.5&	46.4±4.9&
Total number of squares crossed	73.3±6.3	65.0±6.5	41.7±5.5&	48.2±5.0&
Vertical activity				
Rearing number	13.8±2.4	8.7±1.7	6.0±1.4&	4.3±0.9&
Grooming				
Latency of grooming, s	146.5±28.1	133.3±24.5	203.5±18.5	141.5±3.5
Grooming episodes number	0.4±0.1	0.5±0.1	0.1±0.1&	0.2±0.2&
Total grooming duration, s	5.1±2.2	5.6±2.3	1.5±1.1&	0.7±0.5&
Excretory activity				
Defecation number	2.8±0.5	3.9±0.5	3.1±0.6&	3.7±0.7&
Urination number	2.5±0.6	2.1±0.6	2.6±0.6	2.5±0.4

Values represent mean ± SEM; & – difference is significant between two age groups (adolescence vs. adulthood, P < 0.05). Adolescence: n=22/CTRL group, n=21/CF group, adulthood: n=18/CTRL group, n=14/CF group. OF: open field; CTRL: control rats; CF: cross-fostered rats. Modified from Study II.

Table 4. Effect of CF on behaviour in the OF test in adolescent and adult female rats

	Adolescence		Adulthood	
Behavioural parameters	CTRL	CF	CTRL	CF
Horizontal activity				
Latency to leave the central area, s	4.6±0.6	6.9±2.1	5.1±1.3	4.5±0.8
Number of squares crossed in the inner area	2.9±0.6	3.2±0.5	1.9±0.3	4.3±1.0
Number of squares crossed in the outer area	78.4±6.2	70.7±5.4	55.2±5.1[&]	69.4±5.5[&]
Total number of squares crossed	81.3±6.4	73.9±5.6	57.1±5.2[&]	73.7±6.1[&]
Vertical activity				
Rearing number	10.8±1.5	9.4±1.3	9.1±2.0	13.9±2.3
Grooming				
Latency of grooming, s	118.6±14.5	136.5±11.6	-	-
Grooming episodes number	0.5±0.1	0.8±0.1	-	-
Total grooming duration, s	5.7±2.2	9.0±2.2	-	-
Excretory activity				
Defecation number	1.9±0.6	3.0±0.3	0.2±1.1[&]	0.5±0.3[&]
Urination number	2.6±0.6	3.0±0.5	2.1±0.9	1.8±0.3

Values represent mean ± SEM; & – difference is significant between two age groups (adolescence vs. adulthood, $P < 0.05$). Adolescence: n=14/CTRL group, n=24/CF group, adulthood: n=9/CTRL group, n=19/CF group. OF: open field; CTRL: control rats; CF: cross-fostered rats. Modified from Study II.

Thus, we concluded that the activity of CF rats in OF test did not differ significantly from the activity of CTRL rats in any of the studied parameters.

To test the depressive-like behaviour, we performed SPT, which is used in rodent experiments as a measure of sensitivity to reward (Harris et al., 1997) and anhedonia (Castagne et al., 2009; Willner et al., 1987). It has been shown previously in different animal models of depression that sucrose intake is significantly reduced in animals with depressive-like behaviour as compared to controls (Goshen et al., 2008; Grippo et al., 2005; Rygula et al., 2005; Savelyev et al., 2012).

We performed factorial ANOVA to assess the sucrose consumption in our rats, and it revealed no significant effect of CF treatment ($F_{2,54} = 1.74$, $P = 0.19$), sex ($F_{2,54} = 0.05$, $P = 0.83$), or interaction ($F_{2,53} = 1.39$, $P = 0.24$) between them (Figure 15).

Taking together the results from behaviour tests, we conclude that CF rats did not demonstrate anxiety or depressive-like behaviour under normal conditions and their behaviour was similar to the sex-and age-matched control rats. However, taking into account the molecular level changes after CF reported above, it can be suggested that under stressful conditions, brain resilience could be compromised, and behavioural changes would be observed.

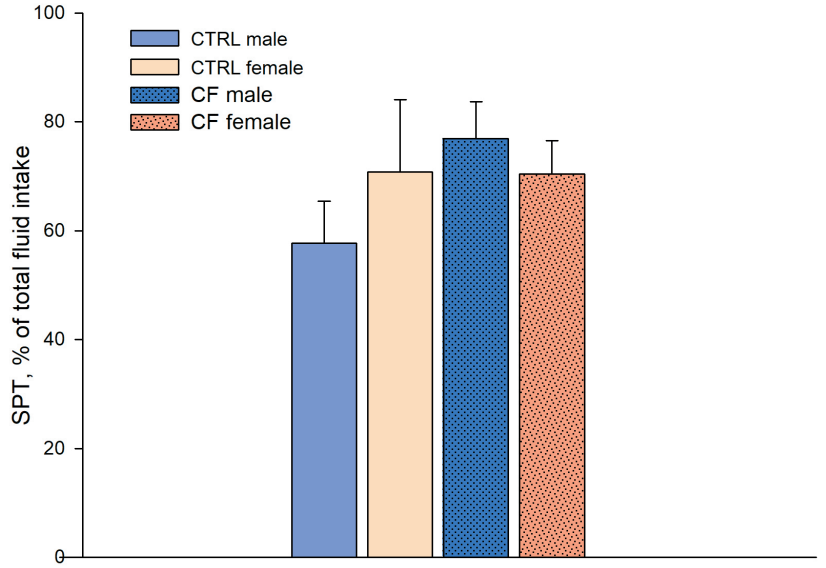

Figure 15. Effect of CF on sucrose preference in adulthood in males and females. Males: n=18/CTRL group, n=13/CF group, females: n=8/CTRL group, n=18/CF group. CTRL: control rats, CF: cross-fostered rats, SPT: sucrose preference test.

Along with behavioural indices, we also assessed weight of the rats. Factorial ANOVA revealed statistically significant effect of all the factors: sex, age, and treatment on weight of the animals.

The body weight was significantly higher in males compared to females ($F_{3,110}$ = 99.99, P < 0.001), and in adulthood compared to adolescence ($F_{3,110}$ = 562.21, P < 0.001) (Table 5). The interaction between sex and age was significant ($F_{3,107}$ = 172.98, P < 0.001). The effect of CF treatment was significant ($F_{3,110}$ = 4.11, P = 0.04).

Adolescent CF males had lower weight than CTRLs ($F_{1,26}$ = 5.63, P = 0.03), but in adulthood this difference faded away ($F_{1,30}$ = 0.46, P = 0.5), whereas CF females had lower body mass during both age periods: adolescence ($F_{1,25}$ = 15.79, P = 0.001) and adulthood ($F_{1,25}$ = 4.61, P = 0.04) (Table 5).

Table 5. Effect of CF on body weight in male and female rats during adolescence and adulthood

Treatment groups	Adolescence (weight, g)	Sample size, n	Adulthood (weight, g)	Sample size, n
Males				
CTRL	134.7±5,8	13	309.1±7.6[&]	18
CF	117.7±4,4*	15	302.4±5.1[&]	14
Females				
CTRL	130.7±2.8	6	203.9±2.7[&]	9
CF	106.9±3.1*	21	197.9±1.5*[&]	18

Values represent mean ± SEM. *- difference is significant between CTRL and CF groups within same-sex, same-age group ($P < 0.05$); [&] – difference is significant between two age groups ($P < 0.05$). CTRL: control rats; CF: cross-fostered rats. Modified from Study II.

Thus, both male and female CF rats had lower body weight in adolescence compared to CTRL rats, which is in line with previous studies on rodent models of depression (Willner, 2017). In adulthood only female CF rats demonstrated lower body weight compared to CTRLs. However, it should be noted that although the difference was statistically significant the real difference in weight between CF and CTRL adult female was rather small.

5.3. Sleep and slow-wave activity in early-onset depression

To evaluate what sleep changes occur in early-onset depression in humans, we assessed sleep macro- and micro-architecture in a clinical sample of depressed adolescent boys in Study III.

5.3.1 Sleep architecture in depressed adolescent boys

All participants underwent detailed clinical and psychiatric evaluation and 8 of them were diagnosed with depressive disorder according to the DSM-IV criteria; among age-matched controls no axis-I diagnoses were found. The details of psychiatric evaluation are presented in the table 6.

Table 6. Details of clinical and psychiatric evaluation in the study participants

Variable	Controls	Patients
Age	16.2 ± 0.7 (14.8-17.2)	16.9 ± 1 (14.7-17.3)
BMI	20.6 ± 1.7 (17.8-23.8)	16.9 ± 3.9 (16.8-27.6)
S-Testo	19.4 ± 3.6 (15.3-27.6)	16.9 ± 4.1 (15.0-26.3)
BDI-21	2.8 ± 4.0 (0-12)	16.9 ± 12.0 (1-33) *
HDRS	0.3 ± 0.7 (0-2)	12.9 ± 4.4 (7-19) **
AIS	2.1 ± 2.2(0-7)	9.4 ± 6.2 (1-18) *

Data are reported as mean ± SD (range). Significant differences between controls and depressed patients are marked with asterisks (*p<0.05, **p<0.001). Controls: n= 10, patients: n=8. BMI, Body Mass Index; S-Testo, serum testosterone level (nmol/l; n=9 for controls); BDI-21: 21-item Beck Depression Inventory; HDRS: Hamilton Depression Rating Scale; AIS: Athens insomnia scale. Modified from Study III.

Sleep was recorded in subjects' home environment during two consecutive nights. First night served as an adaptation night and recordings during the second night were used for assessing sleep macroarchitecture. Different sleep parameters (presented in the table 7) were evaluated. No significant differences in assessed parameters between patients suffering from depression and controls were found, beside shorter TST in the group of patients ($F_{1,15}$=6.61, p=0.02).

Table 7. Sleep parameters in control and depressed adolescents

Sleep parameters	Controls	Patients
TST, min*	481.05 ± 11.84	426.57 ± 25.40
Sleep efficiency	97.15 ± 0.96	93.81 ± 3.42
N1, %	4.00 ± 0.84	4.86 ± 0.80
N2, %	51.68 ± 1.31	49.27 ± 2.71
N3, %	27.47 ± 1.79	29.30 ± 3.77
REM, %	16.86 ± 1.93	16.56 ± 2.20
SWS latency, min	14.85 ± 2.66	35.79 ± 17.56
REM sleep latency, min	151.35 ± 17.26	113.50 ± 22.09
Awakenings, number	11.40 ± 2.50	8.00 ± 1.68

Data are reported as mean ± SEM. Significant difference between control group and depressed patients group *p<0.05 (ANOVA). Controls: n=10, patients: n= 8. TST: total sleep time; N: NREM sleep stage; REM: rapid eye movement sleep; SWS: slow-wave sleep. Modified from Study III.

The shorter TST observed in the group of depressed adolescents may reflect such insomnia symptoms as difficulties in falling asleep, disrupted sleep continuity, and early morning awakenings that are very common for depressive disorder

(Steiger & Kimura, 2010). This is also evidenced by higher AIS scores found in our study in depressed boys compared to controls.

5.3.2 SWA power, dissipation and rise rate in depressed adolescent boys

Slow wave sleep with its typical SWA, is the most restorative stage of sleep (Akerstedt et al., 1997; Dijk, 2009). Temporal distribution of SWA through the night is considered to be a principal physiological marker of sleep homeostatic regulation (process S) (Borbely & Achermann, 1999). To assess sleep homeostasis in depressed boys, we calculated SWA delta power, its temporal distribution through the night (dissipation) and rise rate during the first NREM episode, and compared these parameters to the values found in the control group.

Across all individuals, we observed similar pattern with highest SWA in the first NREM and smallest in the third NREM episode ($F_{2,32}$=22.06, p<0.001), which is the normal course of spontaneous sleep, when the highest SWA is observed in the beginning of the night and then gradually declines towards the end of the night (Dijk, 2009). Moreover, all individuals had higher SWA values in frontal derivation as compared to central one ($F_{1,16}$=63.49, p<0.001).

We found that SWA in depressed patients was lower during the first NREM episode in the frontal derivation ($F_{1,16}$=6.8, p=0.02) and higher during the third NREM episode in the central derivation ($F_{1,16}$=5.37, p=0.03) as compared to healthy subjects (Figure 16).

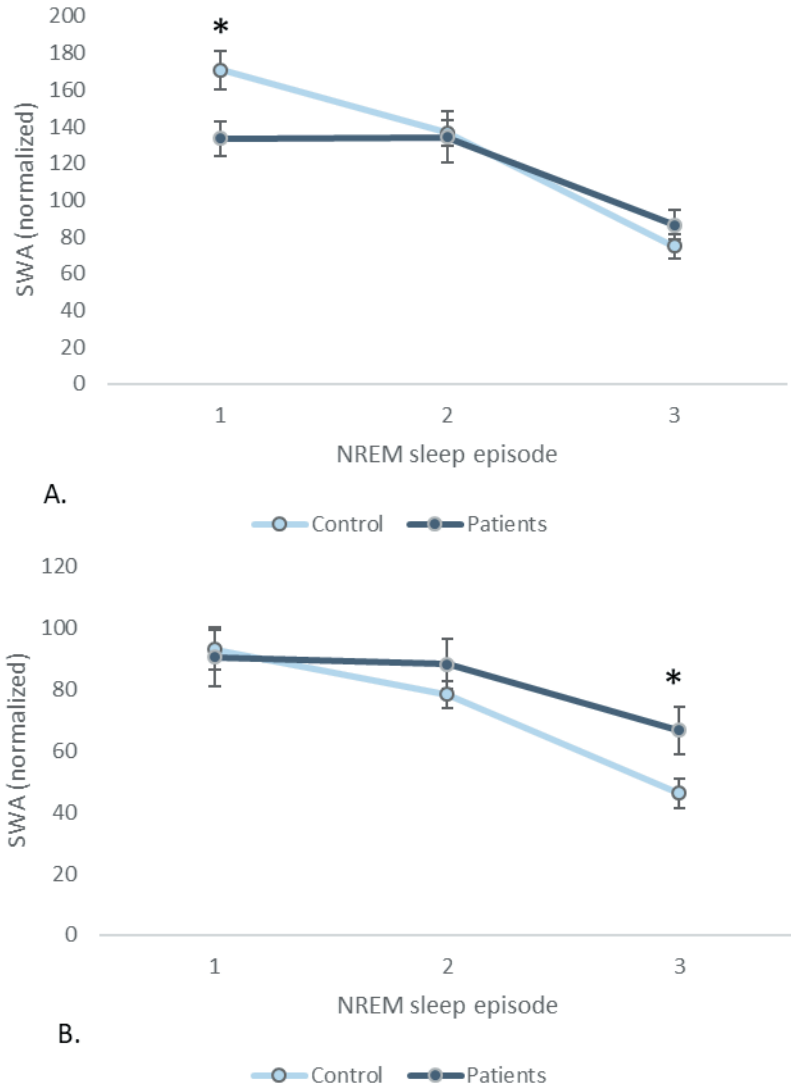

Figure 16. SWA power in frontal (A) and central (B) EEG derivations in first three NREM sleep episodes. Controls: n=10, patients: n= 8. Data are illustrated as mean ± SEM. Repeated measures ANOVA (within-subject factors: derivation and NREM episode, between-subject factor: group), *p<0.05. SWA: slow-wave activity; EEG: electroencephalogram; NREM: non-rapid eye movement sleep. Modified from Study III.

To assess the dissipation pattern of SWA through the night, we estimated the decline of SWA from the first to the third NREM episode and found that it was more prominent in the group of healthy subjects as compared to depressed patients ($F_{1,16}$=5.21, p=0.04) and in the frontal derivation as compared to the central

derivation ($F_{1,16}$=30.93, p<0.001). In the frontal derivation, the difference in SWA power between the first and the third NREM episode was significantly smaller in depressed patients compared to healthy controls ($F_{1,16}$=5.32, p=0.04); in the central derivation it tended to be smaller ($F_{1,16}$=4.09, p=0.06) (Figure 17).

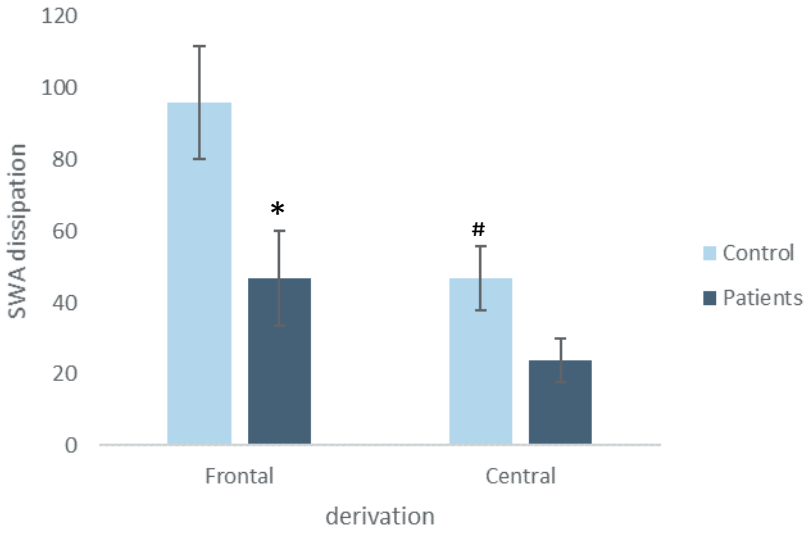

Figure 17. Decline of SWA power from the first to the third NREM episode in frontal and central EEG derivations. Controls: n=10, patients: n=8. Data are illustrated as mean ± SEM. Repeated measures ANOVA (within-subject factors: derivation, between-subject factor: group), *p<0.05, #p=0.06. SWA: slow-wave activity; EEG: electroencephalogram; NREM: non-rapid eye movement sleep. Modified from Study III.

Lower SWA power in the first NREM episode and smaller decline of SWA in the course of sleep found in the group of depressed patients indicate a more evenly distributed SWA power through the night in depressed patients compared to healthy subjects. These observations are in line with earlier studies in adults (Argyropoulos & Wilson, 2005; Armitage, 2007) and adolescents (Armitage et al., 2001; Lopez et al., 2012), showing that depressed patients have lower SWA power in the first NREM sleep episode and an irregular SWA dissipation pattern compared to healthy individuals. Moreover, lower SWA power observed in depressed patients compared to healthy subjects can be also associated with different physical and mental activities during preceding wake period. It has been recently shown in animal and human studies that sensory stimulation, physical exercise and learning

during preceding wake period can locally increase SWA in involved brain areas (Huber et al., 2004; Vyazovskiy et al., 2000; Vyazovskiy et al., 2006). This is in line with the synaptic homeostasis hypothesis of sleep, postulating that synaptic downscaling occurring during sleep selectively favours strengthening of those synaptic connections that were used most during preceding wakefulness (Tononi & Cirelli, 2003). Actigraphic study performed in the same sample of depressed adolescent boys (Merikanto et al., 2017) demonstrated lower activity levels in depressed boys compared to healthy subjects, especially on weekends. Thus, it can be suggested that the difference in SWA power between depressed and control subjects can be at least partially associated with different activity levels during wake periods preceding EEG recordings. However, additional experiments to clarify this issue should be performed, including brief questionnaires evaluating mental and physical activity of the subjects.

We found that in the central derivation SWA in depressed adolescents remained at a higher level after three NREM episodes as compared to healthy controls. This may be interpreted as a partially incomplete dissipation of SWA, which means that the core restorative process of sleep is unsuccessful and part of sleep pressure remains. This resembles the sleep deprivation-like state in healthy individuals, which is known to affect mood and performance (Alhola & Polo-Kantola, 2007; Simon et al., 2015). However, it should be noted that in our study SWA was assessed only during the first three NREM episodes, since all the subjects had at least three NREM episodes during the night; but some of the participants had four (or sometimes even five) episodes of NREM in total. It is therefore possible that during the following episodes the levels of SWA in controls and depressed subjects reached the same level. To verify this assumption a bigger sample size is needed to be able to match control and depressed participants by the number of NREM episodes during the night.

Another marker of sleep homeostatic regulation is a rise rate of SWA during the initial stage of sleep (Dijk et al., 1990), which is calculated during the initial stages of sleep. We found that during the initial 10 min of the first NREM sleep episode depressed patients demonstrated slower buildup of SWA compared to healthy subjects in the frontal derivation ($p=0.03$), but not in the central one ($p=0.16$).

In summary, depressed adolescent boys had lower SWA power and its slower rise in the course of the first NREM episode, and a flatter SWA dissipation through the night in frontal area as compared to healthy boys. These results indicate that homeostatic sleep regulation may be impaired in adolescent depression patients. Indeed, according to the S-deficiency hypothesis of depression (Borbely & Wirz-Justice, 1982), homeostatic sleep regulation is impaired in depression and depressed individuals fail to accumulate a sufficient sleep drive during daytime. Sleep deprivation is considered an efficient (but short-term) tool of depression treatment (Benedetti & Colombo, 2011; Borbely & Wirz-Justice, 1982; Wirz-Justice & Van den Hoofdakker, 1999), which improves mood via suppressing SWA and increasing homeostatic sleep drive. Both slower rise of SWA during initial stage of sleep and incomplete dissipation of SWA found in the group of depressed adolescents indicate lower homeostatic sleep drive.

The analysis of association between severity of depressive symptoms and SWA dissipation revealed that a flatter SWA dissipation was associated with more severe depression symptoms. A negative correlation between SWA dissipation and HDRS was found in the frontal ($r=-0.505$, $p<0.05$) derivation, but not in the central one ($r=-0.437$). This is an interesting finding in terms of the studies showing that the distribution of SWA across the first two NREM sleep episodes is associated with clinical outcome in depressed adult patients (Ehlers et al., 1996; Kupfer et al., 1990).

It should be noted that all the above reported results were more pronounced in frontal derivation compared to central one. Slow waves during sleep are more prevalent in frontal cortex compared to other brain areas (Cajochen et al., 1999; Huber et al., 2004), which was also observed in our study. Furthermore, morphometric and functional brain imaging studies (Arnone et al., 2012; Cooney et al., 2010; Drevets et al., 1997; Myers-Schulz & Koenigs, 2012; Rajkowska et al., 1999), including studies in adolescents and young adults (Ducharme et al., 2014; Reynolds et al., 2014), have shown that the frontal cortex, which plays a key role in emotion regulation, is affected by depression. Thus, our results are in line with earlier findings on the involvement of frontal cortex in depression, and support the idea that sleep regulation and depression might share common neurobiological mechanisms (Adrien, 2002; Chang et al., 2014; Taylor et al., 2005).

5.4. Role of *BDNF* Val66Met polymorphism in early life sleep, temperament and psychomotor development

Several functional polymorphisms in a number of candidate genes have been associated with early development of children's temperament and psychosocial maturation, often predisposing them to various maladaptive outcomes later in life (Caspi et al., 2002; Cicchetti et al., 2011; Cutuli et al., 2013; Holmboe et al., 2011). However, *BDNF* gene and its most common single nucleotide polymorphism (Val66Met) has not been studied in this regard. Since *BDNF* Val66Met polymorphism is related to neuroplasticity, and the expression of the *BDNF* Met allele is related to impaired BDNF secretion, it seems reasonable to assume that this polymorphism could be associated with early life development. Thus, we investigated *BDNF* Val66Met polymorphism in the sample of the Finnish birth cohort CHILD-SLEEP (N=1678) and studied its association with different parameters, including sleep, psychomotor development and temperament.

A total of 1119 infants (572 boys and 547 girls) at the age of 3 months and 994 (507 boys and 487 girls) at the age of 8 months were analyzed in this study (Table 8). The genotype frequencies of *BDNF* Val66Met polymorphism indicated on high prevalence of individuals with Val/Val genotype (~70%) and very low number of Met/Met individuals (~1%).

Table 8. Characteristics of the CHILD-SLEEP birth cohort participants

Variable	Descriptives at the age of 3 months	Descriptives at the age of 8 months
Sample size, n	1119	994
Gender, male, n (%)	572 (51.0)	507 (51.0)
Genotype frequency, n (%)		
Val/Val	796 (71.1)	711 (7.5)
Val/Met	308 (27.5)	271 (27.3)
Met/Met	15 (1.3)	12 (1.2)

Since previous findings have revealed significant differences in development of studied parameters in boys and girls (Gartstein & Rothbart, 2003; Martin et al., 1997), we also assessed the difference between genders in the traits of interest in the CHILD-SLEEP cohort.

At the age of 3 months, girls spent more time in sleep during the day (322.98±3.96 vs. 307.32±3.86 min, P=0.005; P_{Bonf}=0.045), had lower proportion of night sleep respective to TST (higher ratio: night sleep/TST) (P=0.046; P_{Bonf}>0.05) and less number of night awakenings (1.97±1.44 vs. 2.14±1.20, P=0.035; P_{Bonf}>0.05) than boys (Table 9). Regarding psychomotor development, boys had higher scores at gross motor development scale than girls (5.12±0.07 vs. 4.92±0.07, P=0.031; P_{Bonf}>0.05) (Table 9).

At the age of 8 months, we found a significant difference between boys and girls in two psychomotor characteristics (socioemotional and fine motor development) and in temperament scale, particularly in negative affectivity dimension of IBQ-R. Girls showed higher scores in the socioemotional (21.11±0.11 vs. 20.63±0.11, p=0.001, P_{Bonf}=0.016) and fine motor development (25.15±0.16 vs. 24.23±0.15, P=2.9x10^{-5}; P_{Bonf}=0.00046) scales than boys. Girls demonstrated higher scores than boys in negative affectivity scales of IBQ-R (3.07±0.03 vs. 2.95±0.03, P=0.008; P_{Bonf}>0.05 (Table 10). Similar gender differences in IBQ-R have been also reported previously in 6-month old infants (Martin et al., 1997) and in older children and adults (Gartstein & Rothbart, 2003). Thus, we can conclude that our sample was representative as a normative sample of children.

Taking into account these differences, gender was included as a factor in the subsequent analysis for association between *BDNF* polymorphism and above mentioned developmental parameters. However, we found no significant effect of Val66Met *BDNF* polymorphism on studied sleep parameters and psychomotor development of infants at the age of 3 months (P>0.05 for all traits, Table 9) or on sleep, psychomotor development and temperament at the age of 8 months (P>0.05 for all traits, Table 10).

To gain more information about sleep quality of infants at the age of 8 months, a subsample (N=57) of infants underwent polysomnographic recordings. We did not find significant differences in measured polysomnographic parameters (SEI and WASO) between boys and girls, and the analysis of association between *BDNF* Val66Met polymorphism and polysomnographic variables revealed no significant effect (P>0.05 for all parameters, Table 10).

Table 9. Association of *BDNF* gene polymorphism Val66Met with different traits of infants' development at the age of 3 months.

	Effect of gender		Effect of *BDNF* Val66Met polymorphism			Mean values for the *BDNF* genotypes								
	F	P (P_{Bonf})	F	P	P_{Bonf}	Val/Val	SE	N	Val/Met	SE	N	Met/Met	SE	N
Night sleep, h	0.605	0.437	0.549	0.577	1	9.04	0.05	746	9.15	0.09	293	9.18	0.40	15
Daytime sleep, h	7.998	0.005 (0.045)	1.445	0.236	1	5.22	0.05	727	5.29	0.09	289	5.80	0.40	15
TST, h	2.481	0.116	2.44	0.088	0.792	14.18	0.07	740	14.42	0.10	291	14.98	0.52	15
Night sleep/TST [a]	3.99	0.048 (0.432)	0.406	0.666	1	0.636	0.004	745	0.634	0.006	293	0.612	0.028	15
Night awakenings [a]	4.463	0.035 (0.315)	0.396	0.673	1	2.07	0.05	743	2.03	0.09	295	2.32	0.34	15
Vocalization	1.925	0.166	0.148	0.862	1	7.391	0.095	769	7.439	0.152	304	7.067	0.679	15
Socioemotional	0.043	0.836	1.226	0.294	1	12.596	0.105	769	12.384	0.168	304	11.667	0.755	15
Fine motor	1.51	0.219	0.143	0.867	1	12.500	0.129	769	12.557	0.206	304	12.067	0.927	15
Gross motor [a]	4.682	0.031 (0.279)	0.627	0.534	1	5.006	0.055	769	5.067	0.087	304	4.646	0.394	15

[a] – gender is included as an additional factor in the analysis of *BDNF* Val66Met polymorphism effect. TST: total sleep time, BDNF: brain-derived neurotrophic factor.

Table 10. Association of *BDNF* gene polymorphism Val66Met with different traits of infants' development at the age of 8 months.

	Effect of gender		Effect of *BDNF* Val66Met polymorphism			Mean values for the *BDNF* genotypes								
	F	P (P_{Bonf})	F	P	P_{Bonf}	Val/Val	SE	N	Val/Met	SE	N	Met/Met	SE	N
Night sleep, h	1.998	0.158	0.223	0.800	1	9.88	0.04	702	9.92	0.06	274	9.75	0.29	12
Daytime sleep, h	0.019	0.891	0.368	0.692	1	3.45	0.04	702	3.46	0.06	274	3.70	0.32	12
TST, h	2.258	0.133	0.456	0.634	1	13.31	0.05	705	13.38	0.08	271	13.15	0.36	12
Night sleep/TST	0.009	0.923	0.091	0.913	1	0.745	0.003	702	0.743	0.004	274	0.745	0.020	12
TST 3 mo - TST 8 mo, min [a]	4.76	0.029 (0.464)	1.176	0.309	1	51.45	4.41	664	61.31	7.34	255	92.78	33.63	12
Night awakenings	0.0003	0.985	0.160	0.852	1	2.10	0.07	628	2.10	0.10	253	1.82	0.50	11
Vocalization	0.743	0.389	0.325	0.722	1	13.535	0.130	711	13.359	0.212	271	13.449	1.002	12
Socioemotional [a]	10.196	0.001 (0.016)	0.082	0.921	1	20.881	0.089	711	20.829	0.145	271	20.997	0.687	12
Fine motor [a]	17.677	0.000029 (0.00046)	0.559	0.572	1	24.762	0.130	711	24.521	0.212	271	24.322	1.002	12
Gross motor	0.182	0.670	0.176	0.838	1	15.160	0.152	711	15.219	0.247	271	15.833	1.173	12
Positive affectivity	0.798	0.372	0.547	0.579	1	4.758	0.023	709	4.804	0.038	273	4.792	0.179	12
Negative affectivity [a]	7.098	0.008 (0.128)	0.606	0.546	1	3.014	0.027	709	3.002	0.044	273	3.236	0.208	12
Regulatory capacity	0.797	0.372	0.271	0.762	1	5.034	0.025	709	5.017	0.040	273	5.154	0.192	12
REM/NREM ratio	1.710	0.196	0.362	0.698	1	0.486	0.017	34	0.490	0.021	22	0.570	0.098	1
SEI	3.060	0.086	0.541	0.585	1	86.500	0.757	36	87.300	1.015	20	83.000	4.540	1
WASO	3.880	0.054	0.652	0.525	1	67.306	3.735	36	65.795	5.011	20	92.000	22.412	1

[a] – gender is included as an additional factor in the analysis of *BDNF* Val66Met polymorphism effect. TST: total sleep time, BDNF: brain-derived neurotrophic factor, REM: rapid eye movement sleep, NREM: non-rapid eye movement sleep, SEI: sleep efficiency index, WASO: wakefulness time after sleep onset.

From previous studies in adults it is known that *BDNF* Val66Met polymorphism is associated with anxiety/depression-related temperament traits, such as neuroticism (Jylha et al., 2009), but the results of these studies are inconsistent (Huennerkopf et al., 2007; Itoh et al., 2004). Some indicate higher levels of neuroticism in Val allele homozygous individuals than in Met carriers, while others failed to show such association (Tochigi et al., 2006; Wray et al., 2008); one study found the association in females, but not in males (Itoh et al., 2004). Moreover, a link between *BDNF* allelic variation and another temperament trait, extraversion, has been found (Terracciano et al., 2010). Furthermore, the association between *BDNF* genotype and adult depression in context of early life adverse experience has been shown as well (Buchmann et al., 2013; Liu, 2010; van Oostrom et al., 2012). However, our study failed to find an association between *BDNF* genotype and early life temperament or psychomotor development of infants.

Studies regarding the role of Val66Met polymorphism in sleep regulation are limited and performed mostly in adults. Val66Met polymorphism has been link with sleep efficiency: Val/Val genotype has been associated with higher SWA in baseline sleep and post sleep deprivation recovery sleep (Bachmann et al., 2012). Decreased spectral power in alpha and theta ranges were also observed in NREM sleep in Met allele carriers (Guindalini et al., 2014). In our study we found no significant association between Val66Met polymorphism and any of the studied sleep parameters based either on parent reports at the age of 3- and 8 months or on polysomnographic measures in a subsample of infants at the age of 8 months.

BDNF is one of the central regulators of early stages of nervous system maturation, and disturbances in its functions would have severe effects for the development (Binder & Scharfman, 2004; Cohen-Cory et al., 2010; Halepoto et al., 2014). It has been shown that in immature neurons, BDNF is involved in growth, differentiation, maturation and survival processes while in mature neurons it plays an important role in synaptic plasticity, augmentation of neurotransmission and regulation of receptor sensitivity (Calabrese et al., 2009; Gonzalez et al., 2016; Numakawa et al., 2010). Based on our results we can hypothesize that during critical periods of neuronal development *BDNF*

Val66Met polymorphism is not associated with development of infant's temperament, psychomotor maturation or sleep. However, later in life, the effects of the genetic variant may become evident, particularly in connection with traumatic childhood life events (Aguilera et al., 2009; Buchmann et al., 2013; Comasco et al., 2013; Gutierrez et al., 2015).

6. Methodological considerations

I. In the study on exon-specific *BDNF* expression and methylation, the most notable limitation was the small sample size. To estimate the impact of the data, Cohen's d effect sizes were used. In the present data set, group differences showed medium effect sizes. Hence, despite the low n, effect sizes reflect that the significant differences between the groups in the study are relevant. The results should, therefore, be confirmed in studies with larger sample size.

II. In the cross-fostering experiments both male and female rats were used for the study, and females demonstrated high variability in the brain *BDNF* expression. This variability may, at least partially, be due to different BDNF levels in the course of oestrous cycle (Luine & Frankfurt, 2013). This may have been reflected also in the variability of sleep stages, which was reported before (Fang & Fishbein, 1996). In the present study, we did not determine the phase of oestrous cycle because of potential stressful effect of this procedure, and thus potential influence on the results. In future, the studies should be repeated with evaluation of oestrous cycle in females. Another issue of this study is related to corticosterone (stress hormone in rats). Measurements of this hormone could have potentially clarified the stress level induced by the CF treatment.

III. In the study on sleep assessment in depressed adolescent boys, the limitation was the small sample size. The results should, therefore, be confirmed in studies with larger sample size. The study was performed using only male participants. However, it was shown previously that depressed girls may show no changes in SWA (Lopez et al., 2012) or even higher SWA (Frey et al., 2012) as compared to an age- and gender-matched control group. In future, it would be important to be able to have a sufficiently large sample that would enable examining both genders in an identical study setting.

IV. In the genetic study, the main issue is most notably the small number of infants with two Met alleles (Met/Met group) due to its rare occurrence in Caucasian populations. Thus, it is advisable to use bigger sample size to reach meaningful number of Met/Met representatives. On the other

hand, it is known that allele frequencies in *BDNF* Val66Met polymorphism differ by ethnicity (Pivac et al., 2009; Verhagen et al., 2010), so using Asian populations, where the frequency of the Met allele is much higher compared to Caucasians, can be of great interest.

7. Summary and Conclusions

1. During spontaneous sleep-wake cycle the modulation of *BDNF* transcription was at least partially regulated by brain site specific DNA methylation. The dynamics of sleep deprivation-induced *BDNF* up-regulation in basal forebrain and frontal cortex were transcript- and brain area-dependent.

2. Early life stress events leave a persistent molecular trace in the brain, as in the cross-fostering animal study was evidenced by decreased adenosine level in the basal forebrain. Only a tendency to lower *BDNF* gene expression was observed in the basal forebrain of cross-fostered rats. This molecular "scar" may act as a predisposing factor in development of psychopathologies later in life. Sleep, particularly REM sleep, appeared to be the most sensitive behavioural indicator of the early life stress events.

3. Adolescent depression was characterized by lower SWA power and its slower build up in the course of the first NREM episode together with more even pattern of SWA dissipation through the night in frontal area. These findings suggest that homeostatic sleep regulation may be impaired in adolescent depression. Since more pronounced changes were observed in frontal area it support the idea that sleep regulation and depression might share common neurobiological mechanisms.

4. Finally, we could not find any evidence for the involvement of the *BDNF* Val66Met polymorphism neither in early life development of psychomotor functions and sleep in 3- and 8-months old infants, nor in temperament in 8-months old infants.

Our results from animal and human adolescent studies (II, III) indicate that sleep is the most sensitive indicator of early life stress. Adenosine deficiency in the basal forebrain, as was shown in the animal model of early life stress (II), is one of the possible mechanisms leading to high vulnerability to sleep problems, and it is a permanent trace left in the brain by early life stress. In its turn, BDNF being highly involved in sleep regulation (I) may not be a best marker of early life stress (II) and its polymorphism is not associated with sleep and psychomotor development during early life stage (IV).

Assessing adenosine or BDNF levels is difficult in clinical practice and requires blood sampling. Moreover, it is uncertain how well peripheral measurements reflect central concentrations in the brain. In this regard, sleep can be used in clinical practice as a non-invasive biomarker for early diagnosis of depressive disorder as a consequence of early life stress. Early-onset sleep problems should be taken seriously in clinical practise, and access to psychiatric examination should be easily available, in order to address the possible psychiatric problems at their early phase.

8. Acknowledgements

I am grateful to all of those with whom I have had the pleasure to work during this and other related projects.

I want to express my deepest gratitude to my opponent, Associate Professor Janne Grønli, for kindly accepting the task of publicly examining my thesis. It is a big honour for me to be able to discuss my thesis with you.

I also want to thank the pre-examiners, Docents Soili Lehto and Tomi Rantamäki for kindly reviewing the thesis and for their constructive suggestions to improve the manuscript.

My deepest and most sincere gratitude goes to my mentor, Tarja Stenberg (Porkka-Heiskanen). Thank you for your continuous support during my Ph.D study, for your patience, motivation, and immense knowledge. Your guidance helped me in all the time of research and writing of this thesis. I could not have imagined having a better advisor and mentor for my Ph.D study.

I owe my deepest gratitude to Tiina Paunio for her invaluable support and introducing me to the world of genetics.

Besides my advisors, I would like to thank the members of my thesis committee: Professors Erkki Isometsä and Pertti Panula for their insightful comments and encouragement.

Thanks to all the current and former members of the Sleep Team Helsinki. Janneke Zant, for teaching me the methods and introducing to the Sleep Lab. Pirjo Saarelainen, for her excellent assistance in the experiments throughout my Ph.D study. Vilma Aho/Lammi, for her peer-support and encouragement, and for great company in our international conferences. Henna-Kaisa Wigren and Kirsi-Marja Zitting for their guidance and advices. Heidi Ikonen, for her good-humoured company. Anna Sofia Urrila, for involving me in her ADSLEEP study, it was a great pleasure to collaborate with you. Henna Lehtikuja and Eeva Palömäki, for their help in the experiments.

I would like to thank Stanislav Rozov, for the stimulating discussions that we had and for willingness to help in any situations, and Nina Karpova for amazing collaboration that we had. Special thanks to Jussi Virkkala for his help in dealing with technical aspects in ADSLEEP project.

All the co-authors of the papers, thank you for your invaluable comments and help with writing the manuscripts.

I have had the privilege of collaborating with amazing scientists and utilising valuable epidemiological dataset CHILD-SLEEP during my work as a visiting researcher at Tiina Paunio's group, at the National Institute for Health and Welfare (THL). I would like to thank all the member of this group, especially Katri Kantojärvi, Sonja Sulkava, Antti-Jussi Ämmälä, Paula Salo, Johanna Liuhanen for their help in the data analysis of the genetic part of this project.

Completing this work would have been all the more difficult were it not for the support of my dear friends. I thank my friends from Ukraine: Tanya, Evgeniya and Evgeniy, Vladimir, Lera, Vita and Lena for their continuous support in my efforts to come to Finland. I am enormously grateful for my friends from Finland: Julia, Misha, Maija, Chris, Ahmed, Sasha, Paula, Katya, Oxana, Boriss, Olga, Nicole who made my days brighter during the dark winters. Thank you for all the fun that we had and for all the fantastic moments that we shared together.

I would like to thank my family, parents Svitlana and Andriy Ventskovskiy and grandmother Lyudmila Korchagina for supporting me spiritually throughout writing this thesis and my life in general.

And finally, last but by no means least, I thank my loving and devoted husband, Andrea Santangeli, thank you for your encouragement and support, without you I would not have made it. And my daughter Mila for being the main encouragement for finishing this project on time.

Helsinki, February 2018

9. References

Adrien, J. (2002). Neurobiological bases for the relation between sleep and depression. *Sleep medicine reviews, 6*(5), 341-351. doi:10.1053/smrv.2001.0200

Aguilera, M., Arias, B., Wichers, M., Barrantes-Vidal, N., Moya, J., Villa, H., van Os, J., Ibanez, M. I., Ruiperez, M. A., Ortet, G., & Fananas, L. (2009). Early adversity and 5-HTT/BDNF genes: new evidence of gene-environment interactions on depressive symptoms in a general population. *Psychol Med, 39*(9), 1425-1432. doi:10.1017/S0033291709005248

Aid, T., Kazantseva, A., Piirsoo, M., Palm, K., & Timmusk, T. (2007). Mouse and rat BDNF gene structure and expression revisited. *J Neurosci Res, 85*(3), 525-535. doi:10.1002/jnr.21139

Akerstedt, T., Hume, K., Minors, D., & Waterhouse, J. (1997). Good sleep--its timing and physiological sleep characteristics. *Journal of sleep research, 6*(4), 221-229.

Alhola, P., & Polo-Kantola, P. (2007). Sleep deprivation: Impact on cognitive performance. *Neuropsychiatric disease and treatment, 3*(5), 553-567.

American Psychiatric Association. Diagnostic and Statistical Manual of Mental Disorders. (2013). (5 ed.). Arlington, VA.

Anisman, H., & Merali, Z. (2002). Cytokines, stress, and depressive illness. *Brain Behavior and Immunity, 16*(5), 513-524.

Archer, J. (1973). Tests for emotionality in rats and mice: a review. *Anim Behav, 21*(2), 205-235.

Argyropoulos, S. V., & Wilson, S. J. (2005). Sleep disturbances in depression and the effects of antidepressants. *International review of psychiatry, 17*(4), 237-245. doi:10.1080/09540260500104458

Armitage, R. (2007). Sleep and circadian rhythms in mood disorders. *Acta Psychiatr Scand Suppl*(433), 104-115. doi:10.1111/j.1600-0447.2007.00968.x

Armitage, R., Emslie, G. J., Hoffmann, R. F., Rintelmann, J., & Rush, A. J. (2001). Delta sleep EEG in depressed adolescent females and healthy controls. *Journal of affective disorders, 63*(1-3), 139-148.

Arnone, D., McIntosh, A. M., Ebmeier, K. P., Munafo, M. R., & Anderson, I. M. (2012). Magnetic resonance imaging studies in unipolar depression: Systematic review and meta-regression analyses. *European Neuropsychopharmacology, 22*(1), 1-16. doi:10.1016/j.euroneuro.2011.05.003

Arnow, B. A. (2004). Relationships between childhood maltreatment, adult health and psychiatric outcomes, and medical utilization. *Journal of Clinical Psychiatry, 65*, 10-15.

Audet, M. C., & Anisman, H. (2013). Interplay between pro-inflammatory cytokines and growth factors in depressive illnesses. *Frontiers in Cellular Neuroscience, 7*. doi:ARTN 68 10.3389/fncel.2013.00068

Augustinavicius, J. L., Zanjani, A., Zakzanis, K. K., & Shapiro, C. M. (2014). Polysomnographic features of early-onset depression: a meta-analysis. *Journal of affective disorders, 158*, 11-18. doi:10.1016/j.jad.2013.12.009

Autry, A. E., & Monteggia, L. M. (2012). Brain-derived neurotrophic factor and neuropsychiatric disorders. *Pharmacol Rev, 64*(2), 238-258. doi:10.1124/pr.111.005108

Azzi, A., Dallmann, R., Casserly, A., Rehrauer, H., Patrignani, A., Maier, B., Kramer, A., & Brown, S. A. (2014). Circadian behavior is light-reprogrammed by plastic DNA methylation. *Nat Neurosci, 17*(3), 377-382. doi:10.1038/nn.3651

Bachmann, V., Klein, C., Bodenmann, S., Schafer, N., Berger, W., Brugger, P., & Landolt, H. P. (2012). The BDNF Val66Met Polymorphism Modulates Sleep Intensity: EEG Frequency- and State-Specificity. *Sleep, 35*(3), 335-344. doi:10.5665/sleep.1690

Baker, F. C., Turlington, S. R., Hoffman, L. R., Mayer, B. Z., Wagstaff, A. E., & Colrain, I. M. (2011). Developmental Changes in the Sleep Electroencephalogram of Adolescent Boys and Girls. *Sleep, 34*, A293-A293.

Barbazanges, A., Vallee, M., Mayo, W., Day, J., Simon, H., Le Moal, M., & Maccari, S. (1996). Early and later adoptions have different long-term effects on male rat offspring. *J Neurosci, 16*(23), 7783-7790.

Barros, V. G., Berger, M. A., Martijena, I. D., Sarchi, M. I., Perez, A. A., Molina, V. A., Tarazi, F. I., & Antonelli, M. C. (2004). Early adoption modifies the effects of prenatal stress on dopamine and glutamate receptors in adult rat brain. *J Neurosci Res, 76*(4), 488-496. doi:10.1002/jnr.20119

Basheer, R., Strecker, R. E., Thakkar, M. M., & McCarley, R. W. (2004). Adenosine and sleep-wake regulation. *Progress in Neurobiology, 73*(6), 379-396. doi:10.1016/j.pneurobio.2004.06.004

Beck, A. T., Ward, C. H., Mendelson, M., Mock, J., & Erbaugh, J. (1961). An inventory for measuring depression. *Archives of general psychiatry, 4*, 561-571.

Belmaker, R. H., & Agam, G. (2008). Major depressive disorder. *N Engl J Med, 358*(1), 55-68. doi:10.1056/NEJMra073096

Benedetti, F., & Colombo, C. (2011). Sleep deprivation in mood disorders. *Neuropsychobiology, 64*(3), 141-151. doi:10.1159/000328947

Berchtold, N. C., Oliff, H. S., Isackson, P., & Cotman, C. W. (1999). Hippocampal BDNF mRNA shows a diurnal regulation, primarily in the exon III transcript. *Molecular Brain Research, 71*(1), 11-22. doi:Doi 10.1016/S0169-328x(99)00137-0

Bhagya, V., Srikumar, B. N., Raju, T. R., & Shankaranarayana Rao, B. S. (2008). Neonatal clomipramine induced endogenous depression in rats is associated with learning impairment in adulthood. *Behav Brain Res, 187*(1), 190-194. doi:10.1016/j.bbr.2007.08.020

Bhattacharya, S. K., Ramchandani, S., Cervoni, N., & Szyf, M. (1999). A mammalian protein with specific demethylase activity for mCpG DNA. *Nature, 397*(6720), 579-583. doi:10.1038/17533

Binder, D. K., & Scharfman, H. E. (2004). Brain-derived neurotrophic factor. *Growth Factors, 22*(3), 123-131.

Bird, A. (2007). Perceptions of epigenetics. *Nature, 447*(7143), 396-398. doi:10.1038/nature05913

Borbely, A. A. (1982). A two process model of sleep regulation. *Human neurobiology, 1*(3), 195-204.

Borbely, A. A. (1987). The S-deficiency hypothesis of depression and the two-process model of sleep regulation. *Pharmacopsychiatry, 20*(1), 23-29.

Borbely, A. A., & Achermann, P. (1999). Sleep homeostasis and models of sleep regulation. *Journal of biological rhythms, 14*(6), 557-568.

Borbely, A. A., Baumann, F., Brandeis, D., Strauch, I., & Lehmann, D. (1981). Sleep deprivation: effect on sleep stages and EEG power density in man. *Electroencephalography and clinical neurophysiology, 51*(5), 483-495.

Borbely, A. A., Tobler, I., Loepfe, M., Kupfer, D. J., Ulrich, R. F., Grochocinski, V., Doman, J., & Matthews, G. (1984). All-Night Spectral-Analysis of the Sleep Eeg in Untreated Depressives and Normal Controls. *Psychiatry research, 12*(1), 27-33. doi:Doi 10.1016/0165-1781(84)90135-5

Borbely, A. A., & Wirz-Justice, A. (1982). Sleep, sleep deprivation and depression. A hypothesis derived from a model of sleep regulation. *Human neurobiology, 1*(3), 205-210.

Bradley, R. G., Binder, E. B., Epstein, M. P., Tang, Y., Nair, H. P., Liu, W., Gillespie, C. F., Berg, T., Evces, M., Newport, D. J., Stowe, Z. N., Heim, C. M., Nemeroff, C. B., Schwartz, A., Cubells, J. F., & Ressler, K. J. (2008). Influence of child abuse on adult depression - Moderation by the corticotropin-releasing hormone receptor gene. *Archives of general psychiatry, 65*(2), 190-200. doi:DOI 10.1001/archgenpsychiatry.2007.26

Brand, S., & Kirov, R. (2011). Sleep and its importance in adolescence and in common adolescent somatic and psychiatric conditions. *International journal of general medicine, 4*, 425-442. doi:10.2147/IJGM.S11557

Breslau, N., Roth, T., Rosenthal, L., & Andreski, P. (1996). Sleep disturbance and psychiatric disorders: A longitudinal epidemiological study of young adults. *Biological psychiatry, 39*(6), 411-418. doi:Doi 10.1016/0006-3223(95)00188-3

Brigitta, B. (2002). Pathophysiology of depression and mechanisms of treatment. *Dialogues in clinical neuroscience, 4*(1), 7-20.

Bromet, E., Andrade, L. H., Hwang, I., Sampson, N. A., Alonso, J., de Girolamo, G., de Graaf, R., Demyttenaere, K., Hu, C., Iwata, N., Karam, A. N., Kaur, J., Kostyuchenko, S., Lepine, J. P., Levinson, D., Matschinger, H., Mora, M. E., Browne, M. O., Posada-Villa, J., Viana, M. C., Williams, D. R., & Kessler, R. C. (2011). Cross-national epidemiology of DSM-IV major depressive episode. *BMC Med, 9*, 90. doi:10.1186/1741-7015-9-90

Buchmann, A., Ringli, M., Kurth, S., Schaerer, M., Geiger, A., Jenni, O. G., & Huber, R. (2011). EEG Sleep Slow-Wave Activity as a Mirror of Cortical Maturation. *Cereb Cortex, 21*(3), 607-615. doi:10.1093/cercor/bhq129

Buchmann, A. F., Hellweg, R., Rietschel, M., Treutlein, J., Witt, S. H., Zimmermann, U. S., Schmidt, M. H., Esser, G., Banaschewski, T., Laucht, M., & Deuschle, M. (2013). BDNF Val 66 Met and 5-HTTLPR genotype moderate the impact of early psychosocial adversity on plasma brain-derived neurotrophic factor and depressive symptoms: a prospective study. *Eur Neuropsychopharmacol, 23*(8), 902-909. doi:10.1016/j.euroneuro.2012.09.003

Burke, K. C., Burke, J. D., Regier, D. A., & Rae, D. S. (1990). Age at Onset of Selected Mental-Disorders in 5 Community Populations. *Archives of general psychiatry, 47*(6), 511-518.

Cajochen, C., Foy, R., & Dijk, D. J. (1999). Frontal predominance of a relative increase in sleep delta and theta EEG activity after sleep loss in humans. *Sleep Res Online, 2*(3), 65-69.

Calabrese, F., Molteni, R., Racagni, G., & Riva, M. A. (2009). Neuronal plasticity: a link between stress and mood disorders. *Psychoneuroendocrinology, 34 Suppl 1*, S208-216. doi:10.1016/j.psyneuen.2009.05.014

Campbell, I. G., & Feinberg, I. (2009). Longitudinal trajectories of non-rapid eye movement delta and theta EEG as indicators of adolescent brain maturation. *Proceedings of the National Academy of Sciences of the United States of America, 106*(13), 5177-5180. doi:10.1073/pnas.0812947106

Caspi, A., McClay, J., Moffitt, T. E., Mill, J., Martin, J., Craig, I. W., Taylor, A., & Poulton, R. (2002). Role of genotype in the cycle of violence in maltreated children. *Science, 297*(5582), 851-854. doi:DOI 10.1126/science.1072290

Caspi, A., Sugden, K., Moffitt, T. E., Taylor, A., Craig, I. W., Harrington, H., McClay, J., Mill, J., Martin, J., Braithwaite, A., & Poulton, R. (2003). Influence of life stress on depression: Moderation by a polymorphism in the 5-HTT gene. *Science, 301*(5631), 386-389. doi:DOI 10.1126/science.1083968

Castagne, V., Moser, P., & Porsolt, R. D. (2009). Behavioral Assessment of Antidepressant Activity in Rodents. In J. J. Buccafusco (Ed.), *Methods of Behavior Analysis in Neuroscience* (2nd ed.). Boca Raton (FL).

Castren, E., & Rantamaki, T. (2010). The role of BDNF and its receptors in depression and antidepressant drug action: Reactivation of developmental plasticity. *Dev Neurobiol, 70*(5), 289-297. doi:10.1002/dneu.20758

Castren, E., Voikar, V., & Rantamaki, T. (2007). Role of neurotrophic factors in depression. *Curr Opin Pharmacol, 7*(1), 18-21. doi:10.1016/j.coph.2006.08.009

Chacon-Fernandez, P., Sauberli, K., Colzani, M., Moreau, T., Ghevaert, C., & Barde, Y. A. (2016). Brain-derived Neurotrophic Factor in Megakaryocytes. *J Biol Chem, 291*(19), 9872-9881. doi:10.1074/jbc.M116.720029

Chang, C. H., Chen, M. C., Qiu, M. H., & Lu, J. (2014). Ventromedial prefrontal cortex regulates depressive-like behavior and rapid eye movement sleep in the rat. *Neuropharmacology, 86*, 125-132. doi:10.1016/j.neuropharm.2014.07.005

Chao, H. M., Sakai, R. R., Ma, L. Y., & McEwen, B. S. (1998). Adrenal steroid regulation of neurotrophic factor expression in the rat hippocampus. *Endocrinology, 139*(7), 3112-3118. doi:DOI 10.1210/en.139.7.3112

Chapman, D. P., Wheaton, A. G., Anda, R. F., Croft, J. B., Edwards, V. J., Liu, Y., Sturgis, S. L., & Perry, G. S. (2011). Adverse childhood experiences and sleep disturbances in adults. *Sleep Medicine, 12*(8), 773-779. doi:10.1016/j.sleep.2011.03.013

Chen, B., Dowlatshahi, D., MacQueen, G. M., Wang, J. F., & Young, L. T. (2001). Increased hippocampal BDNF immunoreactivity in subjects treated with antidepressant medication. *Biological psychiatry, 50*(4), 260-265.

Chen, K. W., & Chen, L. Y. (2017). Epigenetic Regulation of BDNF Gene during Development and Diseases. *International Journal of Molecular Sciences, 18*(3). doi:ARTN 571 10.3390/ijms18030571

Chen, Z. Y., Jing, D., Bath, K. G., Ieraci, A., Khan, T., Siao, C. J., Herrera, D. G., Toth, M., Yang, C., McEwen, B. S., Hempstead, B. L., & Lee, F. S. (2006). Genetic variant BDNF

(Val66Met) polymorphism alters anxiety-related behavior. *Science, 314*(5796), 140-143. doi:10.1126/science.1129663

Chikahisa, S., & Sei, H. (2011). The role of ATP in sleep regulation. *Front Neurol, 2*, 87. doi:10.3389/fneur.2011.00087

Cicchetti, D., Rogosch, F. A., & Toth, S. L. (2011). The effects of child maltreatment and polymorphisms of the serotonin transporter and dopamine D4 receptor genes on infant attachment and intervention efficacy. *Development and Psychopathology, 23*(2), 357-372. doi:10.1017/S0954579411000113

Cirelli, C. (2009). The genetic and molecular regulation of sleep: from fruit flies to humans. *Nature Reviews Neuroscience, 10*(8), 549-560. doi:10.1038/nrn2683

Cirelli, C., Faraguna, U., & Tononi, G. (2006). Changes in brain gene expression after long-term sleep deprivation. *Journal of Neurochemistry, 98*(5), 1632-1645. doi:10.1111/j.1459-4159.2006.04058.x

Cirelli, C., Gutierrez, C. M., & Tononi, G. (2004). Extensive and divergent effects of sleep and wakefulness on brain gene expression. *Neuron, 41*(1), 35-43. doi:Doi 10.1016/S0896-6273(03)00814-6

Cirelli, C., & Tononi, G. (2000a). Differential expression of plasticity-related genes in waking and sleep and their regulation by the noradrenergic system. *Journal of Neuroscience, 20*(24), 9187-9194.

Cirelli, C., & Tononi, G. (2000b). Gene expression in the brain across the sleep-waking cycle. *Brain research, 885*(2), 303-321.

Cirelli, C., & Tononi, G. (2008). Is sleep essential? *Plos Biology, 6*(8), 1605-1611. doi:ARTN e216 10.1371/journal.pbio.0060216

Cohen-Cory, S., Kidane, A. H., Shirkey, N. J., & Marshak, S. (2010). Brain-derived neurotrophic factor and the development of structural neuronal connectivity. *Dev Neurobiol, 70*(5), 271-288. doi:10.1002/dneu.20774

Colrain, I. M., & Baker, F. C. (2011). Changes in Sleep as a Function of Adolescent Development. *Neuropsychology Review, 21*(1), 5-21. doi:10.1007/s11065-010-9155-5

Comasco, E., Aslund, C., Oreland, L., & Nilsson, K. W. (2013). Three-way interaction effect of 5-HTTLPR, BDNF Val66Met, and childhood adversity on depression: A replication study. *European Neuropsychopharmacology, 23*(10), 1300-1306. doi:10.1016/j.euroneuro.2013.01.010

Cooney, R. E., Joormann, J., Eugene, F., Dennis, E. L., & Gotlib, I. H. (2010). Neural correlates of rumination in depression. *Cognitive Affective & Behavioral Neuroscience, 10*(4), 470-478. doi:10.3758/Cabn.10.4.470

Costello, E. J., Erkanli, A., & Angold, A. (2006). Is there an epidemic of child or adolescent depression? *Journal of Child Psychology and Psychiatry, 47*(12), 1263-1271. doi:10.1111/j.1469-7610.2006.01682.x

Costello, E. J., Mustillo, S., Erkanli, A., Keeler, G., & Angold, A. (2003). Prevalence and development of psychiatric disorders in childhood and adolescence. *Archives of general psychiatry, 60*(8), 837-844. doi:DOI 10.1001/archpsyc.60.8.837

Crick, F., & Mitchison, G. (1983). The function of dream sleep. *Nature, 304*(5922), 111-114.

Cunha, R. A. (2005). Neuroprotection by adenosine in the brain: From A(1) receptor activation to A (2A) receptor blockade. *Purinergic Signal, 1*(2), 111-134. doi:10.1007/s11302-005-0649-1

Cutuli, J. J., Raby, K. L., Cicchetti, D., Englund, M. M., & Egeland, B. (2013). Contributions of maltreatment and serotonin transporter genotype to depression in childhood, adolescence, and early adulthood. *Journal of affective disorders, 149*(1-3), 30-37. doi:10.1016/j.jad.2012.08.011

Dagyte, G., Den Boer, J. A., & Trentani, A. (2011). The cholinergic system and depression. *Behav Brain Res, 221*(2), 574-582. doi:10.1016/j.bbr.2010.02.023

Dahl, R. E., Puig-Antich, J., Ryan, N. D., Nelson, B., Dachille, S., Cunningham, S. L., Trubnick, L., & Klepper, T. P. (1990). EEG sleep in adolescents with major depression: the role of suicidality and inpatient status. *Journal of affective disorders, 19*(1), 63-75.

Darnaudery, M., Koehl, M., Barbazanges, A., Cabib, S., Le Moal, M., & Maccari, S. (2004). Early and later adoptions differently modify mother-pup interactions. *Behav Neurosci, 118*(3), 590-596. doi:10.1037/0735-7044.118.3.590

de Kloet, E. R., DeRijk, R. H., & Meijer, O. C. (2007). Therapy Insight: is there an imbalanced response of mineralocorticoid and glucocorticoid receptors in depression? *Nature Clinical Practice Endocrinology & Metabolism, 3*(2), 168-179. doi:10.1038/ncpendmet0403

de Vivo, L., Bellesi, M., Marshall, W., Bushong, E. A., Ellisman, M. H., Tononi, G., & Cirelli, C. (2017). Ultrastructural evidence for synaptic scaling across the wake/sleep cycle. *Science, 355*(6324), 507-510. doi:10.1126/science.aah5982

de Weerd, A. W., & van den Bossche, R. A. (2003). The development of sleep during the first months of life. *Sleep medicine reviews, 7*(2), 179-191.

Dijk, D. J. (2009). Regulation and functional correlates of slow wave sleep. *J Clin Sleep Med, 5*(2 Suppl), S6-15.

Dijk, D. J., Brunner, D. P., & Borbely, A. A. (1990). Time Course of Eeg Power-Density during Long Sleep in Humans. *American Journal of Physiology, 258*(3), R650-R661.

Dougherty, L. R., Klein, D. N., & Davila, J. (2004). A growth curve analysis of the course of dysthymic disorder: The effects of chronic stress and moderation by adverse parent-child relationships and family history. *J Consult Clin Psychol, 72*(6), 1012-1021. doi:10.1037/0022-006x.72.6.1012

Drevets, W. C., Price, J. L., Simpson, J. R., Todd, R. D., Reich, T., Vannier, M., & Raichle, M. E. (1997). Subgenual prefrontal cortex abnormalities in mood disorders. *Nature, 386*(6627), 824-827. doi:DOI 10.1038/386824a0

Ducharme, S., Albaugh, M. D., Hudziak, J. J., Botteron, K. N., Nguyen, T. V., Truong, C., Evans, A. C., Karama, S., & Brain Development Cooperative, G. (2014). Anxious/depressed symptoms are linked to right ventromedial prefrontal cortical thickness maturation in healthy children and young adults. *Cerebral cortex, 24*(11), 2941-2950. doi:10.1093/cercor/bht151

Duman, C. H., Schlesinger, L., Kodama, M., Russell, D. S., & Duman, R. S. (2007). A role for MAP kinase signaling in behavioral models of depression and antidepressant treatment. *Biological psychiatry, 61*(5), 661-670. doi:10.1016/j.biopsych.2006.05.047

Duman, R. S. (2002). Pathophysiology of depression: the concept of synaptic plasticity. *Eur Psychiatry, 17 Suppl 3*, 306-310.

Duman, R. S. (2004). Depression: a case of neuronal life and death? *Biological psychiatry, 56*(3), 140-145. doi:10.1016/j.biopsych.2004.02.033

Duman, R. S., & Monteggia, L. M. (2006). A neurotrophic model for stress-related mood disorders. *Biological psychiatry, 59*(12), 1116-1127. doi:10.1016/j.biopsych.2006.02.013

Dunwiddie, T. V., & Masino, S. A. (2001). The role and regulation of adenosine in the central nervous system. *Annu Rev Neurosci, 24*, 31-55. doi:10.1146/annurev.neuro.24.1.31

Dunwiddie, T. V., & Worth, T. (1982). Sedative and Anticonvulsant Effects of Adenosine-Analogs in Mouse and Rat. *Journal of Pharmacology and Experimental Therapeutics, 220*(1), 70-76.

Dwivedi, Y. (2009). Brain-derived neurotrophic factor: role in depression and suicide. *Neuropsychiatric disease and treatment, 5*, 433-449.

Dwivedi, Y., Rizavi, H. S., Conley, R. R., Roberts, R. C., Tamminga, C. A., & Pandey, G. N. (2003). Altered gene expression of brain-derived neurotrophic factor and receptor tyrosine kinase B in postmortem brain of suicide subjects. *Archives of general psychiatry, 60*(8), 804-815. doi:10.1001/archpsyc.60.8.804

Dwivedi, Y., Rizavi, H. S., & Pandey, G. N. (2006). Antidepressants reverse corticosterone-mediated decrease in brain-derived neurotrophic factor expression: differential regulation of specific exons by antidepressants and corticosterone. *Neuroscience, 139*(3), 1017-1029. doi:10.1016/j.neuroscience.2005.12.058

Edwards, V. J., Holden, G. W., Felitti, V. J., & Anda, R. F. (2003). Relationship between multiple forms of childhood maltreatment and adult mental health in community respondents: Results from the adverse childhood experiences study. *American Journal of Psychiatry, 160*(8), 1453-1460. doi:DOI 10.1176/appi.ajp.160.8.1453

Egan, M. F., Kojima, M., Callicott, J. H., Goldberg, T. E., Kolachana, B. S., Bertolino, A., Zaitsev, E., Gold, B., Goldman, D., Dean, M., Lu, B., & Weinberger, D. R. (2003). The BDNF val66met polymorphism affects activity-dependent secretion of BDNF and human memory and hippocampal function. *Cell, 112*(2), 257-269.

Ehlers, C. L., Havstad, J. W., & Kupfer, D. J. (1996). Estimation of the time course of slow-wave sleep over the night in depressed patients: effects of clomipramine and clinical response. *Biological psychiatry, 39*(3), 171-181. doi:10.1016/0006-3223(95)00139-5

El Yacoubi, M., Costentin, J., & Vaugeois, J. M. (2003). Adenosine A2A receptors and depression. *Neurology, 61*(11 Suppl 6), S82-87.

Fang, J. D., & Fishbein, W. (1996). Sex differences in paradoxical sleep: Influences of estrus cycle and ovariectomy. *Brain research, 734*(1-2), 275-285. doi:Doi 10.1016/S0006-8993(96)00652-X

Faraguna, U., Vyazovskiy, V. V., Nelson, A. B., Tononi, G., & Cirelli, C. (2008). A causal role for brain-derived neurotrophic factor in the homeostatic regulation of sleep. *J Neurosci, 28*(15), 4088-4095. doi:10.1523/JNEUROSCI.5510-07.2008

Feinberg, I., & Campbell, I. G. (2010). Sleep EEG changes during adolescence: An index of a fundamental brain reorganization. *Brain and Cognition, 72*(1), 56-65. doi:10.1016/j.bandc.2009.09.008

Feinberg, I., Higgins, L. M., Khaw, W. Y., & Campbell, I. G. (2006). The adolescent decline of NREM delta, an indicator of brain maturation, is linked to age and sex but not to pubertal stage. *American Journal of Physiology-Regulatory Integrative and Comparative Physiology, 291*(6), R1724-R1729. doi:10.1152/ajpregu.00293.2006

Feinberg, I., Thode, H. C., Jr., Chugani, H. T., & March, J. D. (1990). Gamma distribution model describes maturational curves for delta wave amplitude, cortical metabolic rate and synaptic density. *J Theor Biol, 142*(2), 149-161.

Feldberg, W., & Sherwood, S. L. (1954). Injections of Drugs into the Lateral Ventricle of the Cat. *Journal of Physiology-London, 123*(1), 148-167.

Fleming, J. A. (1994). REM sleep abnormalities and psychiatry. *J Psychiatry Neurosci, 19*(5), 335-344.

Fox, K., & Stryker, M. (2017). Integrating Hebbian and homeostatic plasticity: introduction. *Philos Trans R Soc Lond B Biol Sci, 372*(1715). doi:10.1098/rstb.2016.0413

Frank, M. G. (2012). Erasing Synapses in Sleep: Is It Time to Be SHY? *Neural Plasticity*. doi:Artn 264378 10.1155/2012/264378

Frank, M. G. (2013). Why I Am Not SHY: A Reply to Tononi and Cirelli. *Neural Plasticity*. doi:Artn 394946 10.1155/2013/394946

Frank, M. G., & Heller, H. C. (1997). Development of REM and slow wave sleep in the rat. *American Journal of Physiology-Regulatory Integrative and Comparative Physiology, 272*(6), R1792-R1799.

Franken, P., Dijk, D. J., Tobler, I., & Borbely, A. A. (1991a). Sleep deprivation in rats: effects on EEG power spectra, vigilance states, and cortical temperature. *Am J Physiol, 261*(1 Pt 2), R198-208.

Franken, P., Tobler, I., & Borbely, A. A. (1991b). Sleep homeostasis in the rat: simulation of the time course of EEG slow-wave activity. *Neurosci Lett, 130*(2), 141-144.

Frazer, A., & Benmansour, S. (2002). Delayed pharmacological effects of antidepressants. *Molecular psychiatry, 7 Suppl 1*, S23-28. doi:10.1038/sj.mp.4001015

Frazer, A., & Morilak, D. A. (2005). What should animal models of depression model? *Neurosci Biobehav Rev, 29*(4-5), 515-523. doi:10.1016/j.neubiorev.2005.03.006

Fredholm, B. B. (2010). Adenosine receptors as drug targets. *Experimental Cell Research, 316*(8), 1284-1288. doi:10.1016/j.yexcr.2010.02.004

Fredholm, B. B., Battig, K., Holmen, J., Nehlig, A., & Zvartau, E. E. (1999). Actions of caffeine in the brain with special reference to factors that contribute to its widespread use. *Pharmacological Reviews, 51*(1), 83-133.

Frey, S., Birchler-Pedross, A., Hofstetter, M., Brunner, P., Gotz, T., Munch, M., Blatter, K., Knoblauch, V., Wirz-Justice, A., & Cajochen, C. (2012). Young women with major depression live on higher homeostatic sleep pressure than healthy controls. *Chronobiol Int, 29*(3), 278-294. doi:10.3109/07420528.2012.656163

Frielingsdorf, H., Bath, K. G., Soliman, F., Difede, J., Casey, B. J., & Lee, F. S. (2010). Variant brain-derived neurotrophic factor Val66Met endophenotypes: implications for posttraumatic stress disorder. *Ann N Y Acad Sci, 1208*, 150-157. doi:10.1111/j.1749-6632.2010.05722.x

Fuchs, E., & Fliugge, G. (2006). Experimental animal models for the simulation of depression and anxiety. *Dialogues in clinical neuroscience, 8*(3), 323-333.

Fujihara, H., Sei, H., Morita, Y., Ueta, Y., & Morita, K. (2003). Short-term sleep disturbance enhances brain-derived neurotrophic factor gene expression in rat hippocampus by acting as internal stressor. *Journal of Molecular Neuroscience, 21*(3), 223-231. doi:Doi 10.1385/Jmn:21:3:223

Gampel, S., & Nomura, Y. (2014). Short and long-term effects of compromised birth weight, head circumference, and Apgar scores on neuropsychological development. *J Psychol Abnorm Child, 3*(127), 1-8. doi:10.4172/2329-9525.1000127

Gartstein, M. A., & Rothbart, M. K. (2003). Studying infant temperament via the Revised Infant Behavior Questionnaire. *Infant Behavior & Development, 26*(1), 64-86. doi:Pii S0163-6383(02)00169-8 Doi 10.1016/S0163-6383(02)00169-8

Gass, N., Ollila, H. M., Utge, S., Partonen, T., Kronholm, E., Pirkola, S., Suhonen, J., Silander, K., Porkka-Heiskanen, T., & Paunio, T. (2010). Contribution of adenosine related genes to the risk of depression with disturbed sleep. *Journal of affective disorders, 126*(1-2), 134-139. doi:10.1016/j.jad.2010.03.009

Gass, N., Porkka-Heiskanen, T., & Kalinchuk, A. V. (2009). The role of the basal forebrain adenosine receptors in sleep homeostasis. *Neuroreport, 20*(11), 1013-1018. doi:10.1097/WNR.0b013e32832d5859

Gatt, J. M., Nemeroff, C. B., Dobson-Stone, C., Paul, R. H., Bryant, R. A., Schofield, P. R., Gordon, E., Kemp, A. H., & Williams, L. M. (2009). Interactions between BDNF Val66Met polymorphism and early life stress predict brain and arousal pathways to syndromal depression and anxiety. *Molecular psychiatry, 14*(7), 681-695. doi:10.1038/mp.2008.143

Germain, A., & Kupfer, D. J. (2008). Circadian rhythm disturbances in depression. *Hum Psychopharmacol, 23*(7), 571-585. doi:10.1002/hup.964

Giedke, H., & Schwarzler, F. (2002). Therapeutic use of sleep deprivation in depression. *Sleep medicine reviews, 6*(5), 361-377. doi:10.1053/smrv.2002.0235

Giese, M., Beck, J., Brand, S., Muheim, F., Hemmeter, U., Hatzinger, M., Holsboer-Trachsler, E., & Eckert, A. (2014a). Fast BDNF serum level increase and diurnal BDNF oscillations are associated with therapeutic response after partial sleep deprivation. *Journal of Psychiatric Research, 59*, 1-7. doi:10.1016/j.jpsychires.2014.09.005

Giese, M., Unternaehrer, E., Brand, S., Calabrese, P., Holsboer-Trachsler, E., & Eckert, A. (2013). The Interplay of Stress and Sleep Impacts BDNF Level. *PLoS One, 8*(10). doi:ARTN e76050 10.1371/journal.pone.0076050

Giese, M., Unternahrer, E., Huttig, H., Beck, J., Brand, S., Calabrese, P., Holsboer-Trachsler, E., & Eckert, A. (2014b). BDNF: an indicator of insomnia? *Molecular psychiatry, 19*(2), 151-152. doi:10.1038/mp.2013.10

Gilestro, G. F., Tononi, G., & Cirelli, C. (2009). Widespread Changes in Synaptic Markers as a Function of Sleep and Wakefulness in Drosophila. *Science, 324*(5923), 109-112. doi:10.1126/science.1166673

Giuditta, A., Ambrosini, M. V., Montagnese, P., Mandile, P., Cotugno, M., Grassi Zucconi, G., & Vescia, S. (1995). The sequential hypothesis of the function of sleep. *Behav Brain Res, 69*(1-2), 157-166.

Goetz, R. R., Puig-Antich, J., Ryan, N., Rabinovich, H., Ambrosini, P. J., Nelson, B., & Krawiec, V. (1987). Electroencephalographic sleep of adolescents with major depression and normal controls. *Archives of general psychiatry, 44*(1), 61-68.

Gold, P. W., & Chrousos, G. P. (1999). The endocrinology of melancholic and atypical depression: Relation to neurocircuitry and somatic consequences. *Proceedings of the Association of American Physicians, 111*(1), 22-34. doi:DOI 10.1046/j.1525-1381.1999.09423.x

Gonul, A. S., Akdeniz, F., Taneli, F., Donat, O., Eker, C., & Vahip, S. (2005). Effect of treatment on serum brain-derived neurotrophic factor levels in depressed patients. *Eur Arch Psychiatry Clin Neurosci, 255*(6), 381-386. doi:10.1007/s00406-005-0578-6

Gonzalez, A., Moya-Alvarado, G., Gonzalez-Billaut, C., & Bronfman, F. C. (2016). Cellular and molecular mechanisms regulating neuronal growth by brain-derived neurotrophic factor. *Cytoskeleton (Hoboken), 73*(10), 612-628. doi:10.1002/cm.21312

Gorgulu, Y., & Caliyurt, O. (2009). Rapid antidepressant effects of sleep deprivation therapy correlates with serum BDNF changes in major depression. *Brain Res Bull, 80*(3), 158-162. doi:10.1016/j.brainresbull.2009.06.016

Goshen, I., Kreisel, T., Ben-Menachem-Zidon, O., Licht, T., Weidenfeld, J., Ben-Hur, T., & Yirmiya, R. (2008). Brain interleukin-1 mediates chronic stress-induced depression in mice via adrenocortical activation and hippocampal neurogenesis suppression. *Molecular psychiatry, 13*(7), 717-728. doi:10.1038/sj.mp.4002055

Gratacos, M., Gonzalez, J. R., Mercader, J. M., de Cid, R., Urretavizcaya, M., & Estivill, X. (2007). Brain-derived neurotrophic factor Val66Met and psychiatric disorders: Meta-analysis of case-control studies confirm association to substance-related disorders, eating disorders, and schizophrenia. *Biological psychiatry, 61*(7), 911-922. doi:10.1016/j.biopsych.2006.08.025

Greden, J. F., Fontaine, P., Lubetsky, M., & Chamberlin, K. (1978). Anxiety and Depression Associated with Caffeinism among Psychiatric-Inpatients. *American Journal of Psychiatry, 135*(8), 963-966.

Greenfield, E. A., Lee, C., Friedman, E. L., & Springer, K. W. (2011). Childhood Abuse as a Risk Factor for Sleep Problems in Adulthood: Evidence from a U.S. National Study. *Annals of Behavioral Medicine, 42*(2), 245-256. doi:10.1007/s12160-011-9285-x

Grippo, A. J., Sullivan, N. R., Damjanoska, K. J., Crane, J. W., Carrasco, G. A., Shi, J., Chen, Z., Garcia, F., Muma, N. A., & Van de Kar, L. D. (2005). Chronic mild stress induces

behavioral and physiological changes, and may alter serotonin 1A receptor function, in male and cycling female rats. *Psychopharmacology (Berl), 179*(4), 769-780. doi:10.1007/s00213-004-2103-4

Guindalini, C., Mazzotti, D. R., Castro, L. S., D'Aurea, C. V. R., Andersen, M. L., Poyares, D., Bittencourt, L. R. A., & Tufik, S. (2014). Brain-Derived Neurotrophic Factor Gene Polymorphism Predicts Interindividual Variation in the Sleep Electroencephalogram. *J Neurosci Res, 92*(8), 1018-1023. doi:10.1002/jnr.23380

Gutierrez, B., Bellon, J. A., Rivera, M., Molina, E., King, M., Marston, L., Torres-Gonzalez, F., Moreno-Kustner, B., Moreno-Peral, P., Motrico, E., Monton-Franco, C., GildeGomez-Barragan, M. J., Sanchez-Celaya, M., Diaz-Barreiros, M. A., Vicens, C., Luna, J. D., Nazareth, I., & Cervilla, J. (2015). The risk for major depression conferred by childhood maltreatment is multiplied by BDNF and SERT genetic vulnerability: a replication study. *Journal of Psychiatry & Neuroscience, 40*(3), 187-196. doi:10.1503/jpn.140097

Haarasilta, L., Marttunen, M., Kaprio, J., & Aro, H. (2001). The 12-month prevalence and characteristics of major depressive episode in a representative nationwide sample of adolescents and young adults. *Psychol Med, 31*(7), 1169-1179.

Hagenauer, M. H., & Lee, T. M. (2013). Adolescent sleep patterns in humans and laboratory animals. *Horm Behav, 64*(2), 270-279. doi:10.1016/j.yhbeh.2013.01.013

Hagenauer, M. H., Perryman, J. I., Lee, T. M., & Carskadon, M. A. (2009). Adolescent changes in the homeostatic and circadian regulation of sleep. *Developmental neuroscience, 31*(4), 276-284. doi:10.1159/000216538

Hairston, I. S., Peyron, C., Denning, D. P., Ruby, N. F., Flores, J., Sapolsky, R. M., Heller, H. C., & O'Hara, B. F. (2004). Sleep deprivation effects on growth factor expression in neonatal rats: A potential role for BDNF in the mediation of delta power. *Journal of Neurophysiology, 91*(4), 1586-1595. doi:10.1152/jn.00894.2003

Hajszan, T., MacLusky, N. J., & Leranth, C. (2005). Short-term treatment with the antidepressant fluoxetine triggers pyramidal dendritic spine synapse formation in rat hippocampus. *European Journal of Neuroscience, 21*(5), 1299-1303. doi:10.1111/j.1460-9568.2005.03968.x

Halepoto, D. M., Bashir, S., & L, A. L. A. (2014). Possible role of brain-derived neurotrophic factor (BDNF) in autism spectrum disorder: current status. *J Coll Physicians Surg Pak, 24*(4), 274-278. doi:04.2014/JCPSP.274278

Hamilton, M. (1960). A rating scale for depression. *Journal of neurology, neurosurgery, and psychiatry, 23*, 56-62.

Hammen, C., Henry, R., & Daley, S. E. (2000). Depression and sensitization to stressors among young women as a function of childhood adversity. *J Consult Clin Psychol, 68*(5), 782-787. doi:Doi 10.1037/0022-006x.68.5.782

Hankin, B. L., Abramson, L. Y., Moffitt, T. E., Silva, P. A., McGee, R., & Angell, K. E. (1998). Development of depression from preadolescence to young adulthood: Emerging gender differences in a 10-year longitudinal study. *Journal of Abnormal Psychology, 107*(1), 128-140. doi:Doi 10.1037/0021-843x.107.1.128

Hanlon, E. C., Faraguna, U., Vyazovskiy, V. V., Tononi, G., & Cirelli, C. (2009). Effects of skilled training on sleep slow wave activity and cortical gene expression in the rat. *Sleep, 32*(6), 719-729.

Hanson, E. A. (2009). Can adenosine substitute for SAM-e as an adjunct in the treatment of depression? *Medical Hypotheses, 73*(5), 857-859. doi:10.1016/j.mehy.2009.04.025

Hariri, A. R., Goldberg, T. E., Mattay, V. S., Kolachana, B. S., Callicott, J. H., Egan, M. F., & Weinberger, D. R. (2003). Brain-derived neurotrophic factor val66met polymorphism affects human memory-related hippocampal activity and predicts memory performance. *J Neurosci, 23*(17), 6690-6694.

Harris, R. B. S., Zhou, J., Youngblood, B. D., Smagin, G. N., & Ryan, D. H. (1997). Failure to change exploration or saccharin preference in rats exposed to chronic mild stress. *Physiol Behav, 63*(1), 91-100. doi:Doi 10.1016/S0031-9384(97)00425-3

Hasler, G. (2010). Pathophysiology of depression: do we have any solid evidence of interest to clinicians? *World Psychiatry, 9*(3), 155-161.

Hasler, G., & Northoff, G. (2011). Discovering imaging endophenotypes for major depression. *Molecular psychiatry, 16*(6), 604-619. doi:10.1038/mp.2011.23

Heim, C., & Nemeroff, C. B. (2001). The role of childhood trauma in the neurobiology of mood and anxiety disorders: preclinical and clinical studies. *Biological psychiatry, 49*(12), 1023-1039.

Heim, C., & Nemeroff, C. B. (2002). Neurobiology of early life stress: clinical studies. *Semin Clin Neuropsychiatry, 7*(2), 147-159.

Heim, C., Newport, D. J., Mletzko, T., Miller, A. H., & Hemeroff, C. B. (2008). The link between childhood trauma and depression: Insights from HPA axis studies in humans. *Psychoneuroendocrinology, 33*(6), 693-710. doi:10.1016/j.psyneuen.2008.03.008

Henderson, J. M. T., France, K. G., & Blampied, N. M. (2011). The consolidation of infants' nocturnal sleep across the first year of life. *Sleep medicine reviews, 15*(4), 211-220. doi:10.1016/j.smrv.2010.08.003

Hengen, K. B., Torrado Pacheco, A., McGregor, J. N., Van Hooser, S. D., & Turrigiano, G. G. (2016). Neuronal Firing Rate Homeostasis Is Inhibited by Sleep and Promoted by Wake. *Cell, 165*(1), 180-191. doi:10.1016/j.cell.2016.01.046

Hobson, J. A. (2005). Sleep is of the brain, by the brain and for the brain. *Nature, 437*(7063), 1254-1256. doi:10.1038/nature04283

Hoffmann, R., Hendrickse, W., Rush, A. J., & Armitage, R. (2000). Slow-wave activity during non-REM sleep in men with schizophrenia and major depressive disorders. *Psychiatry research, 95*(3), 215-225. doi:Doi 10.1016/S0165-1781(00)00181-5

Holmboe, K., Nemoda, Z., Fearon, R. M. P., Sasvari-Szekely, M., & Johnson, M. H. (2011). Dopamine D4 receptor and serotonin transporter gene effects on the longitudinal development of infant temperament. *Genes Brain and Behavior, 10*(5), 513-522. doi:10.1111/j.1601-183X.2010.00669.x

Huber, R., Esser, S. K., Ferrarelli, F., Massimini, M., Peterson, M. J., & Tononi, G. (2007). TMS-induced cortical potentiation during wakefulness locally increases slow wave activity during sleep. *PLoS One, 2*(3), e276. doi:10.1371/journal.pone.0000276

Huber, R., Ghilardi, M. F., Massimini, M., & Tononi, G. (2004). Local sleep and learning. *Nature, 430*(6995), 78-81. doi:10.1038/nature02663

Huennerkopf, R., Strobel, A., Gutknecht, L., Brocke, B., & Lesch, K. P. (2007). Interaction between BDNF Val66Met and dopamine transporter gene variation influences anxiety-related traits. *Neuropsychopharmacology, 32*(12), 2552-2560. doi:10.1038/sj.npp.1301383

Huttenlocher, P. R. (1979). Synaptic Density in Human Frontal-Cortex - Developmental-Changes and Effects of Aging. *Brain research, 163*(2), 195-205.

Itoh, K., Hashimoto, K., Kumakiri, C., Shimizu, E., & Iyo, M. (2004). Association between brain-derived neurotrophic factor 196 G/A polymorphism and personality traits in healthy subjects. *American Journal of Medical Genetics Part B-Neuropsychiatric Genetics, 124B*(1), 61-63. doi:10.1002/ajmg.b.20078

Jenni, O. G., & Carskadon, M. A. (2004). Spectral analysis of the sleep electroencephalogram during adolescence. *Sleep, 27*(4), 774-783.

Juulia Paavonen, E., Saarenpaa-Heikkila, O., Polkki, P., Kylliainen, A., Porkka-Heiskanen, T., & Paunio, T. (2017). Maternal and paternal sleep during pregnancy in the Child-sleep birth cohort. *Sleep Medicine, 29*, 47-56. doi:10.1016/j.sleep.2016.09.011

Jylha, P., Melartin, T., Rytsala, H., & Isometsa, E. (2009). Neuroticism, introversion, and major depressive disorder--traits, states, or scars? *Depression and Anxiety, 26*(4), 325-334. doi:10.1002/da.20385

Kalinchuk, A. V., Lu, Y., Stenberg, D., Rosenberg, P. A., & Porkka-Heiskanen, T. (2006). Nitric oxide production in the basal forebrain is required for recovery sleep. *Journal of Neurochemistry, 99*(2), 483-498. doi:10.1111/j.1471-4159.2006.04077.x

Kalinchuk, A. V., McCarley, R. W., Stenberg, D., Porkka-Heiskanen, T., & Basheer, R. (2008). The role of cholinergic basal forebrain neurons in adenosine-mediated homeostatic control of sleep: lessons from 192 IgG-saporin lesions. *Neuroscience, 157*(1), 238-253. doi:10.1016/j.neuroscience.2008.08.040

Kalueff, A. V., & Tuohimaa, P. (2004). Experimental modeling of anxiety and depression. *Acta Neurobiol Exp (Wars), 64*(4), 439-448.

Kalueff, A. V., Wheaton, M., & Murphy, D. L. (2007). What's wrong with my mouse model? Advances and strategies in animal modeling of anxiety and depression. *Behav Brain Res, 179*(1), 1-18. doi:10.1016/j.bbr.2007.01.023

Karpova, N. N. (2014). Role of BDNF epigenetics in activity-dependent neuronal plasticity. *Neuropharmacology, 76*, 709-718. doi:10.1016/j.neuropharm.2013.04.002

Karpova, N. N., Rantamaki, T., Di Lieto, A., Lindemann, L., Hoener, M. C., & Castren, E. (2010). Darkness reduces BDNF expression in the visual cortex and induces repressive chromatin remodeling at the BDNF gene in both hippocampus and visual cortex. *Cell Mol Neurobiol, 30*(7), 1117-1123. doi:10.1007/s10571-010-9544-6

Kaster, M. P., Rosa, A. O., Rosso, M. M., Goulart, E. C., Santos, A. R. S., & Rodrigues, A. L. S. (2004). Adenosine administration produces an antidepressant-like effect in mice: evidence for the involvement of A(1) and A(2A) receptors. *Neurosci Lett, 355*(1-2), 21-24. doi:10.1016/j.neulet.2003.10.040

Katz, R. J. (1982). Animal model of depression: pharmacological sensitivity of a hedonic deficit. *Pharmacol Biochem Behav, 16*(6), 965-968.

Katz, R. J., Roth, K. A., & Carroll, B. J. (1981). Acute and chronic stress effects on open field activity in the rat: implications for a model of depression. *Neurosci Biobehav Rev, 5*(2), 247-251.

Kaufman, J., Birmaher, B., Brent, D., Rao, U., Flynn, C., Moreci, P., Williamson, D., & Ryan, N. (1997). Schedule for Affective Disorders and Schizophrenia for School-Age Children-Present and Lifetime Version (K-SADS-PL): initial reliability and validity data. *J Am Acad Child Adolesc Psychiatry, 36*(7), 980-988. doi:10.1097/00004583-199707000-00021

Kaufman, J., Yang, B. Z., Douglas-Palumberi, H., Houshyar, S., Lipschitz, D., Krystal, J. H., & Gelernter, J. (2004). Social supports and serotonin transporter gene moderate depression in maltreated children. *Proceedings of the National Academy of Sciences of the United States of America, 101*(49), 17316-17321. doi:DOI 10.1073/pnas.0404376101

Kelly, M. P., & Deadwyler, S. A. (2003). Experience-dependent regulation of the immediate-early gene arc differs across brain regions. *Journal of Neuroscience, 23*(16), 6443-6451.

Kendler, K. S., Bulik, C. M., Silberg, J., Hettema, J. M., Myers, J., & Prescott, C. A. (2000). Childhood sexual abuse and adult psychiatric and substance use disorders in women - An epidemiological and Cotwin control analysis. *Archives of general psychiatry, 57*(10), 953-959. doi:DOI 10.1001/archpsyc.57.10.953

Kendler, K. S., Kuhn, J. W., & Prescott, C. A. (2004). Childhood sexual abuse, stressful life events and risk for major depression in women. *Psychol Med, 34*(8), 1475-1482. doi:10.1017/S0033291704002675

Kessler, R. C. (1997). The effects of stressful life events on depression. *Annu Rev Psychol, 48*, 191-214. doi:10.1146/annurev.psych.48.1.191

Kessler, R. C. (2007). The global burden of anxiety and mood disorders: putting the European Study of the Epidemiology of Mental Disorders (ESEMeD) findings into perspective. *J Clin Psychiatry, 68 Suppl 2*, 10-19.

Kessler, R. C., McLaughlin, K. A., Green, J. G., Gruber, M. J., Sampson, N. A., Zaslavsky, A. M., Aguilar-Gaxiola, S., Alhamzawi, A. O., Alonso, J., Angermeyer, M., Benjet, C., Bromet, E., Chatterji, S., de Girolamo, G., Demyttenaere, K., Fayyad, J., Florescu, S., Gal, G., Gureje, O., Haro, J. M., Hu, C. Y., Karam, E. G., Kawakami, N., Lee, S., Lepine, J. P., Ormel, J., Posada-Villa, J., Sagar, R., Tsang, A., Ustun, T. B., Vassilev, S., Viana, M. C., & Williams, D. R. (2010). Childhood adversities and adult psychopathology in the WHO World Mental Health Surveys. *British Journal of Psychiatry, 197*(5), 378-385. doi:10.1192/bjp.bp.110.080499

Khan, A. U., & Todd, S. (1990). Polysomnographic findings in adolescents with major depression. *Psychiatry research, 33*(3), 313-320.

Klintsova, A. Y., & Greenough, W. T. (1999). Synaptic plasticity in cortical systems. *Curr Opin Neurobiol, 9*(2), 203-208.

Koskenvuo, K., Hublin, C., Partinen, M., Paunio, T., & Koskenvuo, M. (2010). Childhood adversities and quality of sleep in adulthood: A population-based study of 26,000 Finns. *Sleep Medicine, 11*(1), 17-22. doi:10.1016/j.sleep.2009.03.010

Krishnan, V., & Nestler, E. J. (2008). The molecular neurobiology of depression. *Nature, 455*(7215), 894-902. doi:10.1038/nature07455

Kulkarni, S. K., & Mehta, A. K. (1985). Purine Nucleoside Mediated Immobility in Mice - Reversal by Antidepressants. *Psychopharmacology, 85*(4), 460-463. doi:Doi 10.1007/Bf00429665

Kupfer, D. J., Frank, E., McEachran, A. B., & Grochocinski, V. J. (1990). Delta sleep ratio. A biological correlate of early recurrence in unipolar affective disorder. *Archives of general psychiatry, 47*(12), 1100-1105.

Kutcher, S., Williamson, P., Marton, P., & Szalai, J. (1992). REM latency in endogenously depressed adolescents. *Br J Psychiatry, 161*, 399-402.

Kuzumaki, N., Ikegami, D., Tamura, R., Hareyama, N., Imai, S., Narita, M., Torigoe, K., Niikura, K., Takeshima, H., Ando, T., Igarashi, K., Kanno, J., Ushijima, T., Suzuki, T., & Narita, M. (2011). Hippocampal Epigenetic Modification at the Brain-Derived Neurotrophic Factor Gene Induced by an Enriched Environment. *Hippocampus, 21*(2), 127-132. doi:10.1002/hipo.20775

Lang, U. E., Hellweg, R., Kalus, P., Bajbouj, M., Lenzen, K. P., Sander, T., Kunz, D., & Gallinat, J. (2005). Association of a functional BDNF polymorphism and anxiety-related personality traits. *Psychopharmacology, 180*(1), 95-99. doi:10.1007/s00213-004-2137-7

Lee, B. H., Kim, H., Park, S. H., & Kim, Y. K. (2007). Decreased plasma BDNF level in depressive patients. *Journal of affective disorders, 101*(1-3), 239-244. doi:10.1016/j.jad.2006.11.005

Lee, B. H., & Kim, Y. K. (2010). The roles of BDNF in the pathophysiology of major depression and in antidepressant treatment. *Psychiatry Investig, 7*(4), 231-235. doi:10.4306/pi.2010.7.4.231

Levenstein, D., Watson, B. O., Rinzel, J., & Buzsaki, G. (2017). Sleep regulation of the distribution of cortical firing rates. *Curr Opin Neurobiol, 44*, 34-42. doi:10.1016/j.conb.2017.02.013

Levine, S. (1970). The pituitary-adrenal system and the developing brain. *Prog Brain Res, 32*, 79-85. doi:10.1016/S0079-6123(08)61521-6

Lewinsohn, P. M., Clarke, G. N., Seeley, J. R., & Rohde, P. (1994). Major depression in community adolescents: age at onset, episode duration, and time to recurrence. *J Am Acad Child Adolesc Psychiatry, 33*(6), 809-818. doi:10.1097/00004583-199407000-00006

Liu, D., Diorio, J., Tannenbaum, B., Caldji, C., Francis, D., Freedman, A., Sharma, S., Pearson, D., Plotsky, P. M., & Meaney, M. J. (1997). Maternal care, hippocampal glucocorticoid receptors, and hypothalamic-pituitary-adrenal responses to stress. *Science, 277*(5332), 1659-1662.

Liu, R. T. (2010). Early life stressors and genetic influences on the development of bipolar disorder: the roles of childhood abuse and brain-derived neurotrophic factor. *Child Abuse Negl, 34*(7), 516-522. doi:10.1016/j.chiabu.2009.10.009

Liu, X., Buysse, D. J., Gentzler, A. L., Kiss, E., Mayer, L., Kapornai, K., Vetro, A., & Kovacs, M. (2007). Insomnia and hypersomnia associated with depressive phenomenology and comorbidity in childhood depression. *Sleep, 30*(1), 83-90.

Liu, Z. W., Faraguna, U., Cirelli, C., Tononi, G., & Gao, X. B. (2010). Direct Evidence for Wake-Related Increases and Sleep-Related Decreases in Synaptic Strength in Rodent Cortex. *Journal of Neuroscience, 30*(25), 8671-8675. doi:10.1523/Jneurosci.1409-10.2010

Livak, K. J., & Schmittgen, T. D. (2001). Analysis of relative gene expression data using real-time quantitative PCR and the 2(-Delta Delta C(T)) Method. *Methods, 25*(4), 402-408. doi:10.1006/meth.2001.1262

Lohoff, F. W. (2010). Overview of the genetics of major depressive disorder. *Curr Psychiatry Rep, 12*(6), 539-546. doi:10.1007/s11920-010-0150-6

Lopez, J., Hoffmann, R., & Armitage, R. (2010). Reduced sleep spindle activity in early-onset and elevated risk for depression. *J Am Acad Child Adolesc Psychiatry, 49*(9), 934-943. doi:10.1016/j.jaac.2010.05.014

Lopez, J., Hoffmann, R., Emslie, G., & Armitage, R. (2012). Sex Differences in Slow-wave Electroencephalographic Activity (SWA) in Adolescent Depression. *Mental illness, 4*(1), e4. doi:10.4081/mi.2012.e4

Lu, B., Pang, P. T., & Woo, N. H. (2005). The yin and yang of neurotrophin action. *Nature Reviews Neuroscience, 6*(8), 603-614. doi:10.1038/nrn1726

Luine, V., & Frankfurt, M. (2013). Interactions between Estradiol, Bdnf and Dendritic Spines in Promoting Memory. *Neuroscience, 239*, 34-45. doi:10.1016/j.neuroscience.2012.10.019

Lyytinen, P., Ahonen, T., Eklund, K., & Lyytinen, H. . (2000). Äänтelyn ja motoriikan kehityksen seurantamenetelmä. *Niilo Mäki Instituutti*.

Maccari, S., Piazza, P. V., Kabbaj, M., Barbazanges, A., Simon, H., & Le Moal, M. (1995). Adoption reverses the long-term impairment in glucocorticoid feedback induced by prenatal stress. *J Neurosci, 15*(1 Pt 1), 110-116.

Maes, M., Yirmyia, R., Noraberg, J., Brene, S., Hibbeln, J., Perini, G., Kubera, M., Bob, P., Lerer, B., & Maj, M. (2009). The inflammatory & neurodegenerative (I&ND) hypothesis of depression: leads for future research and new drug developments in depression. *Metab Brain Dis, 24*(1), 27-53. doi:10.1007/s11011-008-9118-1

Maletic, V., Robinson, M., Oakes, T., Iyengar, S., Ball, S. G., & Russell, J. (2007). Neurobiology of depression: an integrated view of key findings. *International Journal of Clinical Practice, 61*(12), 2030-2040. doi:10.1111/j.1742-1241.2007.01602.x

Manji, H. K., Quiroz, J. A., Sporn, J., Payne, J. L., Denicoff, K., Gray, N. A., Zarate, C. A., & Charney, D. S. (2003). Enhancing neuronal plasticity and cellular resilience to

develop novel, improved therapeutics for difficult-to-treat depression. *Biological psychiatry, 53*(8), 707-742. doi:10.1016/S0006-3223(03)00117-3

Mann, J. J. (2005). Drug therapy - The medical management of depression. *New England Journal of Medicine, 353*(17), 1819-1834. doi:DOI 10.1056/NEJMra050730

Maret, S., Faraguna, U., Nelson, A. B., Cirelli, C., & Tononi, G. (2011). Sleep and waking modulate spine turnover in the adolescent mouse cortex. *Nat Neurosci, 14*(11), 1418-1420. doi:10.1038/nn.2934

Marks, G. A., Shaffery, J. P., Oksenberg, A., Speciale, S. G., & Roffwarg, H. P. (1995). A Functional-Role for Rem-Sleep in Brain Maturation. *Behav Brain Res, 69*(1-2), 1-11. doi:Doi 10.1016/0166-4328(95)00018-O

Martin, R. P., Wisenbaker, J., Baker, J., & Huttunen, M. O. (1997). Gender differences in temperament at six months and five years. *Infant Behavior & Development, 20*(3), 339-347. doi:Doi 10.1016/S0163-6383(97)90005-9

Martinowich, K., Schloesser, R. J., Jimenez, D. V., Weinberger, D. R., & Lu, B. (2011). Activity-dependent brain-derived neurotrophic factor expression regulates cortistatin-interneurons and sleep behavior. *Mol Brain, 4*. doi:Artn 11 10.1186/1756-6606-4-11

Massart, R., Mongeau, R., & Lanfumey, L. (2012). Beyond the monoaminergic hypothesis: neuroplasticity and epigenetic changes in a transgenic mouse model of depression. *Philosophical Transactions of the Royal Society B-Biological Sciences, 367*(1601), 2485-2494. doi:10.1098/rstb.2012.0212

Maya-Vetencourt, J. F., Sale, A., Viegi, A., Baroncelli, L., De Pasquale, R., O'Leary, O. F., Castren, E., & Maffei, L. (2008). The antidepressant fluoxetine restores plasticity in the adult visual cortex. *Science, 320*(5874), 385-388. doi:10.1126/science.1150516

McCarty, R. (2017). Cross-fostering: Elucidating the effects of genexenvironment interactions on phenotypic development. *Neurosci Biobehav Rev, 73*, 219-254. doi:10.1016/j.neubiorev.2016.12.025

McCormick, C. M., Smith, C., & Mathews, I. Z. (2008). Effects of chronic social stress in adolescence on anxiety and neuroendocrine response to mild stress in male and female rats. *Behav Brain Res, 187*(2), 228-238. doi:10.1016/j.bbr.2007.09.005

McEwen, B. S. (2004). Protection and damage from acute and chronic stress - Allostasis and allostatic overload and relevance to the pathophysiology of psychiatric disorders. *Biobehavioral Stress Response: Protective and Damaging Effects, 1032*, 1-7. doi:10.1193/annals.1314.001

McGowan, P. O., Sasaki, A., D'Alessio, A. C., Dymov, S., Labonte, B., Szyf, M., Turecki, G., & Meaney, M. J. (2009). Epigenetic regulation of the glucocorticoid receptor in human

brain associates with childhood abuse. *Nat Neurosci, 12*(3), 342-348. doi:10.1038/nn.2270

Meaney, M. J., & Szyf, M. (2005). Environmental programming of stress responses through DNA methylation: life at the interface between a dynamic environment and a fixed genome. *Dialogues in clinical neuroscience, 7*(2), 103-123.

Mendelson, W. B., & Bergmann, B. M. (1999). EEG delta power during sleep in young and old rats. *Neurobiology of Aging, 20*(6), 669-673. doi:Doi 10.1016/S0197-4580(99)00062-7

Merikanto, I., Partonen, T., Paunio, T., Castaneda, A. E., Marttunen, M., & Urrila, A. S. (2017). Advanced phases and reduced amplitudes are suggested to characterize the daily rest-activity cycles in depressed adolescent boys. *Chronobiology International, 37*(7), 967-976. doi:10.1080/07420528.2017.1332072

Milaneschi, Y., Hoogendijk, W., Lips, P., Heijboer, A. C., Schoevers, R., van Hemert, A. M., Beekman, A. T. F., Smit, J. H., & Penninx, B. W. J. H. (2014). The association between low vitamin D and depressive disorders. *Molecular psychiatry, 19*(4), 444-451. doi:10.1038/mp.2013.36

Miller, A. H., Maletic, V., & Raison, C. L. (2009). Inflammation and Its Discontents: The Role of Cytokines in the Pathophysiology of Major Depression. *Biological psychiatry, 65*(9), 732-741. doi:10.1016/j.biopsych.2008.11.029

Miller, A. H., & Raison, C. L. (2016). The role of inflammation in depression: from evolutionary imperative to modern treatment target. *Nature Reviews Immunology, 16*(1), 22-34. doi:10.1038/nri.2015.5

Minor, T. R., Rowe, M. K., Job, R. F. S., & Ferguson, E. C. (2001). Escape deficits induced by inescapable shock and metabolic stress are reversed by adenosine receptor antagonists. *Behav Brain Res, 120*(2), 203-212. doi:Doi 10.1016/S0166-4328(00)00376-4

Mirmiran, M., Maas, Y. G., & Ariagno, R. L. (2003). Development of fetal and neonatal sleep and circadian rhythms. *Sleep medicine reviews, 7*(4), 321-334.

Mitchelmore, C., & Gede, L. (2014). Brain derived neurotrophic factor: Epigenetic regulation in psychiatric disorders. *Brain research, 1586*, 162-172. doi:10.1016/j.brainres.2014.06.037

Moore, R. Y. (2007). Suprachiasmatic nucleus in sleep-wake regulation. *Sleep Medicine, 8*, S27-S33. doi:10.1016/j.sleep.2007.10.003

Mrdalj, J., Pallesen, S., Milde, A. M., Jellestad, F. K., Murison, R., Ursin, R., Bjorvatn, B., & Gronli, J. (2013). Early and later life stress alter brain activity and sleep in rats. *PLoS One, 8*(7), e69923. doi:10.1371/journal.pone.0069923

Murgatroyd, C., Patchev, A. V., Wu, Y., Micale, V., Bockmuhl, Y., Fischer, D., Holsboer, F., Wotjak, C. T., Almeida, O. F., & Spengler, D. (2009). Dynamic DNA methylation programs persistent adverse effects of early-life stress. *Nat Neurosci, 12*(12), 1559-1566. doi:10.1038/nn.2436

Myers-Schulz, B., & Koenigs, M. (2012). Functional anatomy of ventromedial prefrontal cortex: implications for mood and anxiety disorders. *Molecular psychiatry, 17*(2), 132-141. doi:10.1038/mp.2011.88

Nelson, E. C., Heath, A. C., Madden, P. A. F., Cooper, M. L., Dinwiddie, S. H., Bucholz, K. K., Glowinski, A., McLaughlin, T., Dunne, M. P., Statham, D. J., & Martin, N. G. (2002). Association between self-reported childhood sexual abuse and adverse psychosocial outcomes - Results from a twin study. *Archives of general psychiatry, 59*(2), 139-145. doi:DOI 10.1001/archpsyc.59.2.139

Nestler, E. J., Barrot, M., DiLeone, R. J., Eisch, A. J., Gold, S. J., & Monteggia, L. M. (2002). Neurobiology of depression. *Neuron, 34*(1), 13-25.

Neumeister, A., Nugent, A. C., Waldeck, T., Geraci, M., Schwarz, M., Bonne, O., Bain, E. E., Luckenbaugh, D. A., Herscovitch, P., Charney, D. S., & Drevets, W. C. (2004). Neural and behavioral responses to tryptophan depletion in unmedicated patients with remitted major depressive disorder and controls. *Archives of general psychiatry, 61*(8), 765-773. doi:DOI 10.1001/archpsyc.61.8.765

Neves-Pereira, M., Mundo, E., Muglia, P., King, N., Macciardi, F., & Kennedy, J. L. (2002). The brain-derived neurotrophic factor gene confers susceptibility to bipolar disorder: Evidence from a family-based association study. *American Journal of Human Genetics, 71*(3), 651-655. doi:Doi 10.1086/342288

Newman, D. L., Moffitt, T. E., Caspi, A., Magdol, L., Silva, P. A., & Stanton, W. R. (1996). Psychiatric disorder in a birth cohort of young adults: Prevalence, comorbidity, clinical significance, and new case incidence from ages 11 to 21. *J Consult Clin Psychol, 64*(3), 552-562. doi:Doi 10.1037//0022-006x.64.3.552

Nibuya, M., Morinobu, S., & Duman, R. S. (1995). Regulation of BDNF and trkB mRNA in rat brain by chronic electroconvulsive seizure and antidepressant drug treatments. *J Neurosci, 15*(11), 7539-7547.

Nieminen, P., Korpela, R. . (2004). Leikkitarkkailu: käsikirja lapsen toiminnallisen kehityksen arvioimiseksi. *Tampereen yliopisto, psykologian laitos*.

Nishichi, R., Nufuji, Y., Washio, M., & Kumagai, S. (2013). Serum Brain-Derived Neurotrophic Factor Levels Are Associated with Dyssomnia in Females, but not Males, among Japanese Workers. *Journal of Clinical Sleep Medicine, 9*(7), 649-654. doi:10.5664/jcsm.2828

Nolen-Hoeksema, S. (2001). Gender differences in depression. *Current Directions in Psychological Science, 10*(5), 173-176. doi:Doi 10.1111/1467-8721.00142

Numakawa, T., Suzuki, S., Kumamaru, E., Adachi, N., Richards, M., & Kunugi, H. (2010). BDNF function and intracellular signaling in neurons. *Histology and Histopathology, 25*(2), 237-258.

Nutt, D., Wilson, S., & Paterson, L. (2008). Sleep disorders as core symptoms of depression. *Dialogues in clinical neuroscience, 10*(3), 329-336.

Oldehinkel, A. J., Wittchen, H. U., & Schuster, P. (1999). Prevalence, 20-month incidence and outcome of unipolar depressive disorders in a community sample of adolescents. *Psychol Med, 29*(3), 655-668. doi:Doi 10.1017/S0033291799008454

Onishchenko, N., Karpova, N., Sabri, F., Castren, E., & Ceccatelli, S. (2008). Long-lasting depression-like behavior and epigenetic changes of BDNF gene expression induced by perinatal exposure to methylmercury. *Journal of Neurochemistry, 106*(3), 1378-1387. doi:10.1111/j.1471-4159.2008.05484.x

Pan, W. H., Banks, W. A., Fasold, M. B., Bluth, J., & Kastin, A. J. (1998). Transport of brain-derived neurotrophic factor across the blood-brain barrier. *Neuropharmacology, 37*(12), 1553-1561. doi:Doi 10.1016/S0028-3908(98)00141-5

Pape, K., Bjorngaard, J. H., Holmen, T. L., & Krokstad, S. (2012). The welfare burden of adolescent anxiety and depression: a prospective study of 7500 young Norwegians and their families: the HUNT study. *Bmj Open, 2*(6). doi:ARTN e001942 10.1136/bmjopen-2012-001942

Parade, S. H., & Leerkes, E. M. (2008). The reliability and validity of the Infant Behavior Questionnaire-Revised. *Infant Behavior & Development, 31*(4), 637-646. doi:10.1016/j.infbeh.2008.07.009

Pariante, C. M., & Lightman, S. L. (2008). The HPA axis in major depression: classical theories and new developments. *Trends Neurosci, 31*(9), 464-468. doi:10.1016/j.tins.2008.06.006

Paunio, T., Korhonen, T., Hublin, C., Partinen, M., Koskenvuo, K., Koskenvuo, M., & Kaprio, J. (2015). Poor sleep predicts symptoms of depression and disability retirement due to depression. *Journal of affective disorders, 172*, 381-389. doi:10.1016/j.jad.2014.10.002

Pechtel, P., & Pizzagalli, D. A. (2011). Effects of early life stress on cognitive and affective function: an integrated review of human literature. *Psychopharmacology, 214*(1), 55-70. doi:10.1007/s00213-010-2009-2

Perez-Morales, M., Fajardo-Valdez, A., Mendez-Diaz, M., Ruiz-Contreras, A. E., & Prospero-Garcia, O. (2014). 2-Arachidonoylglycerol into the lateral hypothalamus improves

reduced sleep in adult rats subjected to maternal separation. *Neuroreport, 25*(18), 1437-1441. doi:10.1097/WNR.0000000000000287

Perlis, M. L., Giles, D. E., Buysse, D. J., Thase, M. E., Tu, X., & Kupfer, D. J. (1997). Which depressive symptoms are related to which sleep electroencephalographic variables? *Biological psychiatry, 42*(10), 904-913. doi:Doi 10.1016/S0006-3223(96)00439-8

Pezawas, L., Verchinski, B. A., Mattay, V. S., Callicott, J. H., Kolachana, B. S., Straub, R. E., Egan, M. F., Meyer-Lindenberg, A., & Weinberger, D. R. (2004). The brain-derived neurotrophic factor val66met polymorphism and variation in human cortical morphology. *J Neurosci, 24*(45), 10099-10102. doi:10.1523/JNEUROSCI.2680-04.2004

Pine, D. S., Cohen, E., Cohen, P., & Brook, J. (1999). Adolescent depressive symptoms as predictors of adult depression: Moodiness or mood disorder? *American Journal of Psychiatry, 156*(1), 133-135.

Pivac, N., Kim, B., Nedic, G., Joo, Y. H., Kozaric-Kovacic, D., Hong, J. P., & Muck-Seler, D. (2009). Ethnic Differences in Brain-derived Neurotrophic Factor Val66Met Polymorphism in Croatian and Korean Healthy Participants. *Croatian Medical Journal, 50*(1), 43-48. doi:10.3325/cmj.2009.50.43

Plotsky, P. M., & Meaney, M. J. (1993). Early, Postnatal Experience Alters Hypothalamic Corticotropin-Releasing Factor (Crf) Messenger-Rna, Median-Eminence Crf Content and Stress-Induced Release in Adult-Rats. *Molecular Brain Research, 18*(3), 195-200. doi:Doi 10.1016/0169-328x(93)90189-V

Poduslo, J. F., & Curran, G. L. (1996). Permeability at the blood-brain and blood-nerve barriers of the neurotrophic factors: NGF, CNTF, NT-3, BDNF. *Molecular Brain Research, 36*(2), 280-286. doi:Doi 10.1016/0169-328x(95)00250-V

Porkka-Heiskanen, T. (2013). Sleep homeostasis. *Curr Opin Neurobiol, 23*(5), 799-805. doi:10.1016/j.conb.2013.02.010

Porkka-Heiskanen, T., & Kalinchuk, A. V. (2011). Adenosine, energy metabolism and sleep homeostasis. *Sleep medicine reviews, 15*(2), 123-135. doi:10.1016/j.smrv.2010.06.005

Porkka-Heiskanen, T., Strecker, R. E., & McCarley, R. W. (2000). Brain site-specificity of extracellular adenosine concentration changes during sleep deprivation and spontaneous sleep: An in vivo microdialysis study. *Neuroscience, 99*(3), 507-517. doi:Doi 10.1016/S0306-4522(00)00220-7

Porkka-Heiskanen, T., Strecker, R. E., Thakkar, M., Bjorkum, A. A., Greene, R. W., & McCarley, R. W. (1997). Adenosine: a mediator of the sleep-inducing effects of prolonged wakefulness. *Science, 276*(5316), 1265-1268.

Prins, J., Olivier, B., & Korte, S. M. (2011). Triple reuptake inhibitors for treating subtypes of major depressive disorder: the monoamine hypothesis revisited. *Expert Opinion on Investigational Drugs, 20*(8), 1107-1130. doi:10.1517/13543784.2011.594039

Prut, L., & Belzung, C. (2003). The open field as a paradigm to measure the effects of drugs on anxiety-like behaviors: a review. *Eur J Pharmacol, 463*(1-3), 3-33.

Pruunsild, P., Kazantseva, A., Aid, T., Palm, K., & Timmusk, T. (2007). Dissecting the human BDNF locus: Bidirectional transcription, complex splicing, and multiple promoters. *Genomics, 90*(3), 397-406. doi:10.1016/j.ygeno.2007.05.004

Pruunsild, P., Sepp, M., Orav, E., Koppel, I., & Timmusk, T. (2011). Identification of cis-elements and transcription factors regulating neuronal activity-dependent transcription of human BDNF gene. *J Neurosci, 31*(9), 3295-3308. doi:10.1523/JNEUROSCI.4540-10.2011

Pryce, C. R., Ruedi-Bettschen, D., Dettling, A. C., Weston, A., Russig, H., Ferger, B., & Feldon, J. (2005). Long-term effects of early-life environmental manipulations in rodents and primates: Potential animal models in depression research. *Neurosci Biobehav Rev, 29*(4-5), 649-674. doi:10.1016/j.neubiorev.2005.03.011

Pull, I., & Mcilwain, H. (1972). Metabolism of [C-14] Adenine and Derivatives by Cerebral Tissues, Superfused and Electrically Stimulated. *Biochemical Journal, 126*(4), 965-&.

Purcell, S., Cherny, S. S., & Sham, P. C. (2003). Genetic Power Calculator: design of linkage and association genetic mapping studies of complex traits. *Bioinformatics, 19*(1), 149-150.

Putnam, S. P., Helbig, A. L., Gartstein, M. A., Rothbart, M. K., & Leerkes, E. (2014). Development and Assessment of Short and Very Short Forms of the Infant Behavior Questionnaire-Revised. *Journal of Personality Assessment, 96*(4), 445-458. doi:10.1080/00223891.2013.841171

Rajkowska, G., Miguel-Hidalgo, J. J., Wei, J. R., Dilley, G., Pittman, S. D., Meltzer, H. Y., Overholser, J. C., Roth, B. L., & Stockmeier, C. A. (1999). Morphometric evidence for neuronal and glial prefrontal cell pathology in major depression. *Biological psychiatry, 45*(9), 1085-1098. doi:Doi 10.1016/S0006-3223(99)00041-4

Rao, U., & Chen, L. A. (2009). Characteristics, correlates, and outcomes of childhood and adolescent depressive disorders. *Dialogues in clinical neuroscience, 11*(1), 45-62.

Rao, U., Hammen, C. L., & Poland, R. E. (2009). Risk markers for depression in adolescents: sleep and HPA measures. *Neuropsychopharmacology, 34*(8), 1936-1945. doi:10.1038/npp.2009.27

Rasch, B., & Born, J. (2013). About sleep's role in memory. *Physiol Rev, 93*(2), 681-766. doi:10.1152/physrev.00032.2012

Redina, O. E., Smolenskaya, S. E., Maslova, L. N., Sakharov, D. G., & Markel, A. L. (2009). The characteristics of motor activity in ISIAH rats in an open field test are controlled by genes on chromosomes 2 and 16. *Neurosci Behav Physiol, 39*(1), 57-64. doi:10.1007/s11055-008-9100-8

Remus, J. L., Stewart, L. T., Camp, R. M., Novak, C. M., & Johnson, J. D. (2015). Interaction of Metabolic Stress With Chronic Mild Stress in Altering Brain Cytokines and Sucrose Preference. *Behav Neurosci, 129*(3), 321-330. doi:10.1037/bne0000056

Reyes Prieto, N. M., Romano Lopez, A., Perez Morales, M., Pech, O., Mendez-Diaz, M., Ruiz Contreras, A. E., & Prospero-Garcia, O. (2012). Oleamide restores sleep in adult rats that were subjected to maternal separation. *Pharmacol Biochem Behav, 103*(2), 308-312. doi:10.1016/j.pbb.2012.08.028

Reynolds, S., Carrey, N., Jaworska, N., Langevin, L. M., Yang, X. R., & Macmaster, F. P. (2014). Cortical thickness in youth with major depressive disorder. *BMC Psychiatry, 14*, 83. doi:10.1186/1471-244X-14-83

Rial, D., Lemos, C., Pinheiro, H., Duarte, J. M., Goncalves, F. Q., Real, J. I., Prediger, R. D., Goncalves, N., Gomes, C. A., Canas, P. M., Agostinho, P., & Cunha, R. A. (2016). Depression as a Glial-Based Synaptic Dysfunction. *Frontiers in Cellular Neuroscience, 9*. doi:ARTN 521 10.3389/fncel.2015.00521

Robert, J. J., Hoffmann, R. F., Emslie, G. J., Hughes, C., Rintelmann, J., Moore, J., & Armitage, R. (2006). Sex and age differences in sleep macroarchitecture in childhood and adolescent depression. *Sleep, 29*(3), 351-358.

Roceri, M., Cirulli, F., Pessina, C., Peretto, P., Racagni, G., & Riva, M. A. (2004). Postnatal repeated maternal deprivation produces age-dependent changes of brain-derived neurotrophic factor expression in selected rat brain regions. *Biological psychiatry, 55*(7), 708-714. doi:10.1016/j.biopsych.2003.12.011

Roceri, M., Hendriks, W., Racagni, G., Ellenbroek, B. A., & Riva, M. A. (2002). Early maternal deprivation reduces the expression of BDNF and NMDA receptor subunits in rat hippocampus. *Molecular psychiatry, 7*(6), 609-616. doi:10.1038/sj.mp.4001036

Roque, S., Mesquita, A. R., Palha, J. A., Sousa, N., & Correia-Neves, M. (2014). The behavioral and immunological impact of maternal separation: a matter of timing. *Front Behav Neurosci, 8*, 192. doi:10.3389/fnbeh.2014.00192

Roth, T. L., Lubin, F. D., Funk, A. J., & Sweatt, J. D. (2009). Lasting epigenetic influence of early-life adversity on the BDNF gene. *Biological psychiatry, 65*(9), 760-769. doi:10.1016/j.biopsych.2008.11.028

Rygula, R., Abumaria, N., Flugge, G., Fuchs, E., Ruther, E., & Havemann-Reinecke, U. (2005). Anhedonia and motivational deficits in rats: impact of chronic social stress. *Behav Brain Res, 162*(1), 127-134. doi:10.1016/j.bbr.2005.03.009

Saarelainen, T., Hendolin, P., Lucas, G., Koponen, E., Sairanen, M., MacDonald, E., Agerman, K., Haapasalo, A., Nawa, H., Aloyz, R., Ernfors, P., & Castren, E. (2003). Activation of the TrkB neurotrophin receptor is induced by antidepressant drugs and is required for antidepressant-induced behavioral effects. *Journal of Neuroscience, 23*(1), 349-357.

Sagatun, A., Wentzel-Larsen, T., Heyerdahl, S., & Lien, L. (2016). Mental health in adolescence and subsequent receipt of medical benefits in young adulthood: The mediating role of upper secondary school completion. *Scandinavian Journal of Public Health, 44*(4), 431-438. doi:10.1177/1403494815622309

Sampath, D., Sabitha, K. R., Hegde, P., Jayakrishnan, H. R., Kutty, B. M., Chattarji, S., Rangarajan, G., & Laxmi, T. R. (2014). A study on fear memory retrieval and REM sleep in maternal separation and isolation stressed rats. *Behav Brain Res, 273*, 144-154. doi:10.1016/j.bbr.2014.07.034

Sanchez, M. M., Ladd, C. O., & Plotsky, P. M. (2001). Early adverse experience as a developmental risk factor for later psychopathology: Evidence from rodent and primate models. *Development and Psychopathology, 13*(3), 419-449. doi:Doi 10.1017/S0954579401003029

Satoh, S., Matsumura, H., & Hayaishi, O. (1998). Involvement of adenosine A2A receptor in sleep promotion. *Eur J Pharmacol, 351*(2), 155-162.

Satomaa, A. L., Saarenpaa-Heikkila, O., Paavonen, E. J., & Himanen, S. L. (2016). The adapted American Academy of Sleep Medicine sleep scoring criteria in one month old infants: A means to improve comparability? *Clin Neurophysiol, 127*(2), 1410-1418. doi:10.1016/j.clinph.2015.08.013

Satou, R., Sugihara, N., Ishizuka, Y., Matsukubo, T., & Onishi, Y. (2013). DNA methylation of the BMAL1 promoter. *Biochemical and Biophysical Research Communications, 440*(3), 449-453. doi:10.1016/j.bbrc.2013.09.124

Savelyev, S. A., Rantamaki, T., Rytkonen, K. M., Castren, E., & Porkka-Heiskanen, T. (2012). Sleep homeostasis and depression: studies with the rat clomipramine model of depression. *Neuroscience, 212*, 149-158. doi:10.1016/j.neuroscience.2012.03.029

Schaaf, M. J., de Jong, J., de Kloet, E. R., & Vreugdenhil, E. (1998). Downregulation of BDNF mRNA and protein in the rat hippocampus by corticosterone. *Brain research, 813*(1), 112-120.

Schmidt, M. V., Enthoven, L., van der Mark, M., Levine, S., de Kloet, E. R., & Oitzl, M. S. (2003). The postnatal development of the hypothalamic-pituitary-adrenal axis in the mouse. *Int J Dev Neurosci, 21*(3), 125-132.

Schmidt, M. V., Wang, X. D., & Meijer, O. C. (2011). Early life stress paradigms in rodents: potential animal models of depression? *Psychopharmacology, 214*(1), 131-140. doi:10.1007/s00213-010-2096-0

Schmitt, K., Holsboer-Trachsler, E., & Eckert, A. (2016). BDNF in sleep, insomnia, and sleep deprivation. *Ann Med, 48*(1-2), 42-51. doi:10.3109/07853890.2015.1131327

Schroeder, M., Krebs, M. O., Bleich, S., & Frieling, H. (2010). Epigenetics and depression: current challenges and new therapeutic options. *Current Opinion in Psychiatry, 23*(6), 588-592. doi:10.1097/YCO.0b013e32833d16c1

Sen, S., Nesse, R. M., Stoltenberg, S. F., Li, S., Gleiberman, L., Chakravarti, A., Weder, A. B., & Burmeister, M. (2003). A BDNF coding variant is associated with the NEO personality inventory domain neuroticism, a risk factor for depression. *Neuropsychopharmacology, 28*(2), 397-401. doi:10.1038/sj.npp.1300053

Shimizu, E., Hashimoto, K., Okamura, N., Koike, K., Komatsu, N., Kumakiri, C., Nakazato, M., Watanabe, H., Shinoda, N., Okada, S., & Iyo, M. (2003). Alterations of serum levels of brain-derived neurotrophic factor (BDNF) in depressed patients with or without antidepressants. *Biological psychiatry, 54*(1), 70-75.

Shirayama, Y., Chen, A. C. H., Nakagawa, S., Russell, D. S., & Duman, R. S. (2002). Brain-derived neurotrophic factor produces antidepressant effects in behavioral models of depression. *Journal of Neuroscience, 22*(8), 3251-3261.

Silberg, J., Pickles, A., Rutter, M., Hewitt, J., Simonoff, E., Maes, H., Carbonneau, R., Murrelle, L., Foley, D., & Eaves, L. (1999). The influence of genetic factors and life stress on depression among adolescent girls. *Archives of general psychiatry, 56*(3), 225-232. doi:DOI 10.1001/archpsyc.56.3.225

Simon, E. B., Oren, N., Sharon, H., Kirschner, A., Goldway, N., Okon-Singer, H., Tauman, R., Deweese, M. M., Keil, A., & Hendler, T. (2015). Losing Neutrality: The Neural Basis of Impaired Emotional Control without Sleep. *J Neurosci, 35*(38), 13194-13205. doi:10.1523/JNEUROSCI.1314-15.2015

Siuciak, J. A., Lewis, D. R., Wiegand, S. J., & Lindsay, R. M. (1997). Antidepressant-like effect of brain-derived neurotrophic factor (BDNF). *Pharmacol Biochem Behav, 56*(1), 131-137. doi:10.1016/S0091-3057(96)00169-4

Sklar, P., Gabriel, S. B., McInnis, M. G., Bennett, P., Lim, Y. M., Tsan, G., Schaffner, S., Kirov, G., Jones, I., Owen, M., Craddock, N., DePaulo, J. R., & Lander, E. S. (2002).

Family-based association study of 76 candidate genes in bipolar disorder: BDNF is a potential risk locus. *Molecular psychiatry, 7*(6), 579-593. doi:10.1038/sj.mp.4001058

Smith, A. P. (2009). Caffeine, cognitive failures and health in a non-working community sample. *Human Psychopharmacology-Clinical and Experimental, 24*(1), 29-34. doi:10.1002/hup.991

Smith, M. A., Makino, S., Kvetnansky, R., & Post, R. M. (1995). Stress and glucocorticoids affect the expression of brain-derived neurotrophic factor and neurotrophin-3 mRNAs in the hippocampus. *J Neurosci, 15*(3 Pt 1), 1768-1777.

Soldatos, C. R., Dikeos, D. G., & Paparrigopoulos, T. J. (2000). Athens Insomnia Scale: validation of an instrument based on ICD-10 criteria. *J Psychosom Res, 48*(6), 555-560.

Sousa, N., Cerqueira, J. J., & Almeida, O. F. X. (2008). Corticosteroid receptors and neuroplasticity. *Brain Research Reviews, 57*(2), 561-570. doi:10.1016/j.brainresrev.2007.06.007

Steiger, A., & Kimura, M. (2010). Wake and sleep EEG provide biomarkers in depression. *Journal of Psychiatric Research, 44*(4), 242-252. doi:10.1016/j.jpsychires.2009.08.013

Stoltenborgh, M., Bakermans-Kranenburg, M. J., & van IJzendoorn, M. H. (2013a). The neglect of child neglect: a meta-analytic review of the prevalence of neglect. *Social Psychiatry and Psychiatric Epidemiology, 48*(3), 345-355. doi:10.1007/s00127-012-0549-y

Stoltenborgh, M., Bakermans-Kranenburg, M. J., van IJzendoorn, M. H., & Alink, L. R. A. (2013b). Cultural-geographical differences in the occurrence of child physical abuse? A meta-analysis of global prevalence. *International Journal of Psychology, 48*(2), 81-94. doi:10.1080/00207594.2012.697165

Stoltenborgh, M., van Ijzendoorn, M. H., Euser, E. M., & Bakermans-Kranenburg, M. J. (2011). A Global Perspective on Child Sexual Abuse: Meta-Analysis of Prevalence Around the World. *Child Maltreatment, 16*(2), 79-101. doi:10.1177/1077559511403920

Strauss, J., Barr, C. L., George, C. J., Devlin, B., Vetro, A., Kiss, E., Baji, I., King, N., Shaikh, S., Lanktree, M., Kovacs, M., Kennedy, J. L., & Moo, I. C. C. O. (2005). Brain-derived neurotrophic factor variants are associated with childhood-onset mood disorder: confirmation in a Hungarian sample. *Molecular psychiatry, 10*(9), 861-867. doi:10.1038/sj.mp.4001685

Strecker, R. E., Morairty, S., Thakkar, M. M., Porkka-Heiskanen, T., Basheer, R., Dauphin, L. J., Rainnie, D. G., Portas, C. M., Greene, R. W., & McCarley, R. W. (2000). Adenosinergic modulation of basal forebrain and preoptic/anterior hypothalamic neuronal activity in the control of behavioral state. *Behav Brain Res, 115*(2), 183-204.

Sullivan, P. F., Neale, M. C., & Kendler, K. S. (2000). Genetic epidemiology of major depression: review and meta-analysis. *Am J Psychiatry, 157*(10), 1552-1562. doi:10.1176/appi.ajp.157.10.1552

Taylor, D. J., Lichstein, K. L., Durrence, H. H., Reidel, B. W., & Bush, A. J. (2005). Epidemiology of insomnia, depression, and anxiety. *Sleep, 28*(11), 1457-1464.

Teicher, M. H., Andersen, S. L., Polcari, A., Anderson, C. M., Navalta, C. P., & Kim, D. M. (2003). The neurobiological consequences of early stress and childhood maltreatment. *Neurosci Biobehav Rev, 27*(1-2), 33-44. doi:10.1016/S0149-7634(03)00007-1

Teicher, M. H., Dumont, N. L., Ito, Y., Vaituzis, C., Giedd, J. N., & Andersen, S. L. (2004). Childhood neglect is associated with reduced corpus callosum area. *Biological psychiatry, 56*(2), 80-85. doi:DOI 10.1016/j.biopsych.2004.03.016

Teicher, M. H., Tomoda, A., & Andersen, S. L. (2006). Neurobiological consequences of early stress and childhood maltreatment: Are results from human and animal studies comparable? *Psychobiology of Posttraumatic Stress Disorder: A Decade of Progress, 1071*, 313-323. doi:10.1196/annals.1364.024

Terracciano, A., Tanaka, T., Sutin, A. R., Deiana, B., Balaci, L., Sanna, S., Olla, N., Maschio, A., Uda, M., Ferrucci, L., Schlessinger, D., & Costa, P. T. (2010). BDNF Val66Met is Associated with Introversion and Interacts with 5-HTTLPR to Influence Neuroticism. *Neuropsychopharmacology, 35*(5), 1083-1089. doi:10.1038/npp.2009.213

Tesler, N., Gerstenberg, M., Franscini, M., Jenni, O. G., Walitza, S., & Huber, R. (2016). Increased frontal sleep slow wave activity in adolescents with major depression. *Neuroimage Clin, 10*, 250-256. doi:10.1016/j.nicl.2015.10.014

Thase, M. E. (2006). Depression and sleep: pathophysiology and treatment. *Dialogues in clinical neuroscience, 8*(2), 217-226.

Thu, K. L., Vucic, E. A., Kennett, J. Y., Heryet, C., Brown, C. J., Lam, W. L., & Wilson, I. M. (2009). Methylated DNA immunoprecipitation. *J Vis Exp*(23). doi:10.3791/935

Tiba, P. A., Palma, B. D., Tufik, S., & Suchecki, D. (2003). Effects of early handling on basal and stress-induced sleep parameters in rats. *Brain research, 975*(1-2), 158-166. doi:10.1016/S0006-8993(03)02630-1

Tiba, P. A., Tufik, S., & Suchecki, D. (2004). Effects of maternal separation on baseline sleep and cold stress-induced sleep rebound in adult Wistar rats. *Sleep, 27*(6), 1146-1153.

Tiba, P. A., Tufik, S., & Suchecki, D. (2008). Long lasting alteration in REM sleep of female rats submitted to long maternal separation. *Physiol Behav, 93*(3), 444-452. doi:10.1016/j.physbeh.2007.10.001

Ticho, S. R., & Radulovacki, M. (1991). Role of Adenosine in Sleep and Temperature Regulation in the Preoptic Area of Rats. *Pharmacology Biochemistry and Behavior, 40*(1), 33-40. doi:Doi 10.1016/0091-3057(91)90317-U

Tobler, I. I., Franken, P., Trachsel, L., & Borbely, A. A. (1992). Models of sleep regulation in mammals. *Journal of sleep research, 1*(2), 125-127.

Tochigi, M., Otowa, T., Suga, M., Rogers, M., Minato, T., Yamasue, H., Kasai, K., Kato, N., & Sasaki, T. (2006). No evidence for an association between the BDNF Val66Met polymorphism and schizophrenia or personality traits. *Schizophrenia Research, 87*(1-3), 45-47. doi:10.1016/j.schres.2006.06.029

Tononi, G., & Cirelli, C. (2003). Sleep and synaptic homeostasis: a hypothesis. *Brain Res Bull, 62*(2), 143-150.

Tononi, G., & Cirelli, C. (2006). Sleep function and synaptic homeostasis. *Sleep medicine reviews, 10*(1), 49-62. doi:10.1016/j.smrv.2005.05.002

Tononi, G., & Cirelli, C. (2014). Sleep and the price of plasticity: from synaptic and cellular homeostasis to memory consolidation and integration. *Neuron, 81*(1), 12-34. doi:10.1016/j.neuron.2013.12.025

Toth, L. A., & Bhargava, P. (2013). Animal models of sleep disorders. *Comp Med, 63*(2), 91-104.

Touma, C., Fenzl, T., Ruschel, J., Palme, R., Holsboer, F., Kimura, M., & Landgraf, R. (2009). Rhythmicity in Mice Selected for Extremes in Stress Reactivity: Behavioural, Endocrine and Sleep Changes Resembling Endophenotypes of Major Depression. *PLoS One, 4*(1). doi:ARTN e4325 10.1371/journal.pone.0004325

Tsankova, N., Renthal, W., Kumar, A., & Nestler, E. J. (2007). Epigenetic regulation in psychiatric disorders. *Nature Reviews Neuroscience, 8*(5), 355-367. doi:10.1038/nrn2132

Tsuno, N., Besset, A., & Ritchie, K. (2005). Sleep and depression. *Journal of Clinical Psychiatry, 66*(10), 1254-1269.

Turek, F. W. (2007). From circadian rhythms to clock genes in depression. *Int Clin Psychopharmacol, 22 Suppl 2*, S1-8. doi:10.1097/01.yic.0000277956.93777.6a

Turrigiano, G. (2011). Too many cooks? Intrinsic and synaptic homeostatic mechanisms in cortical circuit refinement. *Annu Rev Neurosci, 34*, 89-103. doi:10.1146/annurev-neuro-060909-153238

Ueyama, T., Kawai, Y., Nemoto, K., Sekimoto, M., Tone, S., & Senba, E. (1997). Immobilization stress reduced the expression of neurotrophins and their receptors in the rat brain. *Neuroscience Research, 28*(2), 103-110. doi:Doi 10.1016/S0168-0102(97)00030-8

Urrila, A. S., Karlsson, L., Kiviruusu, O., Pelkonen, M., Strandholm, T., & Marttunen, M. (2012). Sleep complaints among adolescent outpatients with major depressive disorder. *Sleep Medicine, 13*(7), 816-823. doi:10.1016/j.sleep.2012.04.012

Van de Velde, S., Bracke, P., & Levecque, K. (2010). Gender differences in depression in 23 European countries. Cross-national variation in the gender gap in depression. *Soc Sci Med, 71*(2), 305-313. doi:10.1016/j.socscimed.2010.03.035

van Oostrom, I., Franke, B., Rijpkema, M., Gerritsen, L., Arias-Vasquez, A., Fernandez, G., & Tendolkar, I. (2012). Interaction between BDNF Val66Met and childhood stressful life events is associated to affective memory bias in men but not women. *Biological Psychology, 89*(1), 214-219. doi:10.1016/j.biopsycho.2011.10.012

Ventskovska, O., Porkka-Heiskanen, T., & Karpova, N. N. (2015). Spontaneous sleep-wake cycle and sleep deprivation differently induce Bdnf1, Bdnf4 and Bdnf9a DNA methylation and transcripts levels in the basal forebrain and frontal cortex in rats. *Journal of sleep research, 24*(2), 124-130. doi:10.1111/jsr.12242

Verhagen, M., van der Meij, A., van Deurzen, P. A. M., Janzing, J. G. E., Arias-Vasquez, A., Buitelaar, J. K., & Franke, B. (2010). Meta-analysis of the BDNF Val66Met polymorphism in major depressive disorder: effects of gender and ethnicity. *Molecular psychiatry, 15*(3), 260-271. doi:10.1038/mp.2008.109

Vetulani, J. (2013). Early maternal separation: a rodent model of depression and a prevailing human condition. *Pharmacological Reports, 65*(6), 1451-1461.

Vyazovskiy, V., Borbely, A. A., & Tobler, I. (2000). Unilateral vibrissae stimulation during waking induces interhemispheric EEG asymmetry during subsequent sleep in the rat. *Journal of sleep research, 9*(4), 367-371. doi:DOI 10.1046/j.1365-2869.2000.00230.x

Vyazovskiy, V. V., Cirelli, C., Pfister-Genskow, M., Faraguna, U., & Tononi, G. (2008). Molecular and electrophysiological evidence for net synaptic potentiation in wake and depression in sleep. *Nat Neurosci, 11*(2), 200-208. doi:10.1038/nn2035

Vyazovskiy, V. V., Ruijgrok, G., Deboer, T., & Tobler, I. (2006). Running wheel accessibility affects the regional electroencephalogram during sleep in mice. *Cerebral cortex, 16*(3), 328-336. doi:10.1093/cercorbhi110

Vyazovskiy, V. V., & Tobler, I. (2005). Theta activity in the waking EEG is a marker of sleep propensity in the rat. *Brain research, 1050*(1-2), 64-71. doi:10.1016/j.brainres.2005.05.022

Wang, Q., Shao, F., & Wang, W. (2015). Maternal separation produces alterations of forebrain brain-derived neurotrophic factor expression in differently aged rats. *Front Mol Neurosci, 8*, 49. doi:10.3389/fnmol.2015.00049

Warner-Schmidt, J. L., & Duman, R. S. (2006). Hippocampal neurogenesis: opposing effects of stress and antidepressant treatment. *Hippocampus, 16*(3), 239-249. doi:10.1002/hipo.20156

Watson, B. O., Levenstein, D., Greene, J. P., Gelinas, J. N., & Buzsaki, G. (2016). Network Homeostasis and State Dynamics of Neocortical Sleep. *Neuron, 90*(4), 839-852. doi:10.1016/j.neuron.2016.03.036

Weaver, I. C., Cervoni, N., Champagne, F. A., D'Alessio, A. C., Sharma, S., Seckl, J. R., Dymov, S., Szyf, M., & Meaney, M. J. (2004). Epigenetic programming by maternal behavior. *Nat Neurosci, 7*(8), 847-854. doi:10.1038/nn1276

Weinhold, B. (2006). Epigenetics - The science of change. *Environmental Health Perspectives, 114*(3), A160-A167.

Weiss, B., & Garber, J. (2003). Developmental differences in the phenomenology of depression. *Development and Psychopathology, 15*(2), 403-430.

Wilber, A. A., & Wellman, C. L. (2009). Neonatal maternal separation alters the development of glucocorticoid receptor expression in the interpositus nucleus of the cerebellum. *Int J Dev Neurosci, 27*(7), 649-654. doi:10.1016/j.ijdevneu.2009.08.001

Willner, P. (2017). The chronic mild stress (CMS) model of depression: History, evaluation and usage. *Neurobiol Stress, 6*, 78-93. doi:10.1016/j.ynstr.2016.08.002

Willner, P., Towell, A., Sampson, D., Sophokleous, S., & Muscat, R. (1987). Reduction of Sucrose Preference by Chronic Unpredictable Mild Stress, and Its Restoration by a Tricyclic Antidepressant. *Psychopharmacology, 93*(3), 358-364.

Wirz-Justice, A., & Van den Hoofdakker, R. H. (1999). Sleep deprivation in depression: what do we know, where do we go? *Biological psychiatry, 46*(4), 445-453.

Wray, N. R., James, M. R., Handoko, H. Y., Dumenil, T., Lind, P. A., Montgomery, G. W., & Martin, N. G. (2008). Association study of candidate variants from brain-derived neurotrophic factor and dystrobrevin-binding protein 1 with neuroticism, anxiety, and depression. *Psychiatric Genetics, 18*(5), 219-225. doi:10.1097/YPG.0b013e3283050aee

Yamada, K., Kobayashi, M., Shiozaki, S., Ohta, T., Mori, A., Jenner, P., & Kanda, T. (2014). Antidepressant activity of the adenosine A2A receptor antagonist, istradefylline (KW-6002) on learned helplessness in rats. *Psychopharmacology (Berl), 231*(14), 2839-2849. doi:10.1007/s00213-014-3454-0

Yang, G., & Gan, W. B. (2012). Sleep contributes to dendritic spine formation and elimination in the developing mouse somatosensory cortex. *Dev Neurobiol, 72*(11), 1391-1398. doi:10.1002/dneu.20996

Zant, J. C., Rozov, S., Wigren, H. K., Panula, P., & Porkka-Heiskanen, T. (2012). Histamine release in the basal forebrain mediates cortical activation through cholinergic neurons. *J Neurosci, 32*(38), 13244-13254. doi:10.1523/JNEUROSCI.5933-11.2012

Zhu, J. K. (2009). Active DNA demethylation mediated by DNA glycosylases. *Annu Rev Genet, 43*, 143-166. doi:10.1146/annurev-genet-102108-134205

Zorrilla, E. P., Luborsky, L., McKay, J. R., Rosenthal, R., Houldin, A., Tax, A., McCorkle, R., Seligman, D. A., & Schmidt, K. (2001). The relationship of depression and stressors to immunological assays: A meta-analytic review. *Brain Behavior and Immunity, 15*(3), 199-226. doi:DOI 10.1006/brbi.2000.0597